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ATBT
الجمعيّة التونسيّة للتقانات الحيويّة

L'ASSOCIATION TUNISIENNE DE BIOTECHNOLOGIE
THE TUNISIAN ASSOCIATION OF BIOTECHNOLOGY

IDB2018

THE 17TH INTERNATIONAL DAYS OF BIOTECHNOLOGY

December 20th - 23th, 2018
Hotel Green Park Concorde *****
SOUSSE, TUNISIA

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TOPICS

INDUSTRIAL BIOTECHNOLOGY | ENVIRONMENTAL BIOTECHNOLOGY. | PLANT BIOTECHNOLOGY | HEALTH BIOTECHNOLOGY | BIOACTIVE MOLECULES AND APPLICATIONS

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Foreword

Dear Colleagues,

The field of biotechnology is growing and playing a leading role in the global economy. Indeed, bioproducts, renewable chemicals and energy, biomass are interesting alternatives to the current problems caused by high prices, climate change and depletion of oil deposits.

The future of biotechnology in developing countries will depend on policy makers, a clear strategic vision and the researchers willingness.

The 17th International Days of Biotechnology (IDB/JIB 2018) represent an opportunity to work in this direction, to sensitize the political leaders of the region and to make biotechnology a national and Mediterranean priority. These 17th International Days of Biotechnology (IDB/JIB 2018), organized by the Tunisian Association of Biotechnology, represent also an opportunity for students, young researchers and professors to present and discuss their research.

The 5 topics to be covered during IDB/JIB 2018 are: 1: Health Biotechnology (vaccine and diagnostics); 2: Industrial biotechnology (enzyme, microorganisms, metagenomics and bioprocesses); 3: Environmental Biotechnology (treatment and recovery of waste, bioremediation, bioenergy and phytoremediation); 4: Bioactive molecules and applications and 5: Plant biotechnology (genetic diversity, environmental constraints and plant breeding, plant-microorganism interaction).

After a selection based on scientific content and relationship with the 5 themes that will be developed during the IDB/JIB 2018, the retained participants that confirmed their participation are 265, in addition to about 46 participants without contribution. The program includes 9 plenary lectures, presented by eminent foreign and Tunisian professors and researchers, who will present the latest novelties in relation with the five retained themes. In addition to these conferences, we have scheduled 11 Thematic Conferences, 62 oral communications and 192 poster communications.

Dear Colleagues,

In order to increase the number of oral communications and to free up more time for poster discussion, we decided to do parallel sessions while keeping the lectures in plenary form. The choice of oral communications was made by the Scientific Committee considering the specificity of the A. T. Biotech and the applied aspects of the proposed works. This choice was very difficult, given the wish of several participants to communicate their work orally. At the same time, we devoted 5 poster sessions to give more importance to poster presentations and we will also award the best posters for each theme.

Finally, the organizing committee and of A.T.Biotech office would like to thank all the speakers and the participants and all those who participated in the organization and the success of these 17th International days of Biotechnology.

The organisation commitee

SUMMARY

AKNOLDGEMENTS

The organizers are very grateful for their moral and / or financial support:

Mr. Minister of Higher Education and Scientific Research
Mr. Secretary of State for Scientific Research
Mr. Director General of the Biotechnology Center of Sfax (CBS)
Mr. Director General of the Biotechnology Center of Borj-Cédria (CBBC)
Mr. Director General of the Pasteur Institute of Tunis (IPT)
Mr. Director General of the National Center for Nuclear Science and Technology (CNSTM)
The Manager and all the staff of the Hotel Concorde Marco Polo Hammamet
The manager of the company F-LAMBDA

The organizers also thank all those who participated in the organization and success of the JIB 2018, in particular:
Mr. Ramzi Rekik
Mrs. Fatma Elgharbi
Mrs. Saousan Kchaou Ben Jemaa
Mrs. Monia Mezghani Abid

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CONFERENCES
(Cf)

Cf1: National Agency for the Promotion of Scientific Research: Interfacing structure for the reinforcement of the interactions between research and the socio-economic environment

Prof. Chedly Abdelly

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Abstract

SWOT analysis for National System of Research and Innovation showed several positive features: main institutional actors are present, human resources (researchers, ...) are important and qualified, funding is based in large part on performance, scientific production is abundant and with good quality, some innovative companies are available as well as excellence structures with international position. The negative aspects concern mainly the low patent production with almost no exploitation, weak private sector contribution to research and innovation, as well as modest interactions between research structures and the socio-economic environment. In this context, a national strategy has been implemented by the Ministry of Higher Education and Scientific Research to bring research closer from its potential users through several financing mechanisms covering different levels of technological ripeness.

The purpose of this conference is to present the new mechanisms dedicated to the valorization of the research and the missions of the National Agency for the Promotion of Scientific Research (ANPR) as an interfacing structure for the reinforcement of the interactions between research and the socio-economic environment. Mechanisms relating to the reinforcement of researcher expertise in terms of international project setting up, the mobility of young researchers to companies, the creation of Spin-offs, the promotion of the process of technology transfer and the establishment of consortia will be presented. Adoption of new relevant approaches for regional development such as smart specialization will be discussed

Cf2: How can technology transfer from lab to market push companies for open innovation?

Prof. Souad Rouis
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Abstract

Economic crisis is mainly manifested by the difficulties for the creation and survival of enterprises. Moreover, proposed strategies to promote economic and social development in our region should take into account the crucial role of these SMEs especially for small countries and transition economic situation like Tunisia and Greece. One of the most commonly adopted solutions is to promote private employment through improvement of the SMEs competitiveness and innovative startups creation. To be competitive these SMEs must absorb all the high quality university/research Know-How so as to increase innovation in their value chains towards higher levels of value creation.

Sharing of knowledge and know-how could be driven by joint collaboration (through collaborative projects) between academic laboratories, techno-parks, private incubators and SMEsbased on a pre-established value chain.Academic and industrial exchange will allow to researchers to acquire the required skills to deal with economic issues through a deep networking with SMEs and support structures for technology transfer.

Cluster is also proposed as the best strategy to allow these SMEs to overcome their financial weakness and cover the cost of technology transfer. Such Clusters will be based on innovative cooperation among Quadruple Helix stakeholders to implement a common vision and strategy. Target groups and beneficiaries are identified as: Enterprises (SMEs, micro, large, industry & services, innovators) that need Money (grants, guarantees, venture capital, etc..), Clients, markets (domestic, global), Partners (development, value chains ...), Support for new forms of innovation and more tailor made measures, National authorities and /or regional (from different departments including policy making, regional development agencies, guidance and financing). Social networks and the civil sector will also play a crucial role as intermediaries for connecting with partners and target groups, such as citizens, consumers and communities – and more institutionalized actors from the public, private and academic sectors.

Cf3: Precision Medicine in the Era of "Big Data" Genetics

Dr. Peter J. Tonellato

Director of the Center for Biomedical Informatics, Professor of Bioinformatics, School of Medicine, University of Missouri, Columbia MO.

Abstract

Essential to a future of preventive and predictive medicine is the use and application of modern technologies and computational approaches in the implementation of precision medicine. A sweeping array of bioinformatics tools, approaches and analysis will impact everything from the integration of whole genome technologies into clinical and health practice to the identification and refinement of pharmacogenetic algorithms designed to vastly improve clinical outcomes. We present examples of the development of precision medicine, improved outcomes including the use of whole genome sequencing and analysis in rare disease, pharmacogenetics and cancer care to create a post-genome paradigm shift in health, disease prevention, and clinical care. We also demonstrate the use of bioinformatics in the accurate dosing of drugs whose metabolism is impacted by the genetics of the individual patient and a new companion Comprehensive Cancer Genetic Test recently approved by the FDA and opportunities to develop validation studies demonstrating the impact of such a test in best practice cancer treatment. These and parallel efforts though difficult, will catalyze the adoption and widespread implementation of precision medicine resulting in dramatically improved patient outcomes.

Cf4: Redefining the specificity of a human protein: the plasma retinol-binding protein case

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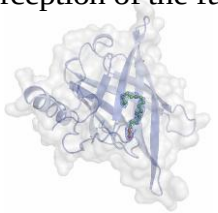
Abstract

RBP4 (plasma retinol-binding protein) is the 21 kDa transporter of all-trans retinol that circulates in serum as a moderately tight 1:1 molar complex of the vitamin with the protein [1]. RBP4 is primarily synthesised in the liver but is also produced by adipose tissue (about 20-40% of the amounts released by the liver) and circulates bound to a larger protein, transthyretin, TTR, that serves to increase its molecular mass to about 80,000 and thus avoid its elimination by glomerular filtration. The RBP-TTR complex dissociates readily upon interaction with the RBP receptor, STRA6, that removes the vitamin from the transporter and facilitates its entrance into the cell. When retinol is not present in the complex, RBP dissociates from TTR and is eliminated in urine. In 1993 the X-ray structure of human holo RBP4 and of what was expected to be the apo form, i.e. after the loss of retinol, to 2.5 Å resolution has been reported [2]. A couple of years ago we tested crystals from the same batch at the ESRF and we collected a full data set of both the liganded and unliganded crystal forms at a resolution of 1.5 Å and 2.0 Å respectively. The result was that we identified a fatty acid in the ligand-binding site of the protein believed to be apo. We also prepared crystals of BP4 purified from human urine and amniotic fluid, two sources that contain non fluorescent RBP4, i.e. not bound to retinol, and for that reason believed to be the apo form of the protein. In every case we found a fatty acid in the central cavity of the RBP4 molecule, a result that we confirmed by GC-MS analysis of the samples used in the crystallization experiments. This result changes substantially our perception of this protein that has so far been considered to be specific for retinol and is a good example of how simply increasing the quality of the diffraction data can change the perception of the function of a protein [3].



Surface and ligand representation of Human RBP4 with palmitic acid bound

Keywords: Human plasma retinol-binding protein, Fatty acid, Palmitic acid RBP4, X-ray structure.

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Cf5: Bacterial Spore Formers: Biology and Biotechnology

Dr. Ezio Ricca

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Abstract

Bacterial spores are peculiar cells, differentiated by members of the *Clostridium* and *Bacillus* genera, in response to nutrient starvation and to other unfavourable conditions. Spores are metabolically quiescent, highly resistant and can persist indefinitely in conditions that would be lethal for normal cells. However, when spores meet environmental conditions favourable for cell grow, they germinate, originating cells able to grow and, eventually, to sporulate again. Spores are ubiquitous in nature and are commonly isolated from soils and aquatic samples, including samples from extreme environments such as deserts, hydrothermal sites and artic ices. Spores are also found associated in large numbers to other organisms, insects, plants and animals, including humans, and have been shown able to perform their entire life cycle inside the animal gastro-intestinal tract (1). Most spore formers are not pathogenic to human and animals and spores of some species are commonly used as health promoters in commercial products for human and animal use (1).

Spores of the non-pathogenic species *Bacillus subtilis* have been exploited as possible vectors for the delivery of drugs, antigens and enzymes (2). *B. subtilis* proteins localized on the spore surface have been used as carriers to fuse and anchor heterologous proteins. This recombinant approach has been used over the years to display several different antigens with various spore surface proteins as carriers (1, 2). In some cases the recombinant spores have been assessed as mucosal vaccines in animal models and proved able to induce specific and protective immune responses (1, 2). More recently, a non-recombinant approach, based on the spontaneous adsorption of molecules on the negatively charged spore, has been also developed and used to display enzymes and antigens (3).

An overview of recent basic reserch and biotechnological applications of *Bacillus* cell and spores will be presented.

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Cf6: Nitric oxide (NO) regulates symbiotic nitrogen fixation in legumes

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Abstract

Nitric oxide (NO) is a gaseous signaling molecule with a broad spectrum of regulatory functions in plant and animal growth and development. It is involved in plant response to abiotic stress, including hypoxia, and biotic interactions such as nitrogen-fixing symbiosis (NFS) between legumes and soil gram-negative bacteria of rhizobium type. The interaction between legumes and rhizobia leads to the formation of new differentiated organs named nodules, which provide a niche for bacterial nitrogen fixation. In the nodules, bacteria differentiate into bacteroids with the ability to fix atmospheric nitrogen via nitrogenase activity. As nitrogenase is strongly inhibited by oxygen, nitrogen fixation is made possible by the microaerophilic conditions prevailing in the nodules. Thus, nodule development occurs in changing oxygen conditions, shifting from a normoxic environment during symbiosis establishment to a microoxic one in functioning nodules. Increasing evidence has been reporting the presence of NO during symbiosis, from early interaction steps between the plant and the bacterial partners, to nitrogen-fixing and senescence steps in mature nodules. The chemical nature, concentration and location of NO determine its either signalling, toxic or metabolic functions.

Using the *Medicagotruncatula – Sinorhizobiummeliloti* symbiotic model, we found that NO is required for an optimal establishment of the interaction, nodule development, nitrogen-fixing metabolism and nodule senescence. This presentation will investigate NO source and degradation pathway, and summarize the different roles of NO throughout the nitrogen-fixing symbiotic process.

Cf7: Proteomics as promosing tool to progress in the elucidation of salt tolerance mechanisms in a model of Extremophile plants: *Cakile maritima*

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Abstract

Halophytes are plants naturally thriving in saline environments where salt concentration exceeds 200 mM NaCl. These species (both dicots and monocots) account for ~1% of the total flora of the world and are distributed mainly in arid, semi-arid inlands but also saline wet lands along the tropical and sub-tropical coasts. Salinity tolerance in halophytes depends on a set of complex responses taking place at different (from the cellular to the whole plant) levels, which allow these extremophile plants to grow and flourish in high saline conditions. Many physiological responses are detectable in terms of protein abundance changes even before physiological responses take place. In addition, many cellular processes in response to salt stress rather take place on the level of protein-protein interactions (PPI), post-translational protein modifications, and enzymatic activities, which can be hardly identified by gene expression studies. This makes protein analysis a pertinent tool to assess functional linking between protein and genomic repertoires in plants challenged with salinity. Using the proteomic tool, many proteins related to salt tolerance have been identified and the related genes cloned and transferred to glycophytes to improve salt tolerance.

Cakile maritima (Brassicaceae) is a seashore-native halophyte with high dispersal aptitude due to long-term seed floating ability. In Tunisia, the native ecosystems of this species are exclusively littoral sandy dunes, extending from humid to semi-arid and even arid areas (personal observations). *C. maritima* shows considerable genetic diversity reflecting to ecological adaptation to local environmental conditions. Here, we try to highlight how the proteomics approach could be used in order to more accurately infer how this promising model of salt-tolerance adapts to salt stress. The contribution of this approach is mainly discussed with respect to (i) the shift in the plant behavior during its ontogeny (at the germination and vegetative stages), (ii) highlighting the anticipatory response to salinity adopted by *C. maritima*, and (iii) understanding the intra-specific variability characterizing local accessions of this halophyte.

Cf8: Carbon monoxide and the Food Packaging in the 21st Century: the Paradox

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Abstract

Due to increased demands for greater expectation in relation to quality, convenience, safety and extended shelf-life, combined with growing demand from retailers for cost-effective extensions of fresh muscle foods' shelf-life, the food packaging industry quickly developed to meet these expectations. During the last few decades, modified atmosphere packaging (MAP) of foods has been a promising area of research, but much remains to be known regarding the use of unconventional gases such carbon monoxide (CO). The use of CO for meat and seafood packaging is not allowed in most countries due to the potential toxic effect, and its use is controversial in some countries. The commercial application of CO in food packaging was not then considered feasible because of possible environmental hazards for workers. CO has previously been reported to mask muscle foods' spoilage, and this was the primary concern raised for the prohibition, as this may mislead consumers. This review was undertaken to present the most comprehensive and current overview of the widely-available, scattered information about the use of CO in the preservation of muscle foods. The advantages of CO and its industrial limits are presented and discussed. The most recent literature on the consumer safety issues related to the use of CO and consumer acceptance of CO especially in meat packaging systems were also discussed. Recommendations and future prospects were addressed for food industries, consumers and regulators on what would be a "best practice" in the use of CO in food packaging. All this promotes high ethical standards in commercial communications by means of effective regulation, for the benefit of consumers and businesses in the world, and this implies that industrialized countries and members of their regulatory agencies must develop coherent and robust systems of regulation and control that can respond effectively to new challenges.

Keywords: muscle foods; modified atmosphere packaging; CO; shelf-life; best practice; regulation.

Cf9: Complementary solutions for dry waste (biomass) and waste water treatment and renewable energy recovery

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Abstract

The rapid increase in population and industrializationcaused a continued production of waste and waste water with high organic content, which creating management problems. The use of traditional methods, for waste treatment, releases gases that may cause global warming. In addition, energy demand is also incresing rapidly in the world. Therefore, owing to access to clean and green energywhichis essential for the sustainabledevelopment of human society. These two challenges, if managed scientifically using biowaste to energy by biological methods combined with solar energy canprovide promisingsolutions.

In this conference, we reviewed the strategies for and status of waste to energy technology used to convert various charged biowastes into bioenergy. Severalstudies have been carried out to characterize organic waste, evaluate their energy potential and to choose the most appropriate process design for converting each of these resources into energy. Also, it is important to study the value chain (collection, sorting, transport to the point of energy production) and to evaluate the cost of electric and thermic kWh produced. All the studies carried out were validated in a multi-scale experimental pilot allowing to produce energy at large scale. For example, two stage anaerobicco-digestion of the most abundant organic wastes with couples CH4 and H2 recuperation may be an important contribution for high-rate Bioenergy recovery. Furthermore, a hybrid energy system, involving organic Rankine cycle (ORC) and a biogas boiler can be investigated.The biogas boiler with a module of Parabolic-Solar Collectors (PSC) can be employed to providea heat source to the ORC via two distinct intermediate circuits in summer and in winter conditions, respectively.

We concluded that various complementary biomass to Bioenergy technologies have been developed efficiently. They open up the possibility of waste treatment with reduced costs and environmental impacts, which could be one of the best strategies for the future of the circular bio - economy. The participation of researchers, scientists, and government agencies, is needed to increase the feasibility of these technologies.

Keywords: Biomass, bioenergy, solar power, circular economy,waste management.

BH Session
Health Biotechnology

ORAL COMMUNICATIONS
(HBO)

TC1-HB/HBO1: A population-based study of copy number variation in healthy Tunisian individuals

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Background and aim: Copy Number Variation (CNV) is a type of genetic variation defined as DNA segments of more than 1kb presenting copy-number differences when comparing two or more genomes. CNVs are generated by one-single or a combination of multiple genomic rearrangements such as deletion, insertion and duplication. They can impact on gene expression and phenotypes. Therefore, CNVs have been reported as associated with several complex traits including neurological diseases such as Autism and Schizophrenia. Recent studies focusing on CNVs in some ethnic groups such as Koreans, Chinese and Europeans had highlighted population-specific enrichment of CNVs. Such data are not available for populations from North Africa as Tunisia. In this work, we conducted a population-based study to detect and characterize CNV in the Tunisian population.
Methods: One hundred and two healthy individuals were genotyped with the Affymetrix 6.0 array.PennCNV software was used to call CNV segments.
Results: We called 3964 CNVs with an average of 39 segments per individual. The CNV mean length was 96.7 kb. Deletions represented 66.5 % of the called CNVs with an average length of 65.98 kb which was less than mean length duplication (173 kb). These CNVs have been clustered in 751 CNVRs (CNV regions) with a mean length of 104 kb and covering 78 Mb of the genome. Sixty percent of these CNVRs have a frequency of less than 1%. Three percent of these CNVRs are not found in the DGV database. About 34% of CNVRs overlapped with 234 RefSeq genes. Gene Ontology annotation revealed that genes overlapping CNVRs are significantly enriched in «calcium ion binding » molecular function (0.4%; p-value=7 10^{-4}), involved in biological processes as «cellular adhesion» (7 %; pvalue=1.45 10^{-4}) and «nervous system developement» (19%; p-value=1.45 10^{-5}). Overrepresentation of cellular components are «synapse» (9.5 %; p-value=6.47 10^{-3}) and «integral component of plasma membrane » (13.2%; p-value=3.6 10^{-2}).
Conclusions: Our preliminary results suggest the need of a complete characterization of the CNVs in the Tunisian population and the map that we build will serve as a useful resource for further medical studies.
Keywords: Copy number variation, Affymetrix 6.0, Tunisian population

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HBO2: High Throughput Analysis for Processed Meat Species Authentication

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Background and aim: Food adulteration has moved from being fraudulence to a highly sophisticated business. As a matter of fact, the identification of meat species is requisite for religious, safety and conservation regulations reasons, among others.
Methods: Mitochondrial DNA (mt-DNA), present with multi-copy by cell, is likely to be the best target and is used in several commercialized kits and normalized protocols. However, the low throughput of the PCR-based methods, the high level of polymorphisms/heteroplasmy, as well as the need for a complete analysis of the cooked ground-meat motivated us to develop new specific protocols with a high analytical throughput and a better discrimination power.
The next generation sequencing of DNA (NGS) could meet these needs by providing parallel sequencing and both qualitative and quantitative data.
Results: In the current study, mt-DNA sequences from different species (i.e., chicken, turkey, ostrich, cattle, sheep, goat, donkey, horse, dog, cat, rat, mouse and Pig) were analyzedby multiple sequence alignment. Conserved sequences flanking short high variable regions of mtDNA were identified and used for primer designs. The three most variable regions were selected (size less than 300 bp) for further investigation using NGSamong Tunisian livestock (including 7 different species)with the purpose to identify local specificity and improve single test sensitivity. Amplicons were sequenced using a Miseq sequencer (Illumina Technology) and a VCF file was built for each included species.
The characterization of Tunisian livestock was performed using a specific program written under the R language. A specific algorithm was implemented with the aim to attribute a relative mutability for each position of the included variable region considering the reference sequences. The algorithm first cleared the duplicate mitochondrial sequences and then calculated the different nucleotide frequencies for each position and at the end the discriminatory positions for each species. Therefore, a specific barcode was generated for each species.
Conclusion: Our work is apreliminary step towards developing accurate single, medium or high-throughput tools for processed and cooked meatauthentification.
Keywords: mtDNA, animal species, adulteration, NGS, algorithm, multiple alignments.

HBO3: Prevalence and characterization of carbapenemase producing Enterobacteriaceae isolated from poultry meat

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Background and aim: Carbapenems are the molecules of choice for the treatment of nosocomial infections. However, in recent years, there is a spread of enterobacteriaceae strains resistant to these molecules. Several mechanisms may have been at the origin of carbapenem resistance, including the production of carbapenemases. The aim of this study is to search the production of carbapenemases by phenotypic methods in bacterial strains isolated from poultry meat.
Methods: The samples were taken from the different butcheries of the city of Tébessa. After isolation, the identification was carried out by the Api 20E strips and the carbapenem susceptibility test was performed according to the diffusion method in agar medium. The production of carbapenemases was investigated by the Carba NP test, a phenotypic test based on the demonstration of the medium acidification, by a colored indicator, during the hydrolysis of imipenem by a carbapenemase.
Results: From 9 samples, we have isolates and identified 75 strains of enterobacteriaceae that belong to different genera: *Serratia* (50. 67%), *Escherichia* (14.67%), *Kluyvera* (13.33%), *Salmonella* (8%), *Klebsiella* (6.67%), *Citrobacter* (4%) and *Enterobacter* (2.67%). The susceptibility test to carbapenems showed a high resistance to ertapenem (58.06%) and a lower rate to imipenem (26.76%). The test of Carba NP was positive in 6/75 of the strains, ie 8% of the enterobacteriacea tested.
Conclusion: Our study shows the presence of enterobacteriaceae producing carbapenemases in poultry meat. Therefore, these resistant bacteria can potentially be transferred to humans across the food chain. This situation raises serious concerns about public health and food safety.
Keywords: enterobacteriaceae, carbapenem resistance, carbapenemases, poultry meat, Carba NP test.

HBO4: Beyond the standard, a Y-chromosome markers rearrangement increases their forensic relevance using Shannon Entropy Approach

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Background: Y-chromosome STRs genotyping becomes the reference tool targeting the male component in forensic investigation and kinship. To increase his discriminatory power (DP), forensic scientists upgraded the Y- STR panel up to the latest megaplexes of 23 and 27 YSTRs. Megaplexes kept the core panel disregarding the impact related to each STR in a given population leading laboratories to acquire the latest chemistry.
Materiel and Methods: In our investigation, we initially explored caseworks using the core YSTRs. Then, we selected nine markers previously described as genetically diverse in different populations including two STRs published in megaplexes kits, two rapidly mutating RM (DYS710, DYS449) and one multicopy Y-STR (DTS464) to increase the DP. To go over technical limits, a rearrangement of Y-STRs was suggested ignoring the core Y-STRs presence. To confirm the strategy, two calculation modalities were done based on linkage disequilibrium (LD) and Shannon entropy (SE).
Results: Technically, the limited DP of the core panel was low (80%) and highlighted an inability to solve complexed crimes. In 200 South Tunisian males, RM STRs were highly diverse and able to show the hidden DP between close individuals. Their addition to the standard panel increased the DP up to 96%. The rearranged multiplexing optimizations confirmed SE and achieved a combination including remarkably limited number of markers increasing DP (96%) in South Tunisians. Statistically, as the LD kept the whole standard panel and revealed the need of the slowly mutating Y-STRs, the SE discarded them and decreased the marker information redundancy related to the single STR characterization which ignored the markers inter-relation yielding a rearranged small set.
Conclusion: Although published kits provided limited information disregarding our population specificities, the standard panel is internationally recommended and mandatory in our court rooms. Population specificities were highlighted with the newly selected sets. An ongoing collaborative work on alternative Y RM markers on Tunisian male buccal swabs held in the Forensic DNA Laboratory of the University of West Cape Town highlighted the need to shift to a new progress considering African populations specificities. Both panels are ongoingly involved in complexed caseworks investigation for a final appraisal.
Key words: Rapidly-mutating Y-STRs, Discriminatory Power, Conditional Entropy, Forensic evidence

HBO5: Synthesis, characterization and biological evaluation of new PEGylated triarylmethanes

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Background and aim: The emergence and dis semination of multidrug resistant (MDR) Gram-negative bacterial pathogens observed in recent years is a major challenge for antimicrobial chemotherapy and is today considered as a major public health issue. This "antibiotic resistance crisis" has been intensified by the gap between the burden of infections due to MDR bacteria and the development of new antibiotics to tackle the problem. Moreover, despite the discovery over the last twenty years of compounds with an interesting antibiotic activity, few of them belong to new chemical classes or have the required properties to become drugs or to avoid resistance problems. That's why the need for new antibiotics is urgent.

Small molecular drugs often suffer some problems, such as low solubility, high toxicity, rapid excretion or untargeted biodistribution. To overcome these obstacles, one promising approach is to use a PEGylation strategy. Thanks to their favorable properties (nontoxic, nonimmunogenic, non-antigenic and amphiphilic), PEG as modifying polymer plays an important role in drug discovery. PEG-drug conjugates have several advantages: prolonged residence in body, decreased degradation by metabolic enzymes, reduction or elimination of protein immunogenicity, and so on.

Methods: The analogues ofbisacodyl were synthesized from commercially available reagents *via* a Friedel-Crafts reaction followed by a functionalization with PEG fragments to increase solubility. The bioassay was performed using polystyrene micro-assay plates (96 well) using levofloxacine, a broad-spectrum antibiotic, as a control.

Results: The antibacterial activity against Gram-positive and Gram-negative pathogens of the compound 4, 4' -(pyridin-2-ylmethylene) diphenol was evaluated. This compound exhibited antilisterial activity, with minimum inhibitory concentration (MIC) ranging from 6.25 to 12.5 µg/mL against *Salmonella typhimurium.* Additionally, it was active against *Escherichia coli*, *Listeria monocytogene* and *Staphylococcus aureus* with MIC values between25-50 µg/mL and 12.5-25 µg/mL. The derivatives bearing PEG groups have also been tested revealing a promising antibacterial activity. In addition, the anticancer activity will be tested towards HeLa cells (cervical epithelial cancer cell line).

Conclusion: The PEGylated compounds displayed a higher bacteriostatic activity compared to bisacodyl.

Keywords: bisacodyl, antibacterial activity, PEGylation, triarymethane.

TC2-HB/HBO6: Effect of polymorphisms in pro-inflammatory cytokine genes in carcinogenesis of gastric mucosa infected with *Helicobacter pylori*

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Background: The Gastric cancer causes a significant amount ofmorbidity and mortality worldwide. Infection with *Helicobacter pylori* is considered a potential risk of developing gastric cancer in association with contributing host genetic factor. IL-1f3, TNF-a and IL-1RN polymorphisms appear to maintain and promote *Helicobacter pylori* infection and to stimulate neoplastic growth ofthe gastric mucosa.

Objective and methods: To elucidate the effect of these polymorphisms in combination with gastric cancer by focusing on the role of *Helicobacterpylori* virulence factors in a population from northwestern Algeria, a case-control study was carried out on 79 patients infected with H. pylori with chronic atrophic gastritis and/or gastric carcinoma, and 32 subjects were recruited as case-control. The bi-allelic polymorphisms of IL-1f3-31, IL-1f3-51 1, TNF-a-308, TNF-a-1031 and and IL-1RN penta-allelic were genotyped. As well as a prospection of the *Helicobacter pylori*: *vac A*, *cag-PAI* and *fla A* genes were carried out.

Results: IL-1f3-31*C homozygous genotype (OR: 4.6) and IL-1RN*2 heterozygous (OR=4.2) were associated with an increased risk of developing gastric carcinoma. However, IL-1 f3-511 *T was associated with an increased risk of development of chronic atrophic gastritis (OR: 4.3). TNF-a-308 type A/A homozygous genotype (OR: 1.567) and heterozygous G/A (OR: 1.272); TNF-a-1031 homozygous genotype C/C (OR: 3,861) and heterozygous T/C (OR: 1,261) were associated with an increased risk of developing gastric carcinoma. *Vac A s1/m1* and the *cag-PAI* integrity of *Helicobacter pylori* were associated with the development of gastric cancer at 93.7%.

Conclusion: IL-1 β, IL-1 RN and TNF-a polymorphisms (The alleles IL-1 β-31 *C, TNF-a103 1*C, TNF-a-308*A and IL-1RN*2) associated with *Helicobacter pylori* infection contribute to the development of chronic atrophic gastritis and gastric carcinomas in an Algerian population. However, IL-1β-51 1*T was associated with an increased risk of developing chronic atrophic gastritis with no significant association of developing gastric carcinoma. The study of virulence factors revealed that genes and specific regions such as *cagA*, *vac A s1/m1*, and the integrity of the pathogenicity island cag-PAI have been linked to the evolution of gastric cancer. As well as the left end of the islet (*vir B1 1*) has been linked to the development of chronic atrophic gastritis.

Keywords: Gastric mucosa, Polymorphism, Proinflammatory cytokines, Helicobacter pylori.

HBO7: HLA-Class II gene polymorphism in Colorectal Cancer

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Background and aim: Colorectal cancer (CRC) is a major cause of morbidity and mortality worldwide. It represents a serious health problem in Tunisia. The research of biomarkers associated to a higher risk to develop CRC is an important area of recent research. The aim of our study was to evaluate the polymorphism of HLA class II antigens as biomarkers for CRC in Tunisian population.
Methods: HLA-DRB1 and HLA- DQB1 gene polymorphisms were analyzed in 100 CRC patients and 100 control subjects from Tunisian population.
Results: The most frequent alleles in patients are DRB1 *03 (24%) and DQB1 * 02 (32%) while DRB1* 13 (19%) and DQB1 *03 (40.5%) are the most frequent alleles in controls. DRB1*03 (P=0.046; OR=1.86) and DRB1*12 (P= 0.024; OR= 1.053) are positively associated with higher risk of CRC. In contrast, DRB1*1 1 (P=0.000; OR=0.24), DQB1*03 (P=0.000; OR= 0.303) and DQB1* 05 (P=0.006; OR=0.38) are negatively linked to CRC. DRB1-DQB1 haplotype analysis identified 20 haplotypes in patients and controls. The haplotype *03*02 is the most frequent in patients (11.9%) while *04*03 is the most frequent haplotype in controls (14.3%). The haplotypes DRB1*04- DQB1*04 and DRB1*09- DQB1*02 are positively associated with higher risk of CRC (P= 0.000; OR= 18.80 and P= 0.027; OR= 8.20 respectively). In contrast, the haplotype DRB1*04- DQB1*03 is negatively linked to CCR (P=0.040; OR= 0.49).
Conclusion: DRB 1*03 and DRB 1 * 1 2are associated with higher risk of CRC. However, DRB 1 * 11, DQB 1*03 and DQB 1 * 05 are associated with a decreased risk of CRC**.**
Keywords: Colorectal cancer- HLA class II- genetic susceptibility.

HBO8: Implication of interleukin-17A variants in colorectal cancer susceptibility

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Background and Aim: Interleukin (IL) 17A is pro-inflammatory cytokine produced by Th17 cells, which play key, but sometimes inconsistent role in autoimmunity and cancer. Polymorphic variants in *IL-1 7A* gene were differentially associated with susceptibility to cancer, including colorectal cancer (CRC). We investigated the association between six *IL1 7A* gene variants (rs3819024, rs2275913, rs3819025, rs10484879, rs7747909, and rs3748067) with CRC susceptibility in Tunisians.
Methods: Retrospective case-control study. Study subjects comprised 293 patients with CRC, and 268 age-, gender-, and BMI-matched healthy controls. *IL1 7A* genotyping was done by real-time PCR, with defined clusters.
Results: Of the seven tested *IL17A* tag-SNPs, minor allele frequency (MAF) of rs10484879 was significantly higher in CRC patients than control subjects. Heterozygous rs10484879 [OR (95% CI) = 2.63 (1.64 – 4.21)] was associated with higher risk, while carriage of heterozygous rs3748067 genotype was associated with reduced risk of CRC [OR (95% CI) = 0.56 (0.37 – 0.84)], respectively. Carriage of rs 10484879 minor allele correlated with positive family history of CRC and other cancers (*P*=0.002), CRC staging (*P*=0.044), CRC treatment (*P*=0.038), and with chemo body reaction (*P*=0.001). Of the 7 *IL-17A* variants, 4 were in linkage disequilibrium, hence allowing for construction of4-locus haplotypes. Varied linkage disequilibrium (LD) was noted between the even tested *IL-1 7A* variants, and further analysis was limited to only 4-locus (rs3819024-rs2275913- rs10484879-rs7747909). Haploview analysis identified the 4-locus *IL-17A* haplotypes AG**T**G (*P* <0.011), and GATG (*P* = 0.036) to be positively associated with CRC, after controlling key covariates.
Conclusion: *IL-17A* rs10484879 SNP and*IL-17A* haplotypes AGG**T**G and **GA**G**T**G constitute independent factors of CRC susceptibility. We propose that IL-1 7A may be a target for future CRC immunotherapy.
Key words: Colorectal cancer; susceptibility; interleukin- 17; variants.

HBO9: Application of the Immunomagnetic Separation (IMS) technique prior, the qPCR in Cutaneous Leishmaniasis Diagnosis

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Background and aim: Leishmaniasis is a disease caused by the protozoa *Leishmania* and is most commonly transmitted by infected sandflies. In Tunisia, the leishmaniasis presents two clinical forms: visceral and cutaneous disease. Cutanenous leishmaniasis (CL) is caused mainly by three species (*Leishmania major*, *Leishmania infantum* and *Leishmania tropica*) with an incidence of more than thousands of cases every year. Diagnosis of CL is routinely based on microscopic observation after MGG staining of the skin fluid smears. However, it has low sensitivity although high specificity. Moreover, the culture suffers contamination and is timeconsuming. Molecular diagnosis (namely PCR and qPCR) is highly sensitive and specific but its performance is limited by inhibitors.
The aim of this study is to improve the molecular diagnosis of CL by specifically separating the parasites from biological samples prior the PCR step. Thus, an Immunomagnetic Separation (IMS) tool was optimized.
Methods: A specific rabbit polyclonal anti-*Leishmania* IgG was produced and conjugated to the magnetic Dynabeads sheep anti-rabbit IgG (Novex). The IMS technique was optimized and applied to skin fluid smears collected from CL suspected patients. The complex beadsparasite is then separated by using appropriate magnetic device and heated at 95°C for 5mn prior to be analyzed by kinetoplastic (k) DNA qPCR.
Results: The IMS technique can isolate the two forms of *Leishmania major* parasite amastigote and promastigote.
Detection of *Leishmania* DNA in skin fluid smears samples using of IMS-qPCR showed a sensitivity of 95.74%, a specificity of 100%, a PPV of 100% and a NPV of 94%.
Conclusion: The optimized IMS technique was for the first time described in the CL diagnosis. Applied to biological samples prior the qPCR, it allowed the parasite detection with high specificity and sensitivity.
Keywords: Cutaneous Leishmaniasis, *Leishmania*, IMS, qPCR, kinetoplastic DNA.

TC3-HB/HBO10: Effects of subchronically dietary supplemented high dosing grape seed powder (GSP) in healthy Wistar rat brain

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Background and aim: Grape seed powder (GSP) is a natural compound of *Vitis vinifera* seeds that contain high amount of polyphenol, at the basis of many positive health effects attributed to its antioxidant properties. GSP is regularly used as a dietary supplement that received the "Generally Recognized As Safe" (GRAS) certification from US FDA [1]. So GSP dosing must be optimized to avoid concomitant side effects and to assure bioavailability particularly when targeting the brain.
Methods: The effect of increasing dosage GSP (w/w) from 0.5%, 5%, 10% and 20%, when administered to Wistar rats for 2 months on oxidative stress and inflammatory parameter's biomarker was evaluated. Ki-67 Immuno-histochemical staining was done within the hippocampal dentate gyrus. Statistically, we have analyzed differences between groups based on Statistica.
Results: At the highest dose of 20% GSP had no effect on plasma IL6, decreased the proinflammatory cytokine IL17A, CRP, and increased the anti-inflammatory cytokine IL10 as well as adiponectin. 20% GSP also decreased lipoperoxidation and protein carbonylation into the brain; moreover, 20% GSP increased the reducing power as assessed by non protein thiols (NPSH), increased catalase (CAT) and glutathione peroxidase (GPx) activity and especially up-regulated the Mn isoform within the brain. Above all, GSP dose dependently increased Ki-67 marking within the hippocampal dentate gyrus, culminating at the highest dose of 20%.
Conclusion: GSP appeared as a good candidate for prevention contra neurodegenerative diseases through neurogenesis improvement.
Keywords: GSP; high dosing range; antioxidant; anti-inflammatory; brain
[1] FDA agency response letter GRAS notice n° GRN000 124, http//www.fda.gov/Food/Food ingredients Packaging/Generally Recognized as Safe GRAS/GRAS Listings/ucm 153940; htm, Accessed 1 june (2011).

HBO11: *Streptococcus agalactiae*: antibiotic susceptibility and genotypic characterization of clinical strains isolated from Guelma, Algeria and Marseille, France

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Background and aim: The Group B *Streptococcus* (GBS) or *Streptococcus agalactiae*, commensal bacteria of the urogenital tract and the digestive tract of humans. This bacterium is usually associated with maternal-fetal and neonatal infections. To prevent these infections in newborns, ß-lactams are recommended as a first line of antibiotic-prophylaxis for women in labor, macrolides-lincosamides are therapeutic alternative in cases of allergy to beta-lactams. However, the use of these antibiotics as an alternative prophylaxis is questioned because of the remarkable increase in the rates of resistance. The objective of our work is to study by phenotypic and molecular techniques distribution of the resistance of the strains circulating in Guelma and Marseille.
Methods: In this perspective, 93 clinical isolates of GBS were collected between January 2011 and February 2012 among pregnant women in Guelma and the month of October 2013 until January 2014, at the microbiology laboratory of the Hospital of Timone Marseille. The genetic basis of the resistance of all strains has been characterized by molecular biology techniques.
Results: The analysis of our results showed that the cMLSB phenotype was significantly higher in Guelma isolates versus Marseille, while the M phenotype was associated with Marseille isolates.The total resistance to erythromycin was 34 of 74 (46%) (including 20 of 44 isolates in Guelma (45.4%) against 14 of 30 in Marseille (46.6%). The clindamycin resistance was found in 37 8% ofthe isolates (19 SGB out of 44 isolated Guelma (43.2%) opposite9 SGB out of 30 isolated in Marseille (30%). In contrast, 100% of Guelma isolates were resistant to tetracycline against 86.6% (SGB 26 of 30) Marseille isolates.
Conclusion: This report presents, for the first time, a detailed molecular analysis of phenotypes and genotypes of *S.agalactaie* circulating in Guelma and Marseille. Finally identification of GBS strains by MALDI-TOF MS offers a powerful tool for the biotyping.
Keywords: *Streptococcus agalactiae,* MALDI-TOF-MS, resistance, biotyping, phenotypes and genotypes.

HBO12: Faecal carriage of CTX-M-producing Enterobacteriaceae in Tunisian communities

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Background and aim: The occurrence of extended-spectrum B-lactamase (ESBL)-producing isolates has increased worldwide. Faecal carriage of ESBL-producing Enterobacteriaceae (ESBL-E) has been detected in the nosocomial setting than in the community. We tried to determine the prevalence of ESBL fecal carriage among healthy volunteers in different governorates of Tunisia.
Methods: The prevalence ofESBL-E in 2136 fecal samples from randomly selected Tunisian inhabitants was studied between February 2015 and January 2017. Each faecal sample was cultured in Mac Conkey agar plates supplemented with cefotaxime (2 µg/ml), colonies were identified, characterized, and ESBL phenotype was confirmed by disk diffusion method. ESBL genes were characterized using multiplex PCR-sequencing. The clonal relationship between ESBL-E was studied by pulse-field gel electrophoresis (PFGE). Conjugation experiments and plasmid identification were conducted to examine the transferability of ESBLs genes.
Results: 411(19.24%) healthy volunteers carried ESBL-producing Enterobacteriaceae. Escherichia coli was the predominant species (376,25.86%) followed by K.pneumoniae (31, 7.02%) and C. koseri (0.9%).Most of ESBL-producing E. coli strains belonged to commensal groups A/B1 (252, 67%). 59 (17.5%) of E. coli strains belonged to the virulent phylogenetic group B2, of which 47 (80%) are associated to the multidrug pandemic clone O25b ST131.Among the 47 O25b-ST131 isolates, H30 subclones were identified in 45 strains (85%). Moreover, H30-Rx was the most common lineage among ST131 strains; it was associated with CTX-M15 ESBL in the community setting. PFGE showed a high diversity (>80%) among the ST13 1 strains. The blaCTX-M-15 gene was frequently located in multiple conjugative IncF plasmids.
Conclusion: High ESBL carriage rate of 19% in the Tunisian community indicated by this study implies the possibility of sustained ESBL carriage even among isolated population, which could serve as a reservoir for enriching the ESBL pool in the hospital. Moreover, this study highlighted that the virulent ESBL-ST131 clone is carried by 2,8 % of the Tunisian community.
Keywords: ESBL-producing Enterobacteriaceae, O25b-ST1 31 isolates, blaCTX-M- 15 gene.

HBO13: The role of Otx2 transcription factor in the control of the proliferation of cerebellum granule cell precursors and medulloblastoma

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Background and aim: The homeobox transcription factor Otx2 is essential for the development of the central nervous system. During cerebellum development, Otx2 is expressed by granule cell precursors (GCPs), which have a high proliferation rate. Deregulation of GCPs proliferation may favor oncogenic processes, as seems to occur in medulloblastoma (MB), a malignant and invasive tumor of the cerebellum. While historically relying on histological criteria, recent studies have shown that genomics-based classification of MB better predicts prognosis and provides valuable information about potential drivers and therapies. The current consensus recognizes four major subgroups: Wingless (WNT, Group 1), Sonic hedgehog (SHH, Group 2), and Group 3 and 4. A recurrent genetic alteration in this tumor is the overexpression of Otx2 in 75% of the cases. Our objective is to study the role of Otx2 in the control of proliferation during normal and oncogenic development of the cerebellum.
Methods: We are investigating the role of Otx2 in the control of GCPs proliferation using a mouse genetic model where Otx2 is co-expressed with the green fluorescent protein GFP, allowing identification and purification of Otx2-expressing (Otx2+) GCPs from developing cerebellum using flow cytometry. Purified cell populations are subjected to different tests of proliferation (Ki67 expression, Edu incorporation) to assess the proliferation rate of Otx2+ versus Otx2- GCPs. We tested the oncogenic potential of Otx2 in Myc-medulloblastoma using the medulloblastoma cell line HD-MB03. We performed gain and loss of function experiments to analyze the effect of Otx2 expression on the proliferation of this cell line.
Results: Our results show that « Otx2+ » GCPs have an increased proliferation rate compared to « Otx2- » GCPs, suggesting that Otx2 may have an oncogenic role in the establishment of medulloblastoma through the regulation of GCPs proliferation. The overexpression of Otx2 increases the proliferation rate of HD-MB03 tumor cells, Otx2 silencing significantly decreases it. These results confirm the mitogenic effect of Otx2 on medulloblastoma cells.
Conclusion: Our project shows a pro-proliferative effect of Otx2 on cerebellum and medulloblastoma cells. To shed the light on the mechanism of action of Otx2 in the control of proliferation in cerebellum and medulloblastoma, the determination of Otx2 partners will be essential to provide information about the mechanisms of tumorigenesis, and new potential therapeutic approaches to fight against cancer.
Keywords: Otx2, granule cell precursors, medulloblastoma.

HBO14: Study of *in vitro* stimulation of bronchial epithelial cells (BEAS -2B) with the major allergen (Cup s1) of *Cupressus sempervirens*

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Background and aim: Allergic diseases have a central place in chronic pathologies. For over 20 years, their frequency has been increasing. Allergies to pollen, at present, are a major public health problem because of pollen diversity. However, all the pollens are not allergenic, their nature and quantity vary significantly depending on the region and climatic conditions. Cupressus sempervirens is one of the most widespread species in Algeria with very high allergenic capacity. The aim of this work is to evaluate the stimulatory effect of the major allergen of this species
Methods: The present study was carried out on human bronchial epithelial cells (BEAS-2B) transformed by an adenovirus 12 SV40 hybrid. In this context we are interested in the stimulation, in vitro, of those cells by different doses of the major allergen Cup s1 to test the viability and the release of IL-8 and IL-6. After a series of culture, the cells were exposed for 24 hours at a concentration of 0.02gg/gl, 0.06gg/gl, 0.1 gg/gl, 0.3 gg/gl and 0.9gg/gl of allergen Cup s1 . The viability was assessed by the MTS assay and the assay of cytokine was carried out in the supernatant using the technology Luminex100.
Results: The MTS test showed that cells exposed to different doses were all viable. The release of IL-8 by the cells exposed to different concentrations of Cup s 1 showed a highly significant decrease in the supernatants of cells treated with 0.02gg/gl of the allergen and a highly significant increase was obtained with cells exposed to 0.1 gg/gl, 0.3gg/gl and 0.9gg/gl of the major allergen. However, cell culture with 0.02gg/gl and 0.06gg/gl of the Cup s 1 induced a highly significant decrease of l'IL-6. Conversely, the exposure to high concentrations (0.1 gg/gl, 0.3 gg/gl and 0.9gg/gl) stimulated significant release of this interleukin.
Conclusion: Our experiments showed that the allergen Cup s 1 represents no risk vitality of the cells and had the potential to stimulate the release of IL-8 and IL-6 in a dose-dependent manner.
Keywords: Allergy, Pollen, Cupressus sempervirens, BEAS-2B, IL-8, IL-6.

POSTER PRESENTATIONS
(HBP)

HBP1: Diversity of the genes involved in Algerian families with hearing loss identified by whole exome sequencing

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Background and aim: Hearing loss is a common sensory defect in humans. The majority of congenital cases are attributable to genetic factors. Autosomal recessive nonsyndromic hearing loss is the most common type and accounts for ~80% of cases of inherited hearing loss. Finding the responsible mutations via traditional methods in families with autosomal recessive nonsyndromic hearing loss is difficult due to a high degree of genetic heterogeneity. Whole exome sequencing provides unprecedented opportunities to identify causative DNA variants in Mendelian disorders. The aim of this work is to identify mutations responsible for autosomal recessive nonsyndromic hearing loss in Algerian families.
Methods: We performed whole exome sequencing of 19 unrelated Algerian families affected by autosomal recessive nonsyndromic hearing loss who were negative for mutations in GJB2 (the gene most frequently involved in autosomal recessive nonsyndromic hearing loss in Mediterranean countries).
Results: We found the causative mutations in all the patients analyzed, either in the homozygous state (seventeen families) or in the compound heterozygous state (one family): (c.100C>T : p. (R34*)), (c.821C>T : p.(Pro 274Leu)) and (c.1534C>T : p (Arg512*)) in TMC1, (c.242G>A : p. (Arg81Gln)) in LRTOMT, (c.709C>T: p.(Arg237*)) and (c. 2122C>T : p. (Arg708*)) in OTOF, (c.1334T>G: p.(Leu445Trp)) and (c.2162C>T : p. (Thr721Met)) in SLC26A4, (c.518T>A: p. (Cys173Ser)) in LHFPL5, (c.5336T>C: p. (Leu1779Pro)) in MYO15A, (c.1807G>T: p.(Val603Phe)) in OTOA, (c.6080dup: p.(Asn2027Lys*9)) in PTPRQ, and (c.601 7del: p.(Gly2006Alafs* 13), (c.7 188_71 89ins14: p.(Val2397Leufs*2)) in ADGRV1 . We also identify an homozygous frame-shift mutation (p.Ser339Alafs*15) in EPS8L2, a new gene implicated in progressive deafness in human. Notably, 7 of these 15 mutations affecting 10 different genes had not been reported in other countries.
Conclusion: These results highlight the genetic heterogeneity of nonsyndromic autosomal recessive hearing loss in Algerian families.
Keywords: Algeria, hearing loss, genetic heterogeneity, whole exome sequencing.

HBP2: The absorption probability of iodine 131 beta ray depending on the thickness of the human body materials

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Background and aim: In this work, the beta ray emitted by iodine 131 was simulated with the Monte Carlo method. The absorption probability of beta particles for normal incidence and materials constituting the human body (bone, lung, muscle, blood, eyes, etc.) was calculated. To determine an empirical formula for absorption probability in function of the film thickness, we have suggested a new function depending on the incidence energy, the absorption probability of semi-infinite target and the range. To the best of our knowledge, no theoretical or experimental work on the dependence of the absorption probability of iodine 131 beta ray on film thickness targets has been reported so far.
Methods: In the present work, we used the Monte Carlo method with the Penelope code (Penetration and ENErgy LOss Positrons and Electron) for modeling the beta ray paths within the targets.
Results: It is showed that the absorption probability increases monotonously with the thickness from zero to the critical value corresponding to that of a semi-infinite target.
Our results showed that the deviation between the two probabilities is less than 0.4 % which allows us to continue the calculations based on the average energy of the beta spectrum. Furthermore, may be, this is the reason why in the radioprotection studies they take into account only the average energy of the emitted beta ray.
Conclusion: This work used Monte Carlo Penelope code to calculate the absorption probability of beta rays for iodine 131 as a function of the thickness of biomaterial targets constituting the human body. All the human body could be simulated as water if the thickness is greater than a critical value elsewhere all the human body could be simulated as water except the bones.
Keywords: Absorption probability; Beta radiation; Monte Carlo simulation; Iodine 131

HBP3: Construction of a VHH antibody fragments library from a camel immunized with a diarrheic *Escherichia coli* strain

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Background and aim: Neonatal diarrhea is a major cause of death in herds and is responsible for considerable economic losses. *Escherichia coli* (*E. coli*) is a major etiological agent of diarrhea in young animals when the immune system is not yet developed. It is part of the intestinal commensal microflora of most warm-blooded animals. However, *E. coli* may also be an opportunistic or obligate pathogen, able to multiply and persist in the host's digestive tract by bypassing immune defenses and inducing cellular damage. Few therapeutic options are available, mainly antibiotic therapy, limited by increasing resistance to commonly used drugs. The aim of this work was to develop immunotherapy based on the use of camel VHH antibody fragments, or nanobodies, to target surface antigens of a pathogenic strain of *E. coli*.
Methods: A formalin killed strain isolated from diarrheic camel calf with the virotype: f17/afa/EastI/papC/iroN/iss/iucD and serotype O64 and belonging to the phylogenetic group B1 was used for the immunization of a young dromedary. We tested the immune response by ELISA on living cells and developed a library of VHH antibody fragments by phage display.
Results: By ELISA on living *E. coli* cells we showed that the immunized camel developed an anti-*E. coli* immune response involving conventional IgG1 antibodies and IgG2 and IgG3 heavy chain antibodies. At the end of the immunization we purified the lymphocytes and isolated the RNA and created a VHH antibody fragment library by the phage display technique of about 10^9 individual clones. The panning of these fragments on living *E. coli* cells allowed the isolation of VHH fragments specific for the surface antigens of E. coli.
Conclusion: The identification of these antigens can lead to the development of new diagnostic and therapeutic tools against diarrhea.
Keywords: *Escherichia coli*; diarrhea; nanobodies; phage display.

HBP4: Selective cytotoxicity of arene tricarbonylchromium towards tumour cell lines

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Background and aim: Metals are cytotoxic and could be harmful to human health; however, as their effects are dose dependent, some of them could be used in chemotherapy.
For this reason, we have examined some arene tricarbonylchromium complexes for their cytotoxic activity.
Methods: Four tumour cell lines and one non-tumour cell line Hek293 (Human embryonic kidney cells 293) are treated with some arene tricarbonylchromium complexes in order to select new potential anticancer drugs.
Results: In the present work, mineral chromium (VI) showed that it is cytotoxic on both tumour (MCF-7, HeLa, Hep2 and Caco-2) and non-tumour (HEK293) cell lines. Interestingly, among seven complexes of arene tricarbonylchromium, chromium (0) becomes more efficient in targeting tumour cell lines with less toxicity to non-tumour cells. Three of complexes (formyl benzene tricarbonylchromium 1, anisol tricarbonylchromium 2 and trimethoxybenzene tricarbonylchromium 3) show a decrease of IC50 values for all tested tumour cells compared to the non-tumour cells HEK293. The remaining compounds have an opposite effect; they are less toxic to tumour cells compared to HEK293. The present work demonstrates that some arene tricarbonylchromium are selectively active against cancer cells by inducing apoptosis.
Conclusion: The role of formyl and methoxy groups in arene tricarbonylchromium is shown in complexes acquiring the selective tumour cytotoxicity.
Keywords: Organometallics, Cytotoxicity, Inorganic chromium (VI), Tricarbonylchromium
Tumour cell lines, 50% inhibitory concentration.

HBP5: Anthelmintic activity of *Menthapulegium* essential oil against *Haemonchus contortus* (gastro-intestinal parasite of sheep)

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Background and aim: The development of resistant strains of helminthes, the residues in animal products and the high cost of conventional anthelminthics has generated much interest in the study of medicinal plants as an alternative source of conventional anthelmintic chemical control. *Mentha pulegium*, a medical plant of the family of *Lamiaceae*, known by his medicinal effects. The present study aimed to investigate the putative anthelminthic effect of essential oil of *Mentha pulegium* against gastro-intestinal nematodes of sheep.
Methods: The anthelmintic efficacy of *Mentha pulegium* against *Haemonchus contortus* was performed using *in vitro* assays. For the first assay, The eggs of the parasite were obtained from Barbarine donor lambs experimentally infested with aqueous suspension of H. contortus third stage larvae. *Menthapulegium* at concentrations (0,25; 50; 100 and 200.ig/ml) was added to 200 eggs in 1 ml of PBS. The test tubes were then covered and kept in an incubator at 27 ^{o}C for 48 h. Then, Eggs and larvae were counted. For worm motility test, the lamb was slaughted six weeks post larval intake. The abomasal contents were examinated and adult worms were recovered, and placed per groups of ten actively moving parasites in Petri dishes in contact with (0.25; 0.5 and 1mg/mL) of *Mentha pulegium* essential oil. PBS and Albendazole were used as negative and positive controls. The mortality rate was determined in units of non- motile worms per intervals of 0, 1, 2, 4 and 8 hours.
Results: *Mentha pulegium* has significant anthelminthic efficacy in inhibiting egg hatch at all concentrations. Total inhibition of eggs hatching was observed at the high concentration (200tg/ml) and the assay showed dose-dependent ovicidal activity in all tested concentrations. The exposure of worms to varied concentration of plant was accompanied with different level of mortality varied from 0 mortality at 0.25mg/ml to 100 % for worms in contact during 2 hours with 1mg/ml of *Menthapulegium* oil.
Keywords: Anthelminthic, *H. contortus, Mentha pulegium.*

HBP6: Effect of extracts from *Zizyphus lotus* in the aggregation and biofilm formation of probiotic and pathogenic bacteria

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Background and aim: Nature has served as a rich repository of medicinal plants for thousands of years and an impressive number of modern drugs have been isolated from natural sources, notably of plant origin.
Zizyphus lotus (*L.*), also known as *Jujube*, is a medicinal plant largely found in the Mediterranean region including Algeria . The *Zizyphus lotus* (jujubier) is a fruitier, thorny shrub belonging to the family Rhamnaceae . Commonly called in North Africa "Sedra".
The present study aims to exploit natural bioactive components and evaluation of the effect of extract of *Zizyphus lotus* (*L.*) as prebiotic on the growth and aggregation of some isolated probiotic bacteria and on growth and biofilm formation of pathogenic bacteria.
Methods: The preliminary evaluation of the phytochemical composition of the various treated parts made it possible to demonstrate the presence of some chemical groups. This was confirmed from a quantitative and qualitative analysis by HPLC based on the determination of total polyphenols, flavonoids, and tannins.
Results: Analysis of the different extracts (Aqueous and methanolic) shows that these extracts are richer in polyphenols, flavonoids and tannins.
This extracts was exhibited a prebiotic activity and antibacterial effect, adhesion tests were shown the stimulatory effect of natural extract on probiotic aggeregation and inhibitory effect of biofilm formation of pathogenic bacteria. The percentage of autoaggregation and adhesion of probiotic bacteria increased in presence of aqueous extract.
Conclusion: Finally, all these results obtained *in vitro* are only a first step in the search for substances and natural source biologically active.
Keywords: *Zizyphus lotus* (*L.*), extracts, aggregationon , biofilm, probiotic, pathogenic bacteria.

HBP7: Protective effect of natural antioxidant "curcumin" agains neurobehavioral disorders in the diabetic rats strain wistar

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Background and aim: Type 1 diabetes is characterized by an immune-mediated depletion of β-cells that results in lifelong dependence on exogenous insulin and by too much glucose (sugar) in the bloodstream. It occurs when the pancreas, a gland behind the stomach, does not produce enough insulin. While both type 1 and type 2 diabetes result in hyperglycemia, the pathophysiology and etiology of the diseases are distinct and require us to consider each type of diabetes independently. As such, this position statement summarizes available data specific to the comprehensive care of individuals with type 1 diabetes. The goal is to enhance our ability to recognize and manage type 1diabetes, to prevent its associated complications, and to eventually cure and prevent this disease.
Methods: Diabetes was induced by a single intra-peritoneal injection of streptozotocin at a dose of (45 mg/kg).
Results: The administration of streptozotocin which is revealed, on one hand, the effects of a single streptozotocinintra-peritoneal injection on anxiety behaviors, hematological biochemical and hormonal parameters in adult Wistar rats, and the other hand the protective role of the curcumin on streptozotocin-induced disorders. Behavioral tests in the Open field (OF) and the Elevated plus-maze (EPM) revealed that diabetic animals exhibited an anxious behavior and an alteration in locomotive and exploratory activities when compared to control.
Conclusion: Our problem is to evaluate the behavioral effects of combination diabetesinfection in adult Wistar rat sand the protective effect of an antioxidant (the curcumin) on neurobehavioral alterations and complications. The administration of the curcumin (60 mg/kg) by gastric gavage reduces anxiety and decreases hyperglycemia-related harm.
Keywords: Rats Wistar, Streptozotocin, Diabetes, Curcumin, Neurobehavioural disorders.

HBP8: Molecular characterization of methicillin resistant *Staphylococcus aureus* isolated from clinical cases in east Algeria

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Background and aim: This study was conducted so as to investigate the phenotypic and genotypic characterization of MRSA involved in east Algeria. The confirmation of the species *S. aureus* was performed by amplifying the gene *gyrA.* Resistance to methicillin was performed by detection of *mecA* gene and several virulence factors including toxin of the PVL coding gene as well as TSST coding gene were searched by polymerase chain reaction (PCR).
Methods: This study has included 12 methicillin resistant *Staphylococcus aureus* isolates. All isolates were previously identified as *Staphylococcus aureus* by a standard microbiological procedure and a detection of methicillin resistance was realized by phenotypic methods. Following genomic DNA extraction, the presence of *gyrA, mecA, lukPV, tst* genes was analyzed by duplex PCR. All retained *S. aureus* species have been found to contain *gyr*A gene.
Results: Precise identification of *S. aureus* is important for successful implementation of a staphylococcal infections control program. Therefore, according to the phenotypic, biochemical properties as well as by amplification of the gyrA gene, all of the isolates obtained in this study were identified as *S. aureus.*
Among the 12 strains of MRSA, 10 were mecA positive, indicating that mecA is responsible for methicillin resistance in those strains. Detection of PVL toxin by amplification of the gene luk-PV from extracted
DNA of the strains reveled that five strains from 12 were positive for amplification of the 533 Pb fragment of luk-PVgene. While none strain had the gene tst encoding the TSST.
Conclusion: The pathogenesis of MRSA infections is related to the expression of a wide variety of virulence factors, including PVL and TSST toxins, which might be considered as potential threats especially that most of them have multidrug resistance, rendering it difficult to treat.
Keywords: MRSA, PVL, TSST 1, multidrug resistance, *mecA.*

HBP9: IL-1β and IL-8 serum levels as biomarkers for Colon cancer

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Background and aim: Colon cancer (CC) remains today a major public health problem in the world and especially in Tunisia because of its high frequency and mortality rates. In Tunisia, epidemiological data indicate a marked increase in CC,it is the third cancer in men after lung and prostate cancer, and the second in women after breast cancer.
Interleukins are a group of cytokines that contribute to growth and differentiation, cell migration, and inflammatory and anti-inflammatory responses by the immune system. Some studies have suggested that increased serum levels of these cytokines may predispose to cancer development in various organs. The aim of this study is to evaluate the association of IL-1β and IL-8 serum levels with the risk of CC.
Subjects and Methods: We performed a case control study including 50 patients with CC recruited from Salah Azaiz Oncology Institute, patients were diagnosed by clinical examination and biopsy findings and 50 healthy controls. IL-1β and IL-8 levels were measured by the immunochimioluminescence technique using Immulite 1000R which uses as solid support polystyrene beads coated with anti-interleukin murine monoclonal antibodies (IL-8 and IL-1β).
Results: Results of the statistical study demonstrated a positive association between serum IL- 8 and CC(*P= 0.0036*), serum IL-8 levels were significantly higher in patients this result suggest that IL-8 may play an important role in the onset of CC. Furthermore, serum level of IL-1 βwas comparable between CC patients and controls showing a negative association (*P= 0,2125).*
Conclusion: IL-8 could be involved in the occurrence of CC.
Keywords: Colon cancer, Interleukinn 8, Interleukin 1 β, Serum level, Tunisia.

HBP10: The healing effect of Fenugreek polysaccharide based films on laser burn

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Background and aim: The fractional CO_2 laser is a very common and effective aesthetic resurfacing technique of rejuvenation, designed to correct skin imperfections. However, a successful result depends upon meticulous post-laser treatment care. So far, no evaluation has been reported about the healing effects, after laser application, of different natural products in the literature. On the one hand, many studies involve the use of extract or oil from medicinal plants to accelerate excision, incision and thermal burn wound healing. On the other hand, their uses for influencing wound healing after exposure to laser energy are not well elucidated.
Methods: Edible and biodegradable films of fenugreek (*Trigonellafoenum graecum*) derived polysaccharide (PSF) reinforced by poly (vinyl alcohol) (PVA) were prepared. Four films in the proportion of PSF/PVA: F1 (70/30), F2 (50/50), F3 (30/70) and PVA (100% PVA) were characterized in terms of physical, optical, morphologic, mechanical and barrier properties. Antioxidant activity as well as healing effect of composite films on CO_2 laser fractional burn in a rat model were evaluated.
Results: Results showed that F1 exhibited the higher antioxidant activity. After eight days, the higher percentage of wound healing contraction was observed among the F1 -treated group (100%) followed by the « Cytol basic® » treated group and untreated group (~ 90%). During the treatment, the F1-treated group showed less erythema, less crusting/scabbing, higher general wound appearance scores and a high content of collagen (967,15±57,72 mg/g of tissue) than the other groups.
Conclusion: The current study has shown, for the first time, the healing effect of biodegradable film of fenugreek derived polysaccharide on CO_2 laser fractional burn. Its wound healing effect could be attributed to its antioxidant effect.
Keywords: Fenugreek; Polysaccharide/PVA films; Fractional CO2 laser; Wound-healing; Antioxidant activity.

HBP11: Evaluation of reproductive toxicity in professional environment in paints workers

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Background and aim: Thousands of chemical substances are produced and used in a wide variety of workplaces around the world. Some of these substances may have negative effects on reproductive function in both men and women exposed to them. Over the past twenty years, many studies have demonstrated the harmful effect of certain occupational exposures on certain sperm parameters in exposed employees. 3% of construction workers are exposed to reprotoxic products compared to 2% in other sectors of professional activity.
Methods: This work aims to study the impact of solvent exposure in the field of BTP in workers on some parameters indicative of male fertility. The experiment involved a cohort of 20 workers against 20 unexposed men (controls).Sperm volume, concentration, motility, speed, vitality and sperm malformations, as well as testosterone levels were studied in both groups.
Results: a non significant decrease in sperm volume in solvent-exposed workers compared to controls, a significant decrease in concentration, mobility, speed, vitality of sperm in the worker group compared to controls. Employees exposed to solvents also show a significant increase in the rate of morphological malformations of sperm, as well as the rate of testosterone marked a significant decrease in workers compared to controls.
Conclusion: To protect the reproductive function of all workers, exposure to chemicals, radiation, biological agents and stressful working conditions must be eliminated or at least reduced as much as possible. Mutagenic, teratogenic and carcinogenic substances should be completely eliminated or isolated so that they do not come into contact with the workers and the working environment. Some industries have adopted a variety of general approaches to protect the reproductive function of workers from workplace exposures. However, many of these approaches are undesirable and are actually discriminatory.
Keywords: Reproductive, Toxicity, Workers, paints, solvents.

HBP12: Evaluation of morphometric and histological parameters in malnourished rats: impact of realimentation with fermented wheat of the Hamoum type

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Background and aim: Historically, traditional fermented wheat such as Hamoum (FWH) has been the subject of a preventive product against many intestinal physiopathological complications. The aim of this work is to verify whether the realimentation protocol with a Hamoum fermented wheat-based diet (FWH) has a beneficial effect on the recovery ofweight growth, morphometry of the intestinal organs and repair of the intestinal mucosa in young malnourished rats.
Methods: We used 48 4-week-old male Wistar rats, divided into five groups. The first group is the control group received a balanced standard diet (20% casein), the second group is the malnourished group (Mal) received a protein-deficient diet for 28 days (2% casein). The third, fourth and fifth groups constitute the malnourished and realimented groups with WFH, non-fermented wheat-based diet (NFW) and control diet (Mal/C) respectively. We evaluated the weight growth of rats, the weight of intestinal segments, the measurement of intestinal villus height and the number of intraepithelial lymphocytes (IEL).
Results : The results obtained show that protein malnutrition induces a very significant loss in the total weight of rats, a decrease in the fresh weight of the intestinal organs, a shortening of the villi suggesting the presence of villositary atrophy as well as a significant infiltration of intraepithelial lymphocytes compared to the control group ($p<0.001$). On the other hand, we observed a significant weight gain in the realimented group with WFH ($p<0.01$), a significant increase in villus height with a decrease in the number of intraepithelial lymphocytes ($p<0.01$). The recovery of the weight of the intestinal segments is strongly observed in the resupplied group with WFH compared to the resupplied groups with NFW and Mal/C.
Conclusion: WFH in its natural state and through its fermentation processes by its endogenous bacterial flora could contribute to the proper functioning of the physiological mechanisms of the intestine in situations of severe protein malnutrition in children.
Keywords: Protein malnutrition, Villus height, IEL, Fermented wheat type Hamoum.

HBP13: Evaluation of the hematotoxicand hepatotoxiceffect of paint solvents in professional environment

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Background and aim: Painting, cleaning metals or textiles, stripping, making perfumes ... many workers are in contact with solvents. Regular exposure, even at low doses, can lead to more or less long-term health damage, some of which is irreversible. Priority must be given to the substitution of dangerous solvents. Solvent exposure is very common in both work and home environments. The toxic effects common to all these compounds are centered on mucocutaneous, hematotoxic and hepatotoxic lesions. The removal of the most toxic solvents, the limitation of emissions and the use of collective and individual protection means are the primary prevention rules that are required. Chemicals, used in addition to our basic products, have significantly improved our quality of life. They eliminate insects for example fluidify the paintings, isolate the houses or even preserve the food. However, these products can be toxic and therefore dangerous for health.
Methods: This work aims to investigate the effect of exposure to paint solvents in building workers in the El Tarf region (north east of Algeria). The study consists to evaluate the variations inhepatotoxic (glucose, LDL cholesterol, total protein, ASAT, ALAT) and hematotoxic parameters (the number of red blood cells, neutrophils; hemoglobin, as well as hematocrit) in workers compared to controls.
Results: Several metabolic and cellular perturbations were observed in the exposed workers compared to the controls, which testify the hematotoxicity effects (decrease in the number of red blood cells, neutrophils; hemoglobin, as well as hematocrit) and hepatotoxicity effects (increased glucose level, LDL cholesterol, total protein, ASAT, ALAT) of solvents.
Conclusion: The whole problem is to determine to what degree a product is defined as hematohepatotoxic and what risk we are willing to accept to benefit from the usefulness of these substances. In industry and especially in the building industry, water-based paints replace more and more often solvent paints.
Keywords: Solvent, Exposure, Health, Workers, Toxicity.

HBP14: Determination of the mutational profile of genetic diseases in Tunisia through the analysis of Whole Exome Sequencing data

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Background and aim: Genetic diseases in Tunisia are a real health public problem given the lack of knowledge concerning their epidemiological data and chronicity as well as the deficiency of patient health care system. This problem is undoubtedly increased by the high rates of consanguinity in the Tunisian population. Hence, our aim was to review and provide a catalogue of genetic data related to this kind of diseases identified in our population.
Methods: Data were collected and updated through a systematic and combined strategy of manuel text mining of published data on Pubmed as well as from gray literature. Collected data on genetic diseases were then classified according to the transmission mode and the World Health Organization International Classification of Diseases.In order to enrich the mutational spectrum of the Tunisian population, we developed a bioinformatic pipeline forpotentially deleterious variant identification from whole exome sequencing (WES) data of our laboratory. Sanger sequencing was then performed to confirm one pathogenic variant in a patient.
Results: The spectrum of genetic diseases is assessed to more than 540 pathological entities among which 60.1% are autosomal recessive, thus reflecting the role of consanguinity in their expression. The CIM-10 classification ofthose diseases showed that the most prevalent group is of congenital malformations. The determination of the mutational spectrum revealed more than 800 mutations have been identified for these diseases. The exome analysis allowed us to identify 52 additional pathogenic variants. Among these, a variant located in the*AIRE* gene was identified in a patient already affected with XPA. This variant was confirmed by Sanger sequencing and could be the cause of the expression of a second pathology for the patient.
Conclusion: This study is a valuable resource for the human genetics community by offering an overview of Mendelian disorders in the Tunisian population. The availability of the database to the scientific community not only will improve the knowledge of these disorders but also help to prevent them.
Keywords: Genetic diseases, Tunisian population, mutation, exome, bioinformatics, database.

HBP15: Identification of a novel missense mutation in NIPAL4 gene in a Tunisian patient with Lamellar Ichthyosis

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Background and aim: The ichthyosis is a group of cornification disorders characterized by abnormal differentiation and desquamation of the epidermis. Lamellar ichthyosis (LI) is a rare and one of the most severe forms of ichthyosis and is inherited in an autosomal recessive state. Several genes were described to be responsible of these ichthyosis forms such as *TGM*1, *ABCA12*, *ALOX12B*, and *NIPAL4*. The *NIPAL4* gene is located on chromosome 5q33.3 and believed to encode a magnesium transporter membrane associated protein involved in epidermal lipid processing and in lamellar body formation. This protein is belonging to a large group of membrane Drug/Metabolite transporters (DMT super family) sharing homology structure with G-protein coupled receptor.
The aim of this work is to search the causative mutation in a patient with sever clinical features of lamellar ichthyosis and belonging to a consanguineous Tunisian family.
Methods: The patient clinically diagnosed for lamellar ichthyosis and his parents were tested as well as 50 Tunisian healthy individuals. After the agreement of the ethics committee and the individuals' consent, blood samples were collected and DNA extraction was performed following the standard phenol–chloroform method. The six exons of *NIPAL4* gene and their flanking exon–intron boundaries were amplified. Each exon was sequenced on both strand in an ABI PRISM 3100-Avant automated DNA sequencer using the BigDye Terminator Cycle Sequencing reaction kit v1. 1. Several bioinformatic tools were also used to predict the variants pathogenicity.
Results: The sequencing analyses of*NIPAL4* gene revealed that our patient carried a novel missense mutation in the exon 4. This mutation was in homozygous state in the patient which was inherited from his heterozygous parents and was absent in 50 tested controls. Bioinformatic tools predicted the pathogenicity of this novel mutation and support its involvement in the phenotype of our patient.
Conclusion: We identified a new homozygous missense mutation in exon 4 in *NIPAL*4 gene in patient with lamellar ichthyosis. Its pthogenicity predicted by several bioinformatic tools and its absence in controls support that it is a causative mutation in our patient.
Keywords: Lamellar Ichthyosis, *NIPAL*4 gene, missense mutation.

HBP16: Evidence of altered cholesterol metabolism associated with oxidative stress in Tunisian demented patients

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Background and aim: Cholesterol metabolism is reported to be altered in several neurodegenerative diseases, including Alzheimer disease (AD) and vascular dementia (VD). In addition, the pivotal role of oxidative stress in brain degeneration is widely accepted, and its increase may cause adverse effects in the cellular macromolecules. Thus, we intended to explore the eventual association between cholesterol metabolism and oxidative stress via the measurements of cholesterol precursor' s levels as well as cholesterol derivatives (oxysterols), resulting from auto-oxidation of cholesterol in oxidative stress conditions, in the periphery of demented patients.
Methods: A case-control study was conducted in 70 individuals (33 demented patients and 37 controls). All the participants (age>60) were classified according to the DSM-IV diagnostic criteria for dementia. Demented patients were stratified into 2 groups; Alzheimer disease group (AD, n=20), vascular dementia group (VD; n=1 3). Plasma levels of cholesterol precursors, lanosterol and lathosterol, as well as oxysterols, especially those oxidized in C7; 7-ketocholesterol (7-KC), 7a-hydroxycholesterol (7a-OHC) and 7f3-hydroxycholesterol (7f3- OHC), were quantified in demented patients by gas chromatography coupled with mass spectrometry.
Results: Analysis of cholesterol precursors revealed a significant decrease in lanosterol level in both AD and VD groups compared to the controls. Besides, a significant positive correlation was observed between the dementia severity, evaluated by MMSE score, and lanosterol level in AD group ($p<0.05$). While no significant variations of 7a-OHC and 7-KC were observed, analysis of oxysterols showed an accumulation of 7f3-OHC in plasma of patients with AD ($p<0.05$).
Conclusion: Our data support the altered cholesterol metabolism in demented patients. Thus, we could suggest a cholesterol synthesis deficiency associated with an increase of its autoxidation.
Keywords: Alzheimer disease, dementia, oxysterols, lanosterol, lathosterol, Cholesterol.

HBP17: Evaluation of pro-inflammatory cytokines in frailty among Tunisian older adults

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Background and aim: Frailty is a multidimensional disease based on the interaction of psychological, physical, and biological factors. Since inflammation has been supported as an important factor in ageing and age-relateddiseases. The present study was undertaken to evaluate serum levels of pro-inflammatory cytokines (Tumor necrosis factor-a (TNF-a), Interleukin-8 (Il-8) and Interleukin-6 (Il-6)) in frail patients and to precise the relationships between inflammatory markers and biochemical or geriatric parameters.
Methods: We conducted a cross-sectional study in a population of Tunisian older adults (N=141, aged 65 and over). The participants were classified according to the short emergency geriatric assessment (SEGA m) on 51 very-frail patients, 40 frail patients, and 50 non-frail patients. An ELISA test was used to analyze serum cytokines (TNF-a,Il-8,Il-6) levels.
Results: TNF-a levels were significantly higher in the group of very-frail subjects than in frail and non- frail subjects (28.71 [27-32.78] pg/ml vs. 25.54 [24.23-28.03] pg/ml vs. 21.16 [25.33-30.97] pg/ml, respectively, p= 0.003,). Similarly, Il-8 levels were significantly higher among very-frail subjects compared with frail and non-frail subjects (22.3 1 [12.63-32.98] pg/ml vs. 9.69 [6.66-17.95] pg/ml vs. 19.91[11.34-24.95] pg/ml, respectively, p< 0.001). An additional marker that was significantly elevated in very-frail group was CRP (p=0.01). No significant differences in Il-6 levels were detected among frailty groups (p=0. 118).
When inflammatory markers values were analyzed for biochemical and geriatric parameters among frailty groups , negative correlations were found between albumin and serum levels of Il-8, Il-6, and CRP (r = -0.240, r =-0.502, and r = -0.563, respectively); ADL scores and serum levels of TNF-a, Il-8, Il-6, and CRP (r = -0.257 , r =-0.265, r = -0.211 and r = -0.3 , respectively).
Conclusion: The key finding of this study collectively supports the role of pro-inflammatory cytokines, especially TNF-a, CRP, and Il-8 in the development of frailty.
Keywords: Frailty, Inflammation, Tumor necrosis factor-a, Interleukin-8, Interleukin-6, Creactive protein.

HBP18: The epidemiology of obesity

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Background and aim: Obesity is a complex metabolic disease. It has become in a few years one of the most significant public health problem according to the World Health Organization reports. The most useful population level measure of obesity is the body mass index (BMI). Measured BMI values are used to define whether an individual is considered to be underweight, healthy, overweight or obese. The prevalence of overweight and obesity has escalated dramatically during last decades in both developed and developing countries.
The origin of the problem is multifactorial and complex. Both genetic and environmental factors are involved in the etiology of obesity.
Methods: Our population included obese and control subjects. Anthropometric and biochemical parameters were determined according to standard protocols using enzymatic methods with commercially available kits.
Results: Obese patients had more individuals with hypertension, dyslipidemia and diabetes. Higher levels of fasting glucose, triglyceride, serum total cholesterol, serum HDL-C, CRP and the mean of the BMI was found in patients than healthy subjects ($p<0.05$). However no difference in age, sex distribution, smoking percentage and LDL-C was shown in our results. Obesity carries significant health implications, including diseases such as diabetes, hypertension, dyslipidemia, cardiometabolic complications, sleep apnea, respiratory problems, hepatic and renal dysfunction.
Conclusion: Obesity rates across Tunisia are alarming. It is important to continue to study the problem of obesity in Tunisia to understand the etiology to help guide future public health strategies and effective interventions to curb rising obesity rates. Without these, the costs of obesity will place a huge financial burden on the public health system.
Keywords: overweight, obesity, public health, Tunisia.

HBP19: Evaluation of Hepatic and Renal dysfunction following Lufenuron administration in Wistar rats

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Background and aim: The widespread use of pesticides in public health and agricultural programs has caused severe environmental pollution and potential health hazards, including acute and chronic cases of human poisoning. Lufenuron is relatively a new member of the benzoyl phenylurea class of chitin-synthesis inhibitors «IGR » (insect growth regulator). Liver and kidney are known as the most important target organs for xenobiotic compounds including environmental pollutants. For this reason, the main objective of our work is based on the study of disturbances in the functioning of the hepatic and renal system following exposure to lufenuron administered by gavage to Wistar rats for 60 days.
Methods: The experiments were conducted on female rats Wistar which were treated daily through orally gavages during 60 days. The animals were divided into two groups: the control group received corn oil while group II received lufenuron. After sacrifice, the blood samples were collected from each rat 2 ml was taken in a glass tube without EDTA and left for 20 minutes to coagulate at room temperature, and then centrifuged at 3000 rpm for 10 min to obtain serum samples used for biochemical assays. The parameters are: transaminases (aspartate aminotransferase (ASAT) and alanine aminotransferase (ALAT)), urea, uric acid and creatinine.
Results: Our work reveals a considerable increase in the plasma levels of creatinine, urea and uric acid, also an elevation in serum AST, ALT activities.
Conclusion: The present study suggests that exposure to lufenuron causes disturbances in renal filtration function and it causes severe acute liver damage in rats treated with lufenuron, a sign of the toxicity of this pesticide.
Keywords: Insect growth regulator, lufenuron, liver damage, renal filtration, toxicity.

HBP20: Influence of moderate leukocytes on sperm nuclear DNA and on seminal interleukin (IL-6 and IL-8) in infertile Tunisian men

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Background and aim: Infertility is defined as the failure of conception after 12 months of sexual relation without the use of contraceptives, affects around 15% of couples in reproductive age, and the male partner is responsible for up to 50% of these cases. Leukcytospermia, present in about 10 to 20% of infertile men is often associated with impaired quality of sperm DNA and might indicate inflammation of the genital tract and influence semen quality. The aim of this work is to confirm the effect of leukocyte on DNA integrity and to determine the IL-6 and IL-8 levels in the seminal plasma of normal and leukocytospermic Tunisian infertile men.
Methods: The amount of sperm nuclear DNA fragmentation was investigated using the chromatin dispersion test and Cytokines secretion were performed on seminal plasma using enzyme-linked immunosorbent assay.
Results: After the sperm chromatin dispersion test procedure, sperm without DNA fragmentation showed haloes of chromatin emerging from the central nuclear core, whereas these haloes appeared small or absent in those sperm containing fragmented DNA. The incidence of DNA damage increased to 31,41% in leukocytospermic group compared to non leukocytospermic group 2 (14,68 %) and this was statistically significant (p<0,001)
Moreover, Sperm DNA fragmentation was negatively correlated with percentage of motility (r= -0,342; p<0,001), sperm vitality (r=-0,334; p=0,001) and positively correlated with concentration of leukocytes (r= 0,222; p=0,024). Also, IL-6 and IL-8 levels were higher in group with leukocytospermia than group without leukocytospermia but not significantly (r= 0,46) and (p= 0,12) respectively and IL-8 was positively correlated with DNA fragmentation (r= 0,42; p< 0,001).
Conclusion: These results confirm the negative effect of the leukocytospermia on semen parameters and on sperm nuclear DNA. Furthermore, inflammatory mediators (Il6, Il8) may be a direct cause of DNA fragmentation of ejaculated spermatozoa and can decrease the fertilization capacity of germ cells
Keywords: Leukocytospermia, Nuclear DNA, IL-6, IL-8, Male infertility.

HBP21: IL-17A play a pivotal role in host defence against *Echinococcus granulosus* infection. Approach diagnostics and therapeutic

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Background and aim: Human cystic echinococcosis is a widely endemic helminthic disease; caused by the larval stage of *Echinococcus granulosus.* It constitutes a serious public health problem in various parts of the world, particularly in Algeria. We have previously shown the role of cytokines Th1, Th17 and Th2 in human Hydatidosis. The present studies aimed to identify the involvement of IL-1 7A in host defense against *Echinococcus granulosus* infection.
Methods: In this way, we investigateIL-17A, IL-6**,** TGF-βand NO production in sera from Algerian patients with cystic echinococcosis and in supernatants culture of peripheral blood mononuclear cells (PBMCs) from the same patients stimulated by a major parasitic antigen. In the same way, the expression of STAT3 and inducible NOS (NOS2) was measured in PBMCs of patients. We also studied NO modulation by IL-17A and TGF-β in PBMCs and monocytes cultures from patients in presence of *Echinococcus granulosus* protoscoleces (larval form of parasite). Protoscoleces viability was assayed microscopically using eosin staining.
Results: Analysis of cytokines and NO production revealed that the levels of IL-17A, IL-6 and NO were significantly higher in all sera and PBMC culture supernatants from patients($p<0.001$).Interestingly, ourresults show a significant positive correlation between NO and IL-17A levels. We observed with interest that both STAT3 and NOS2 expression was up regulated in PBMCs. We noted with interest that co-cultures treatment with IL-17 caused an increased NO production and a decrease in the percentage of viable protoscoleces. However, our results indicate that IL-17A, IL-6 and NO levels was very low in sera and supernatants of PBMC cultures from relapsing patients.
Conclusion: Collectively, our results indicates that the IL-1 7A play a relevant role in host defense against *Echinococcus granulosus*. Our findings may provide an alternative approach to the diagnostics and treatment of patients with hydatic disease.
Keywords: Human cystic Echinococcosis; Interleukin-17A; Nitric oxide;diagnostics; immunoprotection.

HBP22: Expression profile of two mature microRNAs from formalin-fixed paraffin-embedded samples of Tunisian patients with lung adenocarcinoma

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Background and aim: MiRNAs have been identified as potential biomarkers for diagnosis and clinical prognosis of lung cancer, but microRNA signatures varied between different populations and histological subtypes. Adenocarcinoma (AD) is the most common subtype of lung cancer, the leading cause of cancer deaths in the world which urgently needs biomarkers to aid patient management. Here we undertook the first investigation on microRNAs expressions in Tunisian lung adenocarcinoma (LAD) patients.
Methods: MicroRNAs expressions were measured in cancerous and normal lung formalinfixed paraffin-embedded (FFPE) tissue strictly collected from Tunisian LAD patients, who had not been treated with chemotherapy or radiotherapy.
Results: Our findings showed that miR-21 level in patients with lung adenocarcinoma were significantly up regulated, than those in the control group ($P = 0.03$).However, no significant differences were observed in the expression level ofmiR-200a between the cancer and the control groups ($P = 0.192$).The associations with the clinicopathologicalfeatures showed that miR-21 overexpression was associated with the tumor stage and the tumor size ($P=0.002$, 0.003 respectively).
Conclusion: Our results identified three microRNAs as potential diagnostic markers for LAD among Tunisian patients.
Keywords: LAD; microRNAs; FFPE, biomarkers.

HBP23: *In vivo* evaluation of resorption and cell viability of human fresh amniotic membrane using mice subcutaneous implantation model

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Background and aim: The placenta is an easily accessible source of stem cells. The human amniotic membrane is an abundant and readily obtained tissue, it that has a long history in clinical utility; it has been proposed for cell replacement therapy and tissue regeneration. The human amniotic membrane could be an important source of scaffold. The purpose of our study is to evaluate the resorption of fresh human amniotic membrane before implantation in vivo and the viability of its stem cells.
Methods: Placentas were collected under sterile condition from elective caesarean sections of healthy women. Immediately after surgery, fresh amniotic membrane were carefully detached from chorion and rinsed with ringer solution to clean all blood remnants and then stored in ringer solution containing penicillin G (50 mg/mL), streptomycin (50 mg/mL), tobramycin (100 mg/mL) and amphotericin B (2.5 mg/mL) at 4◦C at least for 2 hours . 3 x 3 cm pieces of fresh human amniotic membrane were implanted subcutaneously in the dorsal of mice. Twenty five mice were divided into 5 groups each group was subdivided (n = 5) according to the time interval to sacrifice (5, 10, 20, 30, 40, 60 days). Macroscopic observation, histology (HE staining) and Immunohistochemistry (anti -mitofilin antibody) were evaluated.
Results: The macroscopic and histological results of the present study demonstrate that after implantation fresh human amniotic membrane was present in all groups with significant decrease of its size at 60 days. Immunohistochemistry analysis showing that amniotic cells were still alive at day 10, 20 with high percentage than decreased at day 30 to be absent at days 40 and 60.
Conclusion: After its implantation, the complete resorption of the human fresh amniotic membrane was longer than 60 days while the duration ofviability ofits cells was 30 days.
Keywords: Placenta, stem cells, human amniotic membrane, resorption, *in vivo.*

HBP24: Protective effect of Lentisk's extracts on oxydatif stress, anti-proliferative and anti-bacterial activities

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Background and aim: Ethnopharmacological relevance: Pistacia Lentiscus has been widely used in the Tunisian traditional medicine, for its wide range of medicinal properties. Experimental studies carried out on this plant have demonstrated various biological and pharmacological activities of Various parts, including its fruit, seed, bark, leaves, and root: venous-lymphatic decongestant, antispasmodic, antioxidant effect, anti-ulcerative anti-fungal activity, anti-parasitic, anti-vaginal trichomonas and Ocimum Basilicum, astringent, expectorant, cicatrisant and especially anti-cancer activity... Lentisk is a good source of compounds such as conjugated, poliphenol, gallic acid, linoleic acid and punicic acid... This phenolic compounds are the cause multiple effects. The aim of this study was to evaluate the effect of Lentisk on oxidative stress in liver of rats and prove the anti-cancer and anti-bacterial effect of this plant.
Methods: The trial of oxydatif stress was carried out in the Higher Institute of Biotechnology of Beja. The anti-cancer activity was made in the Pasteur Institute and anti bacterial activity was realized at the biotechnology center of Borge Cedria.
Results: The analyses of oxidative stress were assessed by histopathological and biochemical examinations. Results in vivo showed that the administration of TAM anti-cancer treatment caused an hepatic histological alterations accompanied by an oxidative stress status in rats. Indeed, animals receiving the TAM treatment (2^{cd} group) registered an increase by 43.75% ($p<0.05$) of methylenedioxyamphetamine (MDA) and peroxide hydrogene (H_2O_2) in liver comparatively with the control group (First group). Animals receiving TAM and Lentisk (Fourth group) showed a decrease ($p<0.05$) in MDA and H_2O_2 as compared to the 2^{cd} group treated only with TAM.
Indeed, The antioxidant enzyme in the TAM group (2^{cd} group) registered an increase by 68% ($p<0.05$) of Glutathione Peroxidase (GPx) and Catalase (CAT) in liver comparatively with the control group. Animals receiving TAM and our plant (4^{th} group) showed a decrease ($p<0.05$) in GPx and CAT as compared to TAM group.
Moreover, our result demonstrate a strongly anti-proliferative effect of Lentisk extracts on MDA-MB-231 breast cancer cell lines.However, the antibacterial activities of plant extracts were also improved on few selected bacteria: Enterococcus fecalis, staphylococcus aureus, salmonelle enteritidis, pseudomonnas aeuroginosa, E.coli listeria mnocytogene.
Conclusion: It was concluded that, Lentisk methanolic extracts presented an antioxidative effect, antibacterial and anticancer activity.
Keywords: Lentisk, rat, antioxidant activity, anti-cancer activity, anti-bacterial activity.

HBP25: The Phylodistribution of Drug Resistance in *Ureaplasma* spp. and *Mycoplasma hominis* inTunisia

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Background and aim: Antimicrobial resistance in a number of bacterial pathogens has been shown to evolve clonally. Data about the phylogenetic clustering of antibiotic resistance among genital mycoplasma strains in Tunisia are limited. The aims of this study were to assess the antimicrobial susceptibility of genital mycoplasmas (*Ureaplasma* spp. and *Mycoplasma hominis*) and to investigate the phylodistribution ofresistant strains.
Methods: This study included 101 molecularly typed *Ureaplasma* spp. (serovars 1, 2, 3, 4, 5, 6, 8, 9, 10, 12 and 13) and 59 *M. hominis* (30 expanded sequence types, eSTs) clinical strains isolated in the time period 2000-20 17. The antimicrobial susceptibility was tested against nine antibacterial agents using the broth microdilution method. Neighbor-joining and minimum spanning trees (NJT and MST, respectively) were reconstructed to establish the phylogenetic relationships among isolates.
Results: *M. hominis* isolates were uniformly resistant to azithromycin and erythromycin but susceptible to doxycycline, ofloxacin, ciprofloxacin, levofloxacin, moxifloxacin and josamycin. Resistance to tetracycline was observed among 37.28% of these isolates. Interestingly, the MST showed that tetracycline resistance was associated with eST3, eST6, eST14, eST17, eST20, eST21, eST23, eST24, eST25, and eST26. *Ureaplasma* spp. isolates were uniformly resistant to ciprofloxacin and erythromycin, intermediately resistant to azithromycin, and susceptible to doxycycline, moxifloxacin and josamycin. Ofloxacin and levofloxacin resistance was found in 73.27% and 17.82% of *Ureaplasma* spp. isolates, respectively, while 37.62% of isolates were resistant to tetracycline. Consequently, we detected an elevated multidrug resistance (MDR) rate among *Ureaplasma* spp. strains (37.62%). The NJT revealed that the majority of serovars 2, 5, 8, 9 (77.77%) were MDR, followed by serovars 4, 10, 12, 13 (52.63%) and serovar 3 (48.57%). However, 15.79% of serovar 1 and only one isolate of serovar 6 were MDR.
Conclusion: The present study provides valuable information on the antibiotic susceptibility of genital mycoplasmas and may aid clinicians to prescribe the adequate antimicrobial therapy. Collectively our data lend support to a clonal expansion of antibiotic resistance. This is likely to have important implications in monitoring the spread of drug resistance among genital mycoplasmas.
Keywords: *Ureaplasma* serovars, *Mycoplasma hominis*, Multi-drug resistance.

HBP26: Genetic and mutational heterogeneity in FA Libyan Patients

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Background and aim: Fanconi anemia (FA) is a rare inherited disease characterized by developmental defects, short stature, bone marrow failure, and a high risk of malignancies. FA is heterogeneous: 20 genetic subtypes have been distinguished so far. A clinical diagnosis of FA needs to be confirmed by testing cells for sensitivity to cross-linking agents in a chromosomal breakage test. As a second step, DNA testing can be employed to elucidate the genetic subtype of the patient and to identify the familial mutations. Fanconi anemia (FA) is among the diseases for which the incidence is increased by consanguinity and endogamy. Unfortunately, FA has not been so far investigated in North Africa, except for patients from Tunisia and Egypt. In the present study, we aimed to identify the FA genetic basis in FA Libyan patients in order to develop a strategy for molecular diagnosis applicable to routine clinical use.
Methods: Due to the absence of adequate infrastructure in Libya, 6 patients from 5 unrelated families were referred to our cytogenetic department for FA diagnosis confirmation using clastogen-induced chromosomal breakage test. Following a positive cytogenetic result, molecular analysis of *FANCA* gene was performed using Sanger sequencing of all the 43 coding exons, Multiplex Ligation-dependent Probe Amplification (MLPA), Targeted Gene Sequencing and Whole Exome Sequencing based on Next Generation Sequencing (NGS) techniques.
Results: Causal mutations for all the investigated Libyan FA patients were identified. Five patients carried deleterious variants in *FANCA* gene at a homozygous or a compound heterozygous state: a stop gained mutation c.2749C>T and a missense mutation c.2426G>A found at heterozygous state for two patients; two missense mutations c.1258G>A and c.1304G>A found at homozygous state for two patients and the deletion of exon 15 at homozygous state for one patient. One patient had co-occurrence of DSD (Disorder of Sex Development), COFS (cerebro oculo facio skeletal) syndrome and FA, the latter being due to *FANCJ* mutation.
Conclusion: In the present study, we report for the first time the mutation spectrum of FA in Libyan patients. Due to founder effect, some of these mutations could be shared between several families. Consequently, direct screening for these mutations could provide an accessible tool for diagnosis confirmation. Despite the relatively small number of the studied patients, our results show the genetic and mutational heterogeneity of the disease in this population. Due to the long tradition of consanguinity, the co-occurrence of several inherited disorders in the same patient should be taken into consideration for an appropriate genetic counseling.
Keywords: Bacterial communities, Diversity, Herbicide, Fusilade, Rhizosphere.

HBP27: Evaluation of the subacute toxicity of fruit *Citrullus colocynthis* methanolic extract in male albino rats

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Background and aim: The *Citrullus colocynthis* (CCT) fruit in its dried or fresh forms is consumed by some patients of M'sila region for its antidiabetic, antijaundice, antirheumatic, antiheamorrhroids activities without considering its safety. In this study, we investigated the effects of the subacute administration of the methanolic fruit *Citrullus colocynthis* (CCT) extract in male Albino rats.
Methods: The acute toxicity study was undertaken firstly to determine the acute LD_{50} of the extract. The second step was undertaken to evaluate the toxic effects of the same extract after its oral administration for six weeks (265 mg/kg) to male albino rats.
Results: The LD_{50} of the extract was found to be 1311.45 mg/kg. The subacute study showed a significant reduction in the final body weight and in the relative organ weights of treated rats. These changes tended to return to normal state after stopping the treatment for one week. RBC, HGB, HCT and PLT values of the control group were significantly higher than those of the treated groups. Biochemical analysis revealed a significant elevation in GOT, GPT and ALP activity as well as in urea, uric acid, creatinine and electrolyte levels (Na^+, K^+, PO_4^- and Ca^{+2}) for the first treatment group. Such modifications were totally absent in the second group. A congestive liver and kidneys in the treated animals supported all these modifications.
Conclusion: The present study showed that the intake of the ripe *Citrullus colocynthis* fruit extract presented some adverse effects on the liver, kidney and bone marrow functions in rats.
Keywords: *Citrullus colocynthis*, LD_{50}, Methanolic extract, toxicity.

HBP28: The new model to calculate the beta dose: medical application

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Background and aim: we have determined the deposited beta energy in human body in function of the emitted beta ray. In fact, we have calculated the minimum mass of the human body (to calculate the dose of ^{131}I spectrum beta ray) which allows the use of the above model. We assume that, is valid for all substances constituting the human body. The obtained results are compared with other published works.
Methods: In the present work, we used the Monte Carlo method with the Penelope code (Penetration and ENErgy LOss Positrons and Electron) for modeling the beta ray paths within the targets. Our simulation parameters of the input file used in Penelope code was included as follows:
SIMPAR 1.0e2 1.0e2 1.0e2 0.05 0.05 1.0e2 1.0e2 [EABSs,C1,C2,Wcc,Wcr]
where: *EABSs*: the energies where (electron, photon and positron) are assumed to be effectively stopped and absorbed in the medium. *C1, C2*: the allowed values of the elastic-scattering parameters C1 and C2 are limited to the interval [0,0.2]. Wcc, Wcr: The cutoff values; we consider inelastic collisions with energy loss W < Wcc and emission of bremsstrahlung photons with W < Wcr as soft stopping interactions
Results: We suggest that the average loss energy of backscattered beta ray (primary and secondary) is the half of the incidence energy of emitted beta ray inside the semi-infinite target. The deposited energy compared to that of Penelope code shows a good agreement (the deviation between our results and those of Penelope code is less than 1.1%) which is the proof of the validity of our model.
Conclusion: Besides, we have showed that the deposited energy (used to calculate the beta dose) could be given by <E>=0.5 (Abs+1)E where E is the incidence energy and E is the absorption probability of semi-infinite target. In fact, we have presented the minimum mass of the human body (to calculate the dose of ^{131}I spectrum beta ray) which allows the use of the above equation.
Keywords: Beta radiation; Beta dose; Monte Carlo simulation; human body.

HBP29: Anthelmintic activity of Tunisian *Artemisia campestris* essential oil

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Background and aim: Helminth infections caused by gastrointestinal nematode (GINs) are a major health problem for small ruminants' production. *H. contortus* is one of the most important nematodes. The control of gastro-intestinal nematode (GIN) in livestock is mainly treated with anthelmintic. However the frequent use of anthelmintics to treat nematodes leads to the development of resistance against all major groups. Bioactive substances extracted from plants represent an alternative for controlling gastrointestinal nematodes. The present study aimed at evaluating the *in vitro* and *in vivo* anthelmintic activity of *Artemisia campestris* essential oil aerial parts against two gastrointestinal parasites.
Methods: The *in vitro* anthelmintic activity tests of *A. campestris* essential oil were performed on *Haemonchus contortus* using egg hatch assay (EHA) at concentrations of(0,0.125, 0.25, 0.5, 1 and 2 mg/mL) and adult worm's motility assay (AWMA) at concentration of (0.125, 0.25 and 0.5 mg/mL) compared with a reference drug albendazole. The *in vivo* nematicidal effect of essential oil was evaluated on *Heligmosomoides polygyrus*. It was monitored through faecal egg count reduction (FECR) and total worm count reduction (TWCR). Three doses (2000, 4000 and 5000 mg/kg) were studied using a bioassay.
Results: In the EHA 100% inhibition was observed at 2 mg/ml after 48 hours incubation (IC50=0.93 mg/ml). In the AWMA, essential oil induced 66.6% inhibition at 0.5 mg/ml after 8 hours post exposure. At the *in vivo* tests the dose of 5000 mg/kg showed a high nematicidal activity (72.1% FECR and 72% TWCR), 7 days post-treatment.
Keywords: Anthelmintic, *Haemonchus contortus, Heligmosomoides polygyrus, Artemisia campestris.*

HBP30: Immunophenotyping of digestive tumor infiltrating lymphocytes using flow cytometry and real time PCR approach

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Background and aim: Digestive cancers are one of the most frequent malignant tumors worldwide. It is a common and lethal disease with a high therapeutic need. Also it is essential to develop new diagnosis methods which could lead to personalized medicine. Biotechnology is playing an essential role in cancer, ranging from monoclonal antibodies to immuno-oncology and in between. Recent investigations have focused on the study of the involvement of tumor infiltrate lymphocytes (TILs) in carcinogenesis as well as some transmembrane proteins such as ionic channels as well as checkpoints.
The aim of this study is double: (i) estimating the rates of TILs using flow cytometry, and (ii) the PDCD1 gene expression level coding for the PD1 checkpoint, in patients with digestive cancers.
Methods: We examined samples from 28 patients to evaluate the immunophenotyping of TILs and PDCD1's expression level. MediMachine mechanical digestion technique, monoclonal antibodies targeting lymphocytes membrane antigens and real-time PCR were used. Optimization of mechanical digestion and immunostaining protocols was practiced.
Results: Our investigations reveal a percentage of leukocytes that was at least twice fold higher in healthy tissues, with an average percentage of 12.03% in non-tumoral tissues and 4.9% in tumor tissues. Moreover, the percentages of $CD3^+$ cells, $CD3^+CD4^+$ cells and $CD3^+CD8^+$ cells are greater in tumor tissues, indicating lymphocyte infiltration. In addition, the $CD4^+$ population is more abundant than that of $CD8^+$ in tumoral tissues. The immuno-phenotyping results demonstrate that PD1 is more expressed on the $CD3^+$ $CD8^+$ subpopulation in all tumor samples, compared to percentages recorded in non-tumoral tissues. This trend is less obvious when considering the $CD3^+CD4^+$ subpopulation. In addition, it has been found that immuno-phenotyped TILs $CD8^+$ are mainly $CCR7^+CD45RA^+$ and $CCR7^-CD45RA^-$ cells. Moreover, we studied the PDCD1's expression level by TaqMan-based real-time PCR for 11 patients with gastric or colorectal cancer. Data reveal that five patients in which PDCD1 is up-regulated. However, it is down-regulated in the other six patients.
Conclusion: The preliminary results obtained are very interesting. Work is in progress to increase the cohort of patients and statistical analysis of data in comparative manner.
Keywords: digestif cancer, TILs, PD1, Immunophenotyping, Real time PCR TaqMan.

HBP31: Zebrafish a valuable model to assess *otos* gene otoprotective function

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Background and aim: Otos has been recently identified as a potential gene therapy target for the treatment of cis-platin-induced ototoxicity. In fact, the analysis of variation in exonic target regions of Otos on South African cisplatin-receiving cancer patients indicated a potentially protective role for the variant G alleles of SNPs rs2291767 and rs77124181 against the development of cisplatin-induced ototoxicity. In order to have a better insight about the function of otos proteins we have performed phylogenetic and tissue expression analysis of otos genes in zebrafish.
Methods: Phylogenetic analysis was performed using MEGA7 software. Expression analysis of otos genes in zebrafish was performed using QPCR and *in situ* hybridization.
Results: Phylogenetic tree of otospiralin protein revealed a clear clustering of two groups. The first group "otos" is represented by otos genes from higher and lower vertebrates species whereas the second group "otos-like" is represented only by genes from lower vertebrate. Basing on our analysis we suggest that the otos-like gene has been lost in higher vertebrate during evolution.
To better understand the relation between the two paralogous genes we have analyzed their expression in zebrafish embryos by *in situ* hybridization and we showed that they are both expressed in inner ear starting from 5 days post fertilization (dpf). In addition, we showed by QPCR that only otos is expressed in adult zebrafish inner ear suggesting a primordial role for "otos" in hearing.
Interestingly, we found that otos-like is specifically expressed in inner cells of the notochord of zebrafish embryos at 1 and 2 dpf. Given the fact that notochord inner cells are characterized by the formation of big vacuole and that mice knocked out for the otospiralin gene showed degeneration of type II and IV fibrocytes, characterized also by the formation of big vacuoles, we suggest that the otospiralin function is related to vacuole formation.
Conclusion: Our results show that zebrafish could be a good model to study the function of otospiralin gene and to assess its otoprotective effect.
Keywords: Hearing, inner ear, otospiralin, notochord, zebrafish.

HBP32: Relationship between hospital surfaces and nosocomial infections: *Acinetobacter baumannii*

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Background and aim: The spread of a nosocomial outbreak often originates from cross-contamination and the most common means of pathogen transference occurs between the hands of healthcare professionals, hospital equipment and patients. However, the aim of this study was to investigate the relationship between *Acinetobacter* strains isolated from inanimate surfaces and those isolated from hospitalized patients.
Methods: The study included imipenem-resistant *Acinetobacter baumannii* isolated in 2014 (January to December) from patients hospitalized for at least 48 hours (urine, blood, pus…) and from environmental samples (surfaces and adjacent equipment of nine wards: emergency, operating room, pediatric, gynecology and neonatal, general surgery, general medicine, infectious diseases, hemodialysis and phthisiology) in Ibn-Zohr and El-Hakim Okbi hospitals in Guelma, Algeria. The isolates were identified by microbiological methods (Api 20NE system "BioMérieux") and confirmed by MALDI-TOF MS. Antibiotic susceptibility of the isolates was performed according to the antibiotic susceptibility standard disc diffusion method on Mueller-Hinton agar. Minimum inhibitory concentrations (MICs) of imipenem were determined using the Etest method. Carbapenemase activity was detected via microbiological tests (MHT, EDTA). Carbapenem-resistance determinant were search for by PCR and sequencing. Clonality of the environmental and clinical strains was performed using the MLST method.
Results: A total of 10 imipenem-resistant *A. baumannii* isolated from clinical and surfaces specimens were screened, All *A. baumannii* strains presented a positive MHT 7 of them having positive EDTA test. Seven strains harboring bla_{NDM-1} and 3 isolates harboring bla_{OXA-23}. MLST of the carbapenem-resistant isolates revealed identical sequence types between the environmental and clinical strains, two STs were determined both in clinical and surfaces samples (ST85, ST115).
Conclusion: Here, we highlight the important role of inanimate surfaces in the spread of carbapenem-resistant A. baumannii and clonal relationship between environmental and clinical strains.
Keywords: *A. baumannii*, hospital surfaces, nosocomial infections, multidrug-resistance.

HBP33: Diagnostic value of miR-21 in the plasma of infertile women with low AMH levels

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Background and aim: Female infertility may cause great personal suffering and distress. only an early detection of diminished ovarian reserve seems to be important and crucial in women healthcare. Anti-Müllerian Hormone (AMH) is known as a relevant marker of ovarian reserve status, its serum/plasma assay is by no means predictive of women's natural fertility. However, the existing methods apart from AMH and Follicular Stimulating Hormone (FSH) detection and ultrasound monitoring of ovaries, are not reliable and seem to be insufficient. Therefore, some new biomarkers like microRNAs (miRNAs) are needed.
Methods: In our study we performed the quantification of miR-21-5p in forty-five plasma samples of women with low AMH levels. Then we compared the obtained results to a set of fifteen healthy women with normal AMH measurements using quantitative real-time PCR. Bioinformatic analyses for microRNAs target genes and molecular pathways prediction was performed using Diana mirpath and miRnet tools giving main biological processes, molecular function according to gene ontology (GO) analysis and KEGG pathways.
Results: Our findings showed that miR-21 was significantly under-expressed, in the plasma samples and has great potential to discriminate between women with low AMH rates versus control ones according to ROC curve analysis.
Conclusion: Further studies are still needed to confirm whether the combination of the miR-21expression with other conventional tests like AMH and FSH measurement would improve the diagnosis accuracy for detecting patients with different forms of female infertility.
Keywords: miR-199a, miR-21, female infertility, AMH, biomarkers.

HBP34: Asn680Ser Polymorphism of the *FSHR* gene is associed with premature ovarian failure and polycystic ovary syndrome in Tunisia

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Background and aim: Follicle stimulating hormone (FSH) is a gonadotrophin hormone that plays key roles in the control of oogenesis and follicle development via a specific receptor (*FSHR*). Several polymorphisms of *FSHR* gene are related to ovulation disorders, such as anovulation in ovarian failure or premature ovarian failure (POF) and polycystic ovary syndrome (PCOS). In the present study, we analyzed an Asn680Ser *FSHR* polymorphism in Tunisian infertile women in comparison with controls in order to establish an eventual association of this polymorphism with ovulation disorders.
Methods: We included 50 fertile women aged < 40 years as controls and 90 infertile women in the same average of age; 55 of them had a primary or secondary amenorrhea and 35 had polykystic ovary. Genomic DNA was extracted from the venous blood of all the women and Asn680Ser polymorphism of *FSHR* was analyzed by PCR–RFLP**.**
Results: Statistical calculations indicates that the prevalence of Ser680Ser (GG), Asn680Ser (AG), and Asn680Asn (AA) genotypes was 10.9%, 34.54%, and 54.54% in controls; 12.72%, 56.36%, and 30.9% in POF women; 14.28%, 71.42%, and 14.28% in PCOS women, respectively. Statistically, we have found a significant association between Asn680Ser polymophism and POF ($p = 0.001$) and PCOS ($p = 0.035$). A significant difference was noted between the frequencies of (AA) genotype in these last patients and controls, suggesting the existence of an association between this genotype and ovulation disorders (IOP and PCOS).
Conclusion: Our findings suggest that the Asn680Ser polymorphism of *FSHR* is associated with ovulation disorders in women with POF and with PCOS.
Keywords: FSH receptor, Single nucleotide polymorphism, premature ovarian failure, polycystic ovary syndrome.

IB Session
Industrial Biotechnology

ORAL COMMUNICATIONS
(IBO)

TC1-IB/IBO1: Novel Strategy to combat *Macrococcus caseolyticus* biofilm colonization in dairy processing lines

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Background and aim: The formation of undesirable microbial biofilms in dairy industries is the main cause of chemical cleaning agent inefficiency. Consequently, biofilm-associated contamination represents one of the major threats to the dairy industry. The aim of this study is to propose novel formulation active against *Macrococcus caseolyticucs* biofilm, a selected model to study microbial biofilm elimination in a dairy processing line.
Methods: Biofilm formed on stainless-steel surfaces was quantified using crystal violet method. Biofilm removal was assessed using matrix quantification and cell count methods, and viability was checked using flow cytometry and fluorescent microscopy analysis.
Results: In this study, 197 bacterial strains were isolated from the center of collect of milk and neighboring farms in Sfax, Tunisia. 6 strains were selected according to their ability to form a thick biofilm on polystyrene microplates. Their capacities to form an important monospecies biofilm on stainless steel surfaces were also confirmed. The biofilm formed by *Macrococcus caseolyticus* was selected to be used as a model for the evaluation of eradication efficacy. For that, different hydrolase and polysaccharidase enzymes were applied separately to treat these biofilms. Moreover, the buffer composition has been also improved to increase the diffusion of molecules inside the formed biofilm and to kill bacteria by the addition of selected active biomolecule. Our results revealed that *Macrococcus* monospecies biofilm removal exceeded 50%. Moreover, when a concentrated cocktail (cock2) was applied, the biofilm removal was more efficient compared to the use of the chemical detergent.
Conclusion: This present study is providing novel bio-cleaner formulations, active against biofilms encountered in dairy industries.
Keywords: *Biofilm, stainless steel, dairy industry, biofilm removal, enzymes cleaners.*

IBO2: Effect of the combination MAP fertilizer / *Bacillus amyloliquefaciens* strain C2 on the physiological state and the protection of tomato against verticillium wilt

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Background and aim: The use of chemical fertilizers and pesticides in agriculture had made impressive gains. However continuous use of these products will likely be followed by further environmental and human health damage. In previous work, we have shown that the purified MAP fertilizer and the strain C2 *Bacillus amyloliquefaciens* can be used individually as ecological alternatives for better growth and the protection of tomato against verticillium wilt. In this work, the effect of combination MAP fertilizer / *Bacillus amyloliquefaciens* strain C2 on the physiological state and the protection of tomato against verticillium wilt was studied.
Methods: A study was conducted in potexperiments where C2 was used in combination with different rates of purified and unpurified MAP.
Results: The combinations purified MAP / C2 improved the physiological state of the plants compared to the individual use of the fertilizer and C2. The combination C2 + 0.25P presented the best one. The combinations C2 + 0.5P and C2 + 1P did not reach the effect generated by C2 + 0.25P, this could be explained by the fact that the plant has already reached its maximum growth thanks to the use of0.5P or 1P or C2 (without combination). The study of the protective effect of the combinations purified MAP / C2 showed that the best protection of tomato against verticillium wilt was obtained by the combination C2 / 0.5P.
Conclusion: The combination C2 + 0.25P generated the best physiological state; whereas, the best protection against *Verticiliumdahliae* was registered with the combination C2 + 0.5P.
Keywords: *Bacillus amyloliquefaciens, Verticilliumdahliae*, oxidative stress, combination, purification, MAP fertilizer.

IBO3: Physicochemical and microbiological characterization of two varieties of dates "Hmira and Feggous", of the commune Abadela wilaya of Bechar and test of alcohol production

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Background and aim: In Algeria, date production occupies a large part of the Saharan agricultural production, of which more than half of their production are common dates. In the municipality Abadela, several varieties of dates, including Hmira and Feggous are more popular, but poorly known at the national level. Our work is interested in the valorization of these two varieties, by the study of their physicochemical qualities, microbiological and an ethanol production test
Methods: Physicochemical analyzes determination of ph by phmetre, ash to muffle 5 h to 550. sugar by method of Dubois, fat by soxcelle. Microbiological analyzes Enumeration of all germs by PCA medium, colifromes by mac conkey, yeast and molds by sabouraux.
Raw material and microorganisms The date by-product used in the present study to generate bioethanol is composed essentially of varieties of dates originated from Algerian Sahara. It w as obtained from Abadela-Béchar. The dates w ere dried, kept in bags and stored at room temperature. They were sold in few quantities at local markets or served as feed for animals. The microorganism *S. cerevisiae* used in the fermentation process of date juice.
Bioethanol generation mediumTw o bioreactors w ere prepared for each date variety studied. The first bioreactor is a glass bottle of 3 L capacity and used to follow the fermentation process.
Results: The morphological and physicochemical characteristics of the fruits studied, show that both varieties are semi-soft dates, of significant weight and size, they contain a humidity of 24% for the Hmira variety, and 21.9% for Feggous, a slightly acidic pH 5.7 and 6.7 for the two varieties respectively, an acidity of 0.08% for Hmira and 1.12% for Feggous, and a rate ofash in the order of 1.7% and 1.6% respectively. Both varieties contain a total sugar content exceeding 70% and a fat content of 0.4% for Hmira and 0.1% for Feggous. Microbiological analyzes show the existence of bacteria such as Staphylococcus aureus, faecal coliforms and also a fungal flora such as; *Aspergillus niger*, *Aspergillus flavus*, *Penicillium* and yeasts such as *Saccharomyces cerivicea.*
Conclusion: Thanks to the biotechnological processes, the results of the production of bioethanol obtained show that the variety "Hmira" Provides a yield higher than that provided by the variety Feggous, it is ofthe order of 25% and 16.7%, respectively.
Key words: Hmira and feggous dates, Valuation, Physicochemical analyzes, Microbiological analyzes, Bioethanol.

IBO4: Abitotic factors affecting the larvicidal activity of the *Bacillus thuringiensis* InhA1 metalloprotease against the Lepidopteran pest *Ephestiakuhniella*

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Background and aim: BUPM28 is a *Bacillus thuringiensis* strain producing the InhA1metalloproteasewith an interesting insecticidal activity against the lepidopteran pest, *Ephestiakuhniella*. The aim of this work is to study the effects of some abiotic factors (temperature, UV light,..) on the larvicidal efficacy ofInhA1 towards *E. kuhniella*.
Methods: The InhA1 gene was cloned into pET-28a vector and the corresponding protein fused to a six histidine-tail was over-expressed in the recombinant *Escherchia coli* BL21 (DE3).The purification of this metalloproteasewas carried out using the Amersham "HisTrap" kit. To assess the toxicity of purified InhA1 protein against *E. kuhniella*, a free ingestion technique was used and the activity of the toxin was determined by surface contamination assays.
Results: InhA1 was efficient in killing first instar larvae of the lepidopteran pest *Ephestiakuhniella*. The $_{LC50}$ obtained after 4 days of contact at 28°C, was 234.75 ng/cm^2. For assessing resistance of this metalloprotease to heat or UV, purified InhA1 proteins were heated at 37, 60 and 100°C for 15, 30 and 60 min or exposed to UV (265nm) for the same duration. The obtained LC50were 341.58, 403.37 and 434.20 ng/cm^2 after treating the protoxin for 60 min at 37, 60 and 100°C, respectively, and 402.96 ng/cm^2 after exposure to UV light for 60 min.
Conclusion: The obtained results indicated that InhA1 retained more than 40 % of its activity after these treatments demonstrating that its larvicidal activity is relatively heat stable and insensitive to UV exposure. These properties could be exploited for the formulation of a novel *B. thuringiensis* insecticide for effective biocontrol of undesirable lepidopteran larvae.
Keywords: Abiotic factors, *Bacillus thuringiensis*, *Ephestiakuhniella*, larvicidal activity, metalloprotease.

IBO5: Thermal stability of natural actomyosin extracted from sardine (*Sardina pilchardus*) muscle affects reactivity to cross-linking by microbial and sardine transglutaminases

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Background and aim: Natural actomyosin (NAM) is a major component responsible for gelation of muscle proteins. Pre-incubation of fish NAM in the presence of salt under the appropriate conditions prior to heating results in formation of a stronger gel. This process is typically known as "setting". One of the important mechanisms involved in the setting phenomenon is the formation of inter- and intramolecular isopeptide bonds ofNAM catalyzed by transglutaminases (TGases). Therefore, the objectives of this research were to compare the thermal stability ofNAM extracted from sardine and to investigate the potential differences in the cross-linking of NAM catalyzed by fish (sardine) TGase (FTG) and microbial (MTG).
Methods : Effects of temperature and transglutaminase on the stability of natural actomyosin (NAM) from sardine (*Sardina pilchardus*) were studied using Ca^{2+} ATPase activity, solubility, sulfhydryl group content, disulfide bonds, surface hydrophobicity and electrophoresis SDS-PAGE.
Results: Natural actomyosin (NAM) from sardine exhibited higher stability to thermal treatment at 40°C/2h and microbial transglutaminase (MTGase) than other samples. Lower Ca^{2+} ATPase activity, solubility and sulfhydryl group content, new disulfide bonds and higher surface hydrophobicity mainly formed over 40°C/2h for samples added with MTGase and fish transglutaminase (FTGase) than over 25°C/4h. However, NAM added with MTGase showed the best results suggesting the higher formation of protein aggregates and three-dimensional network structures of actomyosin. MTGase generally catalyzed more extensive cross-linking of sardine myosin heavy chain (MHC) than FTGase.
Conclusion: NAM conformation and types of TGase were important for MHC cross-linking reactions. Unfolding of NAM played a much more critical role for MTGase than FTGase in catalyzing protein cross-linking. It has been found that gelation can be improved by using MTGase while maintaining the treatment temperature close to 40 ° C. The results suggest that if the maturation process is initiated by the addition of TGase enzyme, subsequent controlled heating greatly improves the texture properties of the fish.
Keywords: Sardine, natural actomyosin, transglutaminase, stability.

IBO6: Expression of a Thermo-stable Xylanase from the Thermophilic anaerobic bacterium *Caldicoprobacter algeriensis* sp. TH7C1^T

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Background and aim: Xylanases have attracted considerable research interest because of their potential industrial applications in animal feed. They catalyze the hydrolysis of xylan which is the major component of hemicellulose and they could be produced by diverse hosts, covering filamentous fungi, yeast and bacteria. The present study aimed to clone, express and purify a xylanase from *Caldicoprobacteralgeriensis* sp. nov., strain TH7C1^T, a thermophilic, anaerobic strain isolated from the hydrothermal hot spring of Guelma (Algeria).
Methods: The genomic DNA of *Caldicoprobacteralgeriensis.*sp TH7C1^T was extracted and partially sequenced. The result of the genome annotation shows the presence of a gene coding a xylanase called xynBCA. Amplification of xylanasegene was performed using two primers xylD and xylR. This gene was cloned into the pET-2 1a(+) vector and expressed in *E. coli* BL21 (DE3) pLysS. The purification and characterization of the recombinant activity is also undertaken.
Results: The PCR product was firstly cloned into pUT57 vector and transformed into *E.coli* DH5αthen subsequentlytransferred into thepET-21a(+) vector and introduced in the*E. coli* BL21(DE3)pLysS. The cloned xylanase gene encoded a polypeptide of 451 amino acids residues with a calculated molecular weight of 47,9939 1 kDa. Amino acid sequence homology analysis showed that this enzyme belongs to xynB (Family10). The family 10 contains endo-β-1,4-xylanases with higher molecular weights than those of the xylanase family 11, and have (α / β)8 barrel in their three-dimensional (3D) structures.The XynBCAis optimally active at 80°C and pH7 and with a half-life at 75 °C of 1 hour 10 min.The protein showed no detectable activity in presence of $Zn^{2+}$and $Mn^{2+}$and was strongly inhibited in presence of 2 mM Co2 + and $Cu^{2+}$and highly activated in presence of Li^{2+}, Na^{2+}, Ca^{2+}, Mg^{2+}, Fe^{3+} and EDTA. The nearly purification of the recombinant enzyme at homogeneity was achievedby heat treatment, acetone precipitation and FPLC. Total purification is in progress.
Conclusion: Due to its high level of thermostability, this enzyme could be a good candidate for animal feedbyitsanti-nutritional effects of certain constituents of cereals.
Keywords: Xylanase, thermostability, animal feed, *Caldicoprobacteralgeriensis.*

IBO7: Purification, Characterization and Application in the Bread Making Industry of Acid-stable Alpha Amylases from a New Isolated *Bacillus subtilis* US586 Strain

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Background and aim: Amylases cover about 25 to 33% of the world enzyme market. They are used in various industries, mainly in the starch hydrolysis as an essential step to generate glucose, maltose, and a mixture of malto-oligosaccharides. The search for novel amylases adapted to use in bread baking have been the major extent of recent research. The aim of this work is the purification and the characterization of amylase produced by a new isolated *Bacillus subtilis*US586 strain. The effect of the addition ofAmyUS586 as bread improver was also investigated.
Methods: The purification of Amy586 was achieved through acetone precipitation followed by Superdex200 Gel Filtration column. The effect of AmyUS586 supplementation on rheological dough properties and bread quality was investigated by the measurement of rheological and texture parameters.
Results: A new alpha amylase producing strain was identified as *Bacillus subtilis* US586. The analysis of the crude enzyme proved the presence of three amylases 1, 2, and 3, called AmyUS586, expected to be three isoforms. The purified amylases had molecular masses of 48 kDa, 52 kDa, and 68 kDa with a total specific activity value of 2133 U/mg. AmyUS586 produced maltose, maltotriose, and maltopentaose as main end products from starch hydrolysis. It exhibited a large 4-6 optimal pH and a 60°C temperature activity. Amylases exhibited a moderate thermostability with a half-life time of 45 min at 55°C. The addition of AmyUS586 to a weak local wheat flour, having a dough baking strength (W) of 138 10−4 J, decreased its P/L ratio of elasticity and extensibility from 1.9 to 1.2 and increased its W to 172× 10−4 J. Moreover, additions of 0.06 U/g flour AmyUS586 also improved the bread texturo-parameters by a decrease in its firmness values and a slight increase in the cohesion and elasticity.
Conclusion: In this study we reported the purification of amylases Amy586 with suitable characteristics for bread making. In fact amy586 addition to weak flour can be applied as a corrective action to reduce problems in dough and improve bread texture.
Keywords: Alpha amylase, *Bacillus subtilis*US586, Acid-stable, Bread Making.

IBO8: A novel serine alkaline protease from *Anoxybacilus kamchatkensis* M1V and its application as detergent additive

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Background and aim: Thermophiles have often been proposed as sources of industrially relevant thermostable enzymes. Thanks to their biochemical properties, they are relevant for specific industrial applications that determine the demand for tailor-made enzymes and shift the industrial interest towards biocatalysts from extremophiles including proteases.
Methods: The biochemical and molecular characterization of the pure enzyme were investigated though physico-chemical and kinetic determination as well as spectroscopy analysis.
Results: A new extracellular thermostable serine alkaline protease (designated SAPA) was produced, purified, and characterized from *Anoxybacillus kamchatkensis* M1V. The bacterial strain was found to hyper-produce extracellular protease when grown at 45 °C in optimized media (4,600 U/mL). The purification to homogeneity of the SAPA enzyme was achieved, simultaneously, by precipitation with ammonium sulfate fractionation-dialysis, anion exchange (FPLC), and gel filtration (HPLC) chromatographies. It had a relative molecular mass of 28 kDa as estimated by SDS-PAGE. The sequence of its NH2-terminal amino-acid residues showed high homology with those of *Bacillus* proteases. It showed optimal activity at pH 11 and 70 °C. The thermoactivity and thermostability of SAPA were enhanced in the presence of 2 mM Ca^{2+}. Irreversible inhibition of the enzyme activity by DFP and PMSF confirmed its belonging to the serine proteases family. Interestingly, SAPA displayed higher levels of hydrolysis, substrate specificity, and catalytic efficiency than proteases *viz*. SPVP from *Aeribacillus pallidus* strain VP3, SAPB from *Bacillus pumilus* strain CBS, Subtilisin A from *B. licheniformis*, and Subtilisin 309 from *B. clausii*. More interestingly, SAPA showed a high detergent stability and compatibility and an outstanding stain removal compared to commercial enzymes *viz*. AlcalaseTM and SavinaseTM. The *sapA* gene was cloned, sequenced, and expressed in the extracellular fraction of *E. coli* BL21(DE3)pLysS.
Conclusion: Above all, SAPA exhibited remarkable biochemical proprieties which may be considered as a potential candidate for biotechnological applications such as a cleaning bioadditive in laundry detergent formulations.
Keywords: Protease, *Anoxybacillus kamchatkensis*, Thermophilic, Detergent additive.

POSTER PRESENTATIONS
(IBP)

IBP1: Microbiological quality of Arbia goat's milk and cheese: coagulation by Cy-naracardunculus

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Background and aim: A microbiological study of the milk of local goats and their cheeses obtained after coagulation with an enzymatic extract Cynara cardunculus, is the objective of our work.
Method: The evaluation of the microbiological quality of the milk obtained from goat cheeses was based on the search and enumeration of: total aerobic mesophilic flora (FAMT), total and faecal coliforms, the *streptococcus fécaux*, the staphylocoques dorés (S. aureus), Salmonella, clostridiums sulfioréducteurs. For renneting of the milk with the enzymatic extract of Cynara cardunculuss, 200 ml of each test were heated to 42 O C, and 2 drops of the enzymatic extract placed on a heating magnetic stirrer were added until coagulation.
Results: The results of the microbiological analyzes carried out on milk and goat cheese are shown in Table 1. The results obtained show that the milks and cheeses had satisfactory microbiological qualities, namely the respective FAMT loads of 2.10 ± 0.35 × 10 4 germs / ml and 2.31 ± 0.22 × 10 4 germs / ml, and a total absence. total and faecal coliforms, salmonella and clostridia.
Conclusion: The present microbiological study of goat milks and cheeses prepared with cardoon (Cynara cardunculus) under UV shows the possibility of using this coagulant extract under these conditions without major risk to the health of consumers.
Keywords: Algerian goat, Cynara cardunculus, milk, cheese, hygienic quality, sanitary quality.

IBP2: *In vitro* starch digestibility in sorghum starch by-products from Algerian cultivars

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Background and aim: Grain sorghum (Sorghum bicolor (L.) Moench), one of the minor cereals cultivated in the Algeria (Tidikelt and Hoggar region). The aim of the present study was to assess the nutritive value of by-products obtained after sorghum starch extraction, as dietary starch from feed grains. We use small-scale laboratory steeping in NaOH, and wet-milling process of starch extraction from sorghum whole grain. The fraction yield, starch yield and starch recovery in by-products and starch isolate were determined. Then, the kinetic ofthe starch digestion in sorghum meal and by-products was studied.
Methods: The *in vitro* starch digestion was determined according to the modified method of Goni et al. (1997) by α-amylase (type VI.B from *porcine pancreas*) and amyloglucosidase from *Aspergillus niger* enzymes. Oxidase-Peroxidase Kit (Biomaghreb, Tunisia).
Results: Five by-products differing in their particle sizes and starch contents were collected. The mean values of fraction yields and starch contents, for first, second and third fraction of sorghum gluten feed were respectively 16.13%-52.63%, 01.97%-44.08% and 04.99%- 56.75%, while for sorghum gluten meal the values were 22.27% -71.13%. The substrates from whole grain meal, prepared by dry milling, and from by-products differed in their in vitro starch digestion. The mean values for kinetic parameters ranged from 0.0066 to 0.0147 min^{-1} for the rate constant (k), from 53.66 to 98.58% for the starch hydrolysis at infinite time (C_∞) and from 6.06×10^3 to 8.47×10^3 %.min for the area under the hydrolysis curve (AUC).
Conclusion: Generally, a high digestibility ofby-products of sorghum starch isolation with a great potential for sorghum in livestock and animal feeds are considered in this work.
Keywords: Sorghum, Starch extraction, By-products, Starch digestion, Animal feed.

IBP3: Effect of brine concentration on physico-chemical characteristics, texture, rheological properties and proteolysis level of cheeses produced by *Cynara cardunculus L.* rennet

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Background and aim: Different types of plant coagulants are employed in milk gelation and cheese-making process, such as aqueous extracts from wild cardoon flowers, which have been used for ages in the production of traditional goat's and ewe's cheeses. Their milkclotting activity (MCA) was attributed to aspartic proteases, named cardosins A and B, which resemble to chymosin and pepsin; respectively, in terms of specificity and activity. The aim of this study was to evaluate firstly the quality characteristics of curds produced by a Tunisian wild cardoon rennet, in order to test its efficiency as an appropriate substitute of chymosin, in cheese making process. The second objective was to compare the properties of *Cynara cardunculus* semi-hard cheeses, brined at different salt concentrations (5 %, 7 %, 10 %and15%).
Methods: In this context, physico-chemical, textural, rheological properties and RP-HPLC peptide profiles were investigated.
Results: Results showed that wild cardoon rennet and chymosin produced curds with similar yield, texture, viscoelasticity (G', G") and colour, suggesting that this extract could replace successfully calf rennet. After brining, it was concluded that the use of 15 % of salt in brine was an efficient way to reduce considerably the proteolysis level and bitter peptides concentration in *C. cardunculus* cheeses, stored for 28 days at 4 °C. At this salt level, the highest hardness, gumminess, viscoelasticity and yield of cheeses were also recorded.
Conclusion: Consequently, this study allowed us to select the best salt concentration for *C. cardunculus* cheese brining (15 %), leading to excellent results in terms of physico-chemical properties, yield, texture and viscoelasticity. The satisfactory findings, according to yield, texture, viscoelasticity and proteolysis level, could open new opportunities to produce industrially the *C. cardunculus* rennet and its cheeses in Tunisia.
Keywords: *Cynara cardunculus* rennet, Cheese, Salt, Brine, Rheology, Texture.

IBP4: Kinetic study of Enzymatic Hydrolysis of Starches Isolated from Sorghum and Millet Grains Cultivated in the Desert of Algeria

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Background and Aim: Grain sorghum (Sorghum bicolor (L.) Moench) and pearl millet (Pennisetum glaucum (L.) R. Br.) are staple foods in the desert of Algeria (Tidikelt and Hoggar region).This work aims at evaluating of starch obtained after sorghum and millet isolation and enzymatic hydrolysis has been studied for its potential industrial biotechnology applications.
Methods: Starch substrates are isolated from six sample grains; starch was isolated using wetmilling process in different steeping conditions. The substrates were prepared with different concentrations ranging from 1.25 to 20 g/l. The kinetic hydrolysis of various starches by amyloglucosidase (EC 3.2.1.3) has been studied.
Results: The kinetics of hydrolysis of the extracted starches obey the Michaelis – Menten model. The values of the kinetic parameters K_m (g L^{-1}) and the maximum velocity $_{Vmax}$ (g L-1 min^{-1}) calculated from the curves of Lineweaver–Burk plots. The michaelis constant K_m ranged between 18.84 to 97.9 for sorghum and 33.28 to 46.16 g L^{-1} for millet, maximum velocity $_{Vmax}$ ranged from 1.72 to 6.82 for sorghum and 3.73 to 4.45 for pearl millet g L-1 min^{-1} g L^{-1} , the catalytic constant $_{Kcat}$ (min^{-1}) ranged from 0.59 to 2.4 and from 1.28 to 1.5 min^{-1} for pearl millet.
Conclusion: The results show that the kinetic parameters of the hydrolysis of starch by amyloglucosidase are dependent on the conditions of starch extraction and source of starch and the conditions of starch isolation from grains. This study confirmed that sorghum and millet with a high susceptibility towards glucoamylase hydrolysis to glucose for industrial applications.
Keywords: sorghum, pearl millet, starch, amyloglucosidase, hydrolysis, kinetic.

IBP5: Swelling power and Water Solubility Index of Algerian Sorghum Starch Isolated by Alkaline Wet Milling Procedure

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Background and aim: Sorghum (*Sorghum bicolor* (L.) Moench) is an important cereal that can be grown in the semi-arid regions, it is considered one of the minor cereals cultivated in the Tidikelt region of Algerian Sahara. A little research has been done on improving sorghum grains for nutritional quality or on their uses as industrial raw material. The aim of this work was to determine the swelling power and water solubility index of Algerian sorghum starch isolated by steeping in NaOH solution, and wet milling procedure.
Methods: Starch was isolated from five sorghum landraces grains using alkaline procedures involving steeping, wet milling, filtration, centrifugation, separation, and drying. swelling power and water solubility index of starch samples were determined at 55 °C, 65 °C, 75 °C, 85 °C and 95 °C according to the method described by Li and Yeh (2001).
Results: The recovery starch isolation of both cultivars ranged between 58.06% and 70.60%. Total starch and protein content of starch isolate ranged from 95.35% to 98.75%, and 0.35% to 0.77%, respectively. The swelling power and water solubility index of starch samples at different temperatures showed a non-linear increase with temperature. The results show that the SP and WSI of starch samples ranged from 3.41 (g/g) to 38.83(g/g), and 2.15 % and 38.6 %, respectively.
Conclusion: The swelling power increased with increasing temperatures. When the temperature above 75 °C, the swelling powers of starch samples were higher than those of amylopectin. The water solubility index has illustrated the similar trend, increasing with increasing temperature. When the temperature above 65 °C, the water solubility index was very lower than those of amylopectin.
Keywords: sorghum; starch; isolation; wet-milling; swelling power; water solubility index.

IBP6: Serine protease from *Bacillus licheniformis* strain K7A: Optimization of production parameters, purification, and biochemical characterization

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Background and aim: Proteases are hydrolytic enzymes capable to cleave the peptides linkages in protein, generating soluble peptides and free amino-acids. They are the paramount hydrolytic enzymes widely used in industrial sector accounting ~65% of the aggregate worldwide enzyme market. Proteases are extensively exploited commercially, in detergent industry, food, pharmaceutical, and leather. Given their potential use, there has been renewed interest the discovery of novel proteases with novel properties. In this study an attempt was made for, the optimization, purification and biochemical characterization of an extracellular alkaline protease named SAPHM secreted from the culture supernatant of the thermophilic bacterium strain K7A, isolated from the Hassi Messaoud region, of the Algerian desert to find its suitability detergent bio-additive.
Methods: Optimization of protease production conditions *viz*. time, pH, temperature, different carbon and nitrogen sources and their concentrations were studied. The biochemical characterizations carried on include physico-chemical determination and spectroscopic analysis.
Results: The results showed that the optimum conditions for maximum protease production by *Bacillus licheniformis* strain K7A isolated from the Hassi Messaoud region, Southeast of Ouargla in Algeria. K7A exhibited a high extracellular protease activity (about 12,500 U/mL) after 24 h incubation in an optimized medium at 45 °C. Protease was purified by heat treatment (30 min at 70 °C) and ammonium sulfate precipitation (40-70%)-dialysis followed by Q-12 FPLC and ZORBAX PSM 300 HPLC column. The MALDI-TOF/MS analysis revealed that the purified enzyme was a monomer with a molecular mass of 30,325.12 Da. The sequence of the 26 NH2-terminal residues of SAPHM showed high homology with those of Bacillus proteases. Optimal activity was achieved at pH 10 and 70 °C. SAPHM was completely inhibited by PMSF and DFP suggested its belonging to the serine alkaline family. It exhibited excellent stability to detergents and wash performance analysis revealed that it could remove blood-stains effectively. Data suggest also that SAPHM may be considered as potential candidate for future applications in non-aqueous peptide biocatalysis because it possesses an elevated organic solvent resistance.
Conclusion: These properties make SAPHM a potential, promising and eco-friendly alternative to the conventional chemicals used for laundry detergents.
Keywords: *Bacillus licheniformis*, Optimization, Alkaline protease, Characterization.

IBP7: Pullulanase immobilization in functionalized alginate hydrogels

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Background and aim: The control of enzyme activity appears of primary importance due to increasing number of application (biomedical, food, and so on). Enzyme may undergo a variety of reactions during production, storage and/or application that can lead to the loss of their activity. Therefore the use of free enzymes in industrial application has been limited, so physical or chemical Immobilization of such enzyme in entangled network of water soluble polymer or in suitable hydrogel can be of great interest in order to control and understand the enzymatic activity. Interpenetrated network of polymer (mainly polysaccharides or proteins) constitute most of real environment for enzyme.
Methods: Enzyme activity was estimated by the determination of number–average molar masses using two different methods: a colorimetric assay of reducing ends (REs) and a size exclusion chromatography/multiangle light scattering/differential refractive index. The second method also provided weight–average molar masses of hydrolyzed pullulan and the quantity of maltotriose (DP3) and its multiples (DP6 and DP9) produced by the enzymatic treatment.
Results: In this work, we have investigated the behavior ofPullulanase enzyme, immobilized in hydrogels (calcium Alginate-grafted-Jeffamine® beads and calcium alginate beads), towards its substrate (pullulan). The mass ratio of oligosaccharides (oligo%) and slightly degraded pullulan (PnH%) in the hydrolysis medium obtained from SEC/MALS/ DRI data as a function of time was verified. Complete diffusion of pullulan into the Alg-Ca beads was achieved within 7 hours, whereas it takes longer time with the Alg-g-Jeff®-Ca system. The kinetic downturn in the case of thermosensitive beads may be due to the reason that Pullulanase will be in friendlier environment where hydrophobic microenvironment comes from Jeff®-Jeff® interactions. The comparison of the mechanism mode of entrapped enzyme on Alg-g-Jeff®-Ca and Alg-Ca systems was achieved. The presence of dimer and trimer in the reaction medium confirms that the enzymatic process of immobilized pullulanase in Algg-Jeff®-Ca beads is still an “endo” process.
Conclusion: The presence of Jeffamine®M2005 does not therefore change the enzymatic process of pullulanase or its activity at the optimum conditions (60°C and pH 5.5).
Keywords: Pullulanase, pullulan, alginate-grafted-Jeffamine®, immobilization, hydrolysis.

IBP8: Physico-chemical and microbiological qualities of milk from cows raised extensively in North-East Algeria

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Background and aim: Our previous work has made it possible to develop a typology of farms in the far north-east of Algeria. The majority of breeders seem to opt for the breeding of local cows "Brune de l'Atlas" in extensive system. The milk is consumed by the families and still transformed in a traditional way at the farm level into fermented milk (d’hen, lben, raïb) and then sent directly to the markets. This informal circuit is estimated at 1.6 billion liters in Algeria, or more than 69% ofraw milk bovine production.
In this context, the physicochemical and hygienic qualities of raw milks from local cows in the far northeastern region of Algeria were studied.
Methods: hirty samples of raw milk of small mixture (ten females) of local cows of six periurban farms of extensive breeding situated in the Algerian Northeast were analyzed for their physico-chemical and microbiological parameters 5 times every two or three days over a period of two weeks
Results: At the exit of the udder, milk was characterized by a temperature of 36,6°C and a pH of 6,53. On average the content of the milk in fat and in protein material (subject) was respectively equal to 33,4 and 32,8 g/l. After collection, the milk presented a variable load of the FMAT from 2.10 3 to 49.105 ufc / ml, 4,7.104 ufc / ml of total coliforms and 1,1.104 ufc / ml of fecal coliforms, with a total absence of Salmonellas, Clostridiums and antibiotics.The content in coliforms (total, fecal) was superior to those mentioned by the Algerian gazette**.**
Conclusion: The raw consummate cow's milk may present some sanitary risk for the population of the zone of study; its hygienic quality is bad. So it should be boiled or pasteurized.
Keywords: Brune de l’Atlas, El-Tarf, extensive breeding, fat, hygienic quality, local cow, protein.

IBP9: Effect of iron oxide nanoparticles on cytosolic enzymes in the brain and cerebellum of rabbits *oryctolagus cuniculus*

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Background and aim: The nanoparticles (NPs) measure between 1 and 100 nm. Their nanometric size gives them new properties which are particularly interesting for industrialists and scientists. If man is exposed to an increasing number of sources, the impact on health and, in particular, on the nervous system remains poorly assessed. The aim of our research is to evaluate the effect of Fe_3O_4 nanoparticles on some parameters of oxidative stress in two central nervous system regions.
Methods: For this study, Treatment of rabbits for three months with 75µg/ kg/day and 175 µg/kg/day of iron oxide nanoparticles, and after three months the enzymatic activity of GPx was measured by the method ofFlohe and Günzler (1984). The spectrophotometric assay of catalase (CAT) activity was performed according to the method of Cakmak and Horst (1991). The activity of glutathione S-transferase (GST) was determined according to the method of Habig and *al*. (1974) , finally, Glutathione (GSH) level was determined according to the method of Weckbeker and Cory (1988).
Results: The Administration of 75µg /kg/day of iron oxide nanoparticles results a nonsignificant increase in activity Enzymatic of glutathione S-transferase in both brain regions. In contrast, the dose 175 µg/ kg/day results a highly significant increase in the brain region and a very highly significant increase in the cerebellar region. Results show a non-significant ($p \geq 0.05$) increase in cortex catalase activity (with both doses) and cerebellum (with low dose), whereas the high dose leads to a significant increase ($p \leq 0.05$). results a non-significant decrease in enzymatic activity of GPx. Whereas, a very highly significant decrease ($p \leq 0.001$) in GSH levels after the treatment of rabbits by iron oxide nanoparticles was clearly conceivable in the two brain regions.
Conclusion: this work showed a toxic effect of these nanoparticles on enzymes of brain and cerebellum, induced many effects conducting to the death of cells (neurons) and cause a neurodegenerative disease.
Keywords: iron nanoparticles, brain, cerebellum, neurotoxicity, cytosolic enzymes.

IBP10: Enhanced synthesis of isoamyl acetate using liquid-gas biphasic system by the transesterification reaction of isoamyl alcohol obtained from fusel oil

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Background and aim: Isoamyl acetate, represents one of the most important flavor compounds used in food industries with an annual demand of about 74,000 Kg. In this work, the transesterification reaction of isoamyl alcohol obtained from fusel oil and leading to the synthesis of isoamyl acetate was conducted simultaneously with in situ ethanol removal, which allows to shift the reaction equilibrium toward ester synthesis.
Methods: Immobilized lipase from *A. oryzae* catalyzing the transesterification reaction of isoamyl alcohol and ethyl acetate to produce isoamyl acetate was applied in fluidized bed reactor.
Results: The effects of the most influent parameters affecting the reaction have been also investigated using a Doehlert matrix design. The better operating conditions for isoamyl acetate synthesis were: a temperature of 68.5°C and a respective isoamyl alcohol and *A. oryzae* lipase concentration of 0.72 M and 2.39 g/L. At these conditions, the resulting reaction conversion and ethanol extraction yields were of 89.55 and 69.60%, respectively. The use of the fluidized bed reactor with continuous ethanol removal has allowed to improve the reaction conversion which was two times than the conversion higher obtained in batch reactor. Furthermore, under the optimized conditions in the fluidized bed reactor, the reaction conversion and the ethanol extraction yields were increased by 44.8 and 3 6.2%, respectively.
Conclusion: A liquid-gas bi-phasic system has been developed by using fluidized bed in a solvent free-system with continuous ethanol extraction. The nitrogen was shown to be efficient for ethanol extraction in this system.
Keywords: *Aspergillus oryzae* lipase, ethanol removal, kinetic model, transesterification.

IBP11: Production and valorization of a purified biopolymer from *Leuconostoc* sp. With rheological potential

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Background and aim: Exopolysaccharides (EPS) produced by lactic acid bacteria (LAB) have gained increasing attention over the last few years because of their contribution to the rheology and texture of food products, increasing the possibility to replace or reduce the use of external hydrocolloids, it's can be used as viscosifying, stabilizing, emulsifying, sweetening, gelling and water-binding agent in the food.
Methods: For this purpose, the use of a ruthenium red medium revealed the production of exopolysaccharides from different animal and vegetable samples by LAB strains. 1 6S rDNA gene sequencing was used to identify species level of these strains. In addition, for the EPS production and preparation, a selective sucrose medium has been used. Total sugar estimation and appearent viscosity were determined. Moreover, Componential analysis of hydrolyzed EPS by thin layer chromatography and the structural characterization of these polymers by FTIR, and NMR (H^1 and C^{13}) analysis were investigated.
Results: A total of 3 strains were identified as *Leuconostoc citreum* and *Leuconostoc me*senteroides, which show a highly viscous growth on selective sucrose medium. The results showed that the production of EPS varied from 666,1 mg /l to 1254 mg /l for the highest production for *Leuconostoc mesenteroides* LGM-L14 strain and the apparent viscosity ranged from 201 to 1393 centipoise (cP).Componential analysis of hydrolyzed EPS by thin layer chromatography indicated that it is a dextran, consisting of glucose monomer. Furthermore, the structural characterization of these polymers by FTIR, and NMR (H^1 and C^{13}) analysis confirmed that it is α (1, 6) dextran with a low percentage of α branching (1, 3).
Conclusion: The isolated strains of *Leuconostoc*sp. are a potent producer of dextran, which find its applications in various industries; it can be used as natural, safe additives to enhance the rheology and texture ofnovel food products.
Keywords: Exopolysaccharides, *Leuconostoc* sp., Dextran, viscosity.

IBP12: Action of probiotics on *Salmonella* spp. inoculated in minced meat packaged under vacuum and modified atmosphere

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Background and aim: Considering the prevalence of *Salmonella* in minced meat and the frequency of its consumption via many traditional products and, taking into account that packaging of meat is the most common method of food preservation, there is a need to explore the effect of packaging methods on *Salmonella* spp. survival, especially in mixed minced meat inoculated with probiotic bacteria. Therefore, the aim of this study was to compare the effects of vacuum and two initial headspace-modified atmosphere conditions (20% O_2/50% CO_2) and 20% O_2/30% CO_2) on the survival of *Salmonella* spp., total viable bacteria and probiotic lactic acid bacteria in minced meat stored at 4 °C.
Methods: Minced meat was analyzed for *Salmonella* spp., total viable count and probiotic *Lactobacillus fermentum* strain count immediately on days 3, 7 and 9 of storage. The meat pH was measured. Measurement of headspace gas composition in the minced meat packaging was also conducted.
Results: *Salmonella* spp. counts decreased during storage in all packaging types, with reductions of about 2 log CFU/g. A significant difference ($P < 0.05$) was noted between *Salmonella* spp.counts in meat packaged in vacuum and modified atmospheres, although there was no significant difference in *Salmonella* spp. count between meat packaged in 50% CO2, and meat packaged in 30% CO2. At the end of the study, there were significant differences ($P < 0.05$) in total viable and probiotic bacteria counts between meat packaged in vacuum and modified atmosphere, and the lowest counts were noted in meat packaged in modified atmosphere with 50% CO_2. In all meat sampled, the pH was 5.7 at the beginning of the study and then increased during storage.
Keywords: Minced meat; Vacuum, Modified atmosphere packaging; Probiotic; *Salmonella* spp.

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IBP13: Biochemical and molecular characterization of a recombinant α-amylase from *Bacillus subtilis*

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Background and aim: Alpha amylase (EC3.2.1.1), one of more widespread enzymes in the industrial world, is a glycoside hydrolase. It hydrolyzes the α- (1,4) glucoside linkage between the glucose units of a polysaccharide. Microbial amylases, particularly those from *Bacillus* genus are more in demand than those from other sources. Alpha-amylases have potential application in wide number of industrial applications such as textile, paper, detergent, food, fermentation and pharmaceutical industries. Recombinant DNA technology for amylase production involves the selection of an efficient amylase gene, its insertion into an appropriate vector system, transformation in an efficient bacterial system to produce high amount of recombinant protein. In this context, the aim of this work is the overexpression of anαamylase gene from *Bacillus subtilis*US572in *E. coli* strain and the characterization of the recombinant amylase which is an interesting candidate for biotechnological applications.
Methods: PCR "Polymerase Chain Reaction", extraction of Plasmid DNA, amylolytic activity measured by the DNS method. Ni-NTA column affinity TM column affinity, Westrern Blot
Abstract: An α-amylase gene, called AmyKS, was cloned from *Bacillus subtilis* strain US572 and expressed in *Escherichia coli* BL21 cells. Two variants ofthe gene were investigated: the first one with the signal peptide (SPS+) and the second without it (SPS-). The analysis of recombinant bacteria expressing of these gene forms, showed that the gene without SPS gives higher level of amylase than the SPS+ gene. The His-tagged recombinant AmyKSSPS named r-42-AmyKS was purified in one step using Ni-NTA column affinity with a specific activity of 664.28 U mg^{-1}. The comparison between r42-AmyKS and the wild type AmyKSSPS+ (Wt-AmyKS), showed that there is variation in certain biochemical characteristics. Indeed, the optimum temperature of the wild type and recombinant enzyme has changed from 60 °C to 70 °C respectively. In addition, r42-AmyKS is more resistant to certain additives than the Wt-AmyKS. The hypothesis that C-terminal His-tag could be responsible of this variation was confirmed by experiment tests and molecular modeling.
Conclusion: According to our results, r-42-AmyKS has the suitable characteristics to be applied in bread-making or other industrial applications.
Keywords: Alpha-amylase, *Bacillus subtilis*, *E.coli* BL21 cloning and expression, purification, biochemical characterization, molecular modeling.

IBP14: Expression of a copper activated xylanase for bioethanol production in yeast: Differential properties of c-tagged or untagged recombinant xylanases

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Background and aim: Endo-xylanase (EC 3.2.1.8), which hydrolyze the backbone structure of β-1,4-xylans, can be used together with laccase (activated by copper) to hydrolyse lignocellulosic and hemicellulosic materials in bioethanol production. The previous study conducted in our laboratory described the cloning of the xylanase gene from *Aspergillus niger* US368 and obtaining the recombinant His-tagged protein using *E. coli* expression system. This recombinant xylanase (His-tagged r-XAn11) was activated by copper which is a strong inhibitor for xylanases activities. The activation of the enzyme toward copper is due to the 36 extra amino acids provided by the vector pET28a. However, the expression of this xylanase was relatively low and intracellular that limits its potential industrial application. Hence, we aimed to over-express this interesting enzyme using the *Pichia pastoris* system for use in biofuel production.
Methods: The recombinant xylanase (His-tagged r-XAn11) produced by *E. coli* BL21 which was activated by copper, was expressed in *Pichia pastoris* using the pGAPZαB expression vector.
Results: The recombinant xylanase (His-tagged r-XAn11) produced by *E. coli* BL21, which was activated by copper, was expressed in *Pichia pastoris* using the pGAPZαB expression vector. Two recombinant xylanases forms were obtained (non-C-terminal tagged and C-terminal tagged His-tagged r-XAn11). The findings revealed that the two recombinant xylanases displayed different behaviors toward the copper. In the presence of 3 mM Cu^{2+}, the relative activity of the non C-terminal tagged- His-tagged r-XAn11 was enhanced by about 52%. However, the xylanase activity of the C-terminal tagged was strongly inhibited by copper. The non C-terminal tagged recombinant enzyme was more thermostable, in the presence of 3 mM Cu^{2+}, than the C-terminal tagged one with a half-live of 10 min at 60 °C. 3D models of the two recombinant forms were constructed. The results showed that the created copper site in the non-C terminal tagged protein was loosed in the C terminal tagged protein due to new interactions established between the aminoacids of the C and N-terminal tag.
Conclusion: The present work reports the expression and purification of two recombinant xylanases forms (non C-terminal tagged and C-terminal tagged His-tagged r-XAn11).
Keywords: Xylanase; Expression; *Pichia pastoris*; Copper; Biofuel Production.

IBP15: Effect of acid whey on the quality of gluten-free pasta for celiac patients

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Background and aim: The replacement of gluten functionality in gluten-free products represents a major technological challenge. Gluten-free products are seeing a growing demand, due to the increase in the incidence of pathologies of various type, all linked to some form of intolerance to gluten. The production of hight-quality gluten-free products can be seen as a very important issue and can contribute with the development of puplic policies for individuals with special feed requirements. The objective of this study is to valorize whey by incorporating it into the production of "spaghetti type" gluten-free pasta made from rice and corn, then to see its effect on the technological feasibility and culinary properties of the finished product.
Methods: A physico-chemical characterization of raw materials is realized. The culinary quality of the pasta manufactured was appreciated by the determination of the Optimum Cooking Time, Water Absorption, and Cooking Losses according to the incorporation rates (25%, 50%, 75%, and 100%) in acid whey.
Results: The results obtained indicate that the pasta enriched with acidic whey is characterized by a significantly lower Cooking Time ($p < 0.05$) than that of the Commercial Wheat and Commercial Gluten Free Control. Water absorption is inversely proportional to the incorporation rates of acidic whey. However, pasta, enriched with acidic whey, absorbs less water than the hard wheat and commercial controls without gluten. Cooking losses are affected by the incorporation of acid whey, the best cooking losses are observed for incorporation rates between 25% and 50%.
Conclusion: Acidic whey has an effect on gluten-free pasta quality.PCA results show that cooking losses are not significantly correlated with protein and ash levels.
Keywords: Gluten free pasta, acid whey, culinary quality.

IBP16: Evaluation of *Photorhabdus temperata* antimicrobial activities

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Background and aim: *Photorhabdus temperata* is a bioluminescent Gram negative bacterium living in symbiosis with an entomopathogenic nematode. A critical challenge of this symbiosis is the maintenance of monoxenic infection of the insect by the production of small molecules involved in the inhibition of other microorganisms that could potentially invade the insect carcass.The objective of this work is to investigate the *Photorhabdus temperata* metabolites having potential for pharmaceutical and agricultural bioprospecting.
Methods: The antifungal and antibacterial activity of *Photorhabdus* extract was investigated in order to evaluate its efficacity against a panel of standard reference strains, using disc diffusion, minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) methods.
Results: Among various organic solvents with increasing polarities, used in liquid-liquid extraction of *P. temperata* secondary metabolites, ethyl acetate is the most effective one. This extract has the strongest antibacterial activity against *Staphylococcus aureus*, *Bacillus subtilis*, *Bacillus cereus*, *Agrobactérium tumefaciens (1)* and *Agrobactérium tumefaciens (2)* with an inhibition diameter of 28, 24, 15, 15 and 1 2mm respectively. In contrast, the butanol extract shows antibacterial activity only against *Agrobactérium tumefaciens (1)*, *Agrobactérium tumefaciens (2)* and *Bacillus cereus*, with an inhibition diameter of 18, 12 and 11 mm respectively. The MIC and MBC values showed that ethyl acetate extract is bacteriostatic for *Bacillus cereus*, *Staphylococcus aureus*, and bactericidal for *Pseudomonas aeruginosa*. However, butanol extract is only bactericidal for phytopathogenic bacteria. Investigation of antifungal activity showed that the ethyl acetate extract has the highest inhibitory effect against *Alternaria alternata* and *Fusarium graminearum* with an inhibition zone of 11 to 15 mm, respectively.
Conclusion: The obtained results suggest that *P. temperata* extract displays remarkable antimicrobial activities that could be exploited as novel tools for pest management.
Keywords: *P. temperata*; antimicrobial activity; antifungal activity.

IBP17: Screening and identification of microorganisms isolated from hot spring producing lipases for industrial and environmental interests

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Background and aim: Enzymes remain the key of biotechnology. The aim of this work is screening, identification and purification of new lipases from microorganisms isolated from extreme environments and which would be most powerful possible in order to fulfill the requirements of the industrial applications.
Methods: the Screening of lipolytic microorganisms was carried out using a plate assay on a solid medium containing 1‰ olive oil, 1 % lysogeny broth, 1.6 % agar and 1‰ rhodamin B. The culture plates were incubated at 55 °C. Free fatty acids form a fluorescent complex with Rhodamine B that is incorporated in the medium give a fluorescent halo that is visible under UV light. These fluorescent colonies were regarded as putative lipase producers. The lipolytic activity is measured using a pH-stat (Metrohm) which compensates for the drop in pH caused by the release of fatty acids by the addition of sodium hydroxide ([NaOH] = 0.1 N) allowing thereafter the conservation of the pH in the reaction medium (optimum pH of action of the enzyme).
Results: Five of twenty thermophilic strains were chosen *Melghirimyces algeriensis* strain NariEX^T sp nov. gene nov, *Melghiribacillus thermophilus* strain Nari2A^T sp nov. gene nov, *Hydrogenophilus hirshii* strain KB-DZ40, *Albidovulum inexpectatum* strain KB-DZ44, and *Anoxybacillus kamchatkensis* strain M1V. Lipase and esterase activities of these strains were measured at different pH and temperature. Our results showed that the maximum of activity was obtained at pH 8 and 55°C. Lipase from Nari2a^T showed the highest crude enzyme activity of 67 U/mL, towards long chain triglycerides (olive oil), under the same conditions lipases from Nari EX^T, KB-DZ44, and KB-DZ40 exhibits their maximum activity around 54, 35, and 15 U/mL, respectively, while, M1V seems to prefer short chain triglycerides tributyrin (TC4) with 22 U/mL. The maximum activity was observed in the case of Nari2A^T so it may be the potential candidate to continue our biochemical and molecular study's.
Conclusion: These thermophilic strains secret lipases can shows high activity in alkaline and thermophilic conditions with future industrial and environmental applications.
Keywords: Screening, Lipase, pH-stat, thermophilic strains, alkaline conditions.

IBP18: The modulatory effect of synthetic heterocyles on the activity of *Saccharomyces cerevisiae* invertase

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Background: Invert sugar (mixture of glucose and fructose) production is one of the main applications of the enzyme called invertase. This enzyme has the advantage of forming invert sugar preferred over sucrose in food and pharmaceutical industry and it is extensively used in confectionaries and food industries. The aim of this study was to investigate the variation of the kinetic parameters of *Saccharomyces cerevisiae* invertase in different conditions and the evaluation of the modulatory effect of six synthetic heterocycles.
Methods: The invertase enzyme was extracted from *Saccharomyces cerevisiae*.. The activity of invertase was studied by varying buffer, pH, temperature, the concentration of enzyme extract and the concentration of sucrose. The kinetic parameters were determined by Lineweaver-Burk plot using various the concentration of sucrose. The modulatory effect was established by incubating the enzyme with six the synthetic heterocycles.
Results: The protein concentration in our extract was 16,35 g/ l. The study of the activity of invertase revealed that the enzyme has a maximum activity at pH 4.4 and a temperature of 55°C. The phosphate citrate buffer gives the best activity (1596 IU/ ml). The highest activities were obtained with the conditions: dilution 1/150, concentration of sucrose 0.1 M and concentration of the phosphate citrate buffer 0.1 M. The Lineweaver-Burk plot allowed us to determine the substrate affinity (Km) value of 1.75 mM and a maximum velocity (Vm) of 2389.5 µmol / min.ml.
The study of the synthesis compounds showed that the products named P1, 2, 3, 4 and 5 had an activating effect, and the product P6 had an inhibitory effect against our enzymatic extract. The activation percentages are between 6.3 and 14.5 %.
Conclusion: the conditions found allowed to have a stable activity that can have potential applications in food, beverage and confectionary industries. The position of hydroxyl on 4H-chromene 3 carbonitrile is very important in the activity of heterocycle ring. The grafting of the hydroxyl groups in the 4-position of the methoxyphenyl at the 3-position, the phenol at the 7-position or the benzene at the 7-position influences the activity of the 4-H-chromene 3 carbonitrile unit.
Keywords: *Saccharomyces cerevisiae* invertase, chemical synthesis, heterocycles, enzymatic activity, kinetic parameters.

IBP19: Lipases enzymes: production by microorganisms

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Background and aim : The study of butter, fat of dairy origin, has shown that the storage conditions lead to a significant change essentially characterized by a hydrolysis of the fat mainly attributed to the lipolytic flora. This transformation is to obtain a new product: the smen, sought to raise the taste of some traditional dishes. It is in this context that our work fits and for this we have been interested in the demonstration of strains producing lipase enzymes and the comparative study of the lipolytic activities obtained.
Methods: The microbiological study focused on six samples (farm butter: 1 and smen: 5) from different regions (El-Oued, Setif, Jijel and Bejaia). After the enumeration of the lipolytic strains and the selection of the potentially lipase-producing isolates, cultures in a stirred Marie bath at 150 rpm for 5 days were carried out on a basic liquid medium containing tween 80 or olive oil with / or without tween 80. The assay of the lipase activity is carried out by titration, by determining the rate of production of fatty acids.
Results: We confirmed the lipolytic activity of 159 isolates and ranked according to the degree of lipolysis on a solid medium, which allowed us to select 49 potentially lipolytic bacteria. The results of the hierarchical ascending classification coupled with the main composite analysis made it possible to select eight strains (SG8, BG32, SG5, SG14, SG9, SG26, SG25, SS46) as bacteria with high lipolytic activity. However, the results of the analysis of variance as well as the hierarchy of lipase production revealed the significant effect of the lipid source contained in the culture medium on the production of lipases. Indeed, the two strains SG8, BG32 have an extreme lipase activity whose lipolytic activity equal to 15.3 ± 0.14 U / mL and 13.10 ± 0.70 U / mL respectively.
Conclusion: These findings confirm the possibility of isolation.
Keywords: microorganisms, lipases, lipolytic activity, smen.

IBP20: Effects of oregano essential oil or chlorine on the quality of modified atmosphere packaged fresh-cut iceberg lettuce

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Background and aim: With the objective to improve quality and safety of minimally-processed vegetables (MPV) products and reduce preservatives, the industry is seeking novel and alternative technologies with the objective of improving quality and safety of food products. The present work focused on the effects of the Modified Atmosphere Packaging (MAP) and the previous addition of Oregano essential oil or chlorine washing on the quality of fresh-cut lettuce during refrigerated storage at +4°C.
Methods: Microbiological analyses such as mesophilic aerobic bacteria, psychrotrophic bacteria total coliforms, lactic acid bacteria, molds and yeast, physico-chimical parameters such as pH, weight loss, water content, chlorophyll content, colorimetric measurements and head space gas composition and sensory evaluation were carried out in this study. All samples were taken at the following stages: 0 (initial stage immediately after treatment), 1^{st}, 2^{nd}, 3^{rd}, 5^{th}, 7^{th} and 9^{th} day of refrigerated storage at + 4 °C.
Results: Our results showed the significant effect of this combined treatment on lettuce quality improvement during refrigerated storage. Thus, mesophilic bacteria was reduced in treated samples compared to those untreated with number which not exceeding the critical of 5.10^7 UFC.g^{-1} ($p < 0.05$). In addition, the O_2 and CO_2 levels created were not significantly different between the two treatments ($p > 0.05$). Brightness of lettuce samples was significantly reduced during storage. These results draw attention to modified atmosphere packaging lettuce and the addition of bio-preservatives which could be an alternative of choice to replace chlorine to preserve the sanitary quality of green products.
Keywords: Lettuce; Oregano; Chlorine; Quality; Conservation.

IBP21: Fungal bioconversion potential of pretreated olivemill solid by-product throughsolid state fermentation

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Background and aim: Food wastevalorization through solid state fermentation toward valuable biomolecules have received increasing attention in recent years. Olive oil production is worldwide widespread but generate a huge amounts of solid residues for which biotechnological management is a challenge issue for the olive oil producer. In this context, the main goalof the present study was to assess cellulases production through fungal bioconversion olive mill solid by-product.
Methods: Olive mill solid by-product was used as solid substrate for bioconversion process by two fungal stains, *Aspergillus niger* and *Penicillium janthinellum*. Solid state fermentations were carried out in Erlenmeyer flasks containing 10 g of dried solid substrate, moisture was adjusted using Mendel and Weber solution. The initial moisture and spores inoculum were respectively 75% and 10^8 spores/mL. Pretreatment with NaOH 2N was carried in order to optimize enzymes production. Phenols and protein quantification was performed after extraction from fermentation flasks. The total reducing sugar as glucose equivalent, protein and total phenol quantifications were assessed respectively, using dinitrosalicylic acid, Bradford and Folin–Ciocalteu methods. DPPH free radical scavenging ability of polyphenol extracts before and after fermentationwas quantified.
Results: Enzymatic activities results after NaOH pretreatment showed that the maximal activities were respectively 0.202U.ml^{-1} at 50°Cand 0.198U.ml^{-1} at 60°C for *Aspergillus niger* and *Penicillum janthinellum.* Antioxidant activity of polyphenol extracts, using DPPH test, recordedbefore and after fermentation for *Aspergillus niger* was (IC 50 value)70±1.96µg/ml for fermentedsubstrate and it was below 50 % before fermentation.
Conclusion: Lignocellulosic biomass such as olive mill solid by-product is complex recalcitrant residue, which requires powerful microorganism degraders. Pretreatment, except for *Aspergillus niger*, is an important first step for efficient carbohydrates conversion processes, and it is required to alter the structure of lignocellulose biomass making polysaccharides more accessible to the enzymes that breakdown them into fermentable sugars. This study provided encouraging results in respect for olive mill solid by-product bioconversion toward valuable biomolecules production.
Keywords: Bioconversion, Olive mill solid by-product, Pretreatment, Cellulases.

Session EB
Environmental Biotechnology

ORAL COMMUNICATIONS
(EBO)

TC1-EB/EBO1: Biogas potential from organic fraction of municipal solid waste: hybrid solid co-digestion batch process

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Background and aim: Municipal solid waste (MSW) generation is significantly increasing in Tunisian urban areas and started creating enormous waste disposal problems. In Tunisia, MSW management is the duty of the local municipalities. More than 90% of the MSW generated is dumped in an unsatisfactory way, what creates environmental hazards to water, air and land. In general the organic fraction of MSW in Tunisia is about 50–68%. The anaerobic digestion is an attractive option for energy generation from the putrescible fraction of MSW as well as for reducing the disposal problem. It has reduced environmental impact, especially with respect to the greenhouse effect and global warming. The aim of this work was to analyze the biogas potential of organic fraction of MSW co-digested with poultry manure. The objectives were to characterize substrates by chemical-fluorescence fractionation method, investigate the performance of OFMSW co-digestion in a hybrid solid batch bioreactor and evaluate the fertilization characteristics of digestate.
Methods: Substrates were characterized with physico-chemical methods and by the combined method of chemical-fluorescence fractionation. Biogas potential was determined by different tools (batch and Flash BMP (biochemical methane potential)). Semi-pilot co-digestion experiment was performed by using hybrid solid batch bioreactor with percolation.
Results: Fractionation and fluorescence spectroscopy analysis have demonstrated the high biodegradability and bioaccessibility of OFMSW in comparison to poultry manure. The determination of the methane potential of substrates in mono- and co-digestion showed a correlation between the biogas yield and the bioaccessibility as well as the complexity index of the organic matter. The enhancement of biogas production from the co-digestion of OFMSW with poultry manure was confirmed at pilot-scale. The co-fermentation was conducted in a bi-phasic reactor with percolate recirculation. An improvement of biogas yield about 22.5% and high stability of anaerobic system were observed. The chemical and phytotoxicity analysis have shown the fertilizing quality of digestate.
Conclusion: The exhaustive characterization performed by chemical-fluorescence fractionation method confirms that co-digesting OFMSW with poultry manure in hybrid solid batch bioreactor with percolation is a feasible option for recovering energy from OFMSW.
Keywords: Anaerobic digestion, Fractionation, Fluorescence, Hybrid bioreactor, Biomethane potential, Digestate.

EBO2: Biohydrogen production from onion wastes by the extreme thermophile *Thermotoga maritima* in a simplified culture medium

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Background and aim: The increased production and consumption of onions (*Allium cepa L.*) generated a surplus of onion wastes due to the rapid growth of phytopathogenic agents. The acidogenic fermentation of onion wastes to hydrogen is an interesting solution for the production of bioenergy and for the reduction of wastes disposal. The aim of this work is to evaluate the potential of hydrogen production from onion wastes by the hyperthermophilic and halophilic bacterium *Thermotoga maritima* in a simplified culture medium based on seawater.
Methods: The batch fermentations were performed in a 2.5 L stirred tank reactor (STR). The hydrogen concentration was monitored by gas chromatograph (GC). The concentration of soluble sugars and fermentation end-products were analyzed by high performance liquid chromatography (HPLC).
Results: The effect of onion wastes addition on H_2 production by *T. maritima* from fruit and vegetable wastes was evaluated. They have significantly improved the production of hydrogen. The cumulative H_2 production and the maximum H_2 productivity were 281.4 mmol/L and 23.7 mmol/h.L, respectively. The consumption of carbohydrates was 79% allowing H_2 yield of 3.75 mol H_2/mol hexose. This improvement could be attributed to the ability of *T. maritima* to utilize the compounds supplied by onion wastes. These results encouraged to use onion wastes alone as feedstock for hydrogen production in a simplified culture medium based on seawater. Using 202 g/kg of onion wastes allowed a cumulative H_2 production of 124 mmol/L. The consumption of glucose and fructose by *T. maritima* was totally allowing H_2 yield of 3.76 mol H_2/mol hexose. Therefore, hydrogen production from onion wastes was attributed to its composition rich in sugars, sulfur compounds, vitamins, and minerals.
Conclusion: The feasibility of the hyperthermophilic production of hydrogen by *T. maritima* from onion wastes in seawater was approved. This process could be an economical and effective solution to valorize an agricultural waste characterized by its strong odor in a cheap medium.
Keywords: Biohydrogen, onion wastes, *Thermotoga maritima*, hyperthermophilic dark fermentation, seawater.

EBO3: Fermentative hydrogen and methane co-production from anaerobic co-digestion of organic wastes at high loading rate coupling continuously and sequencing batch digesters

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Background and aim: Study for the first time the anaerobic co-digestion of the major available organic wastes in the two-stage anaerobic system, coupling continuously stirred tank reactor (CSTR) and anaerobic sequencing batch reactor (ASBR). This approach met to produce bio-hydrogen by dark fermentation and methane by anaerobic digestion with the aim to use this gas separately or to constitute the bio-hythane.
Methods: Anaerobic co-digestion of organic waste was carried in first step in single stage reactors (ASBRs) and in the second step in two stage reactor. The biogas production rate, the mass balance and energy recovery assessments were also performed using physical and chemical analysis.
Results: The anaerobic co-digestion of the most abundant organic wastes was investigated for enhancing biogas production rate and quality. The used feedstock was composed of fruit and vegetable waste (FVW), waste-activated sludge (WAS), olive mill wastewater (OMW) and cattle manure (CM). A considerable methane yield of 340 L/kg volatile solid (VS) $_{inlet}$ was obtained using single stage (ASBRs). However, VS biodegradation becomes difficult at high organic loading rate (OLR). Therefore, the reactor (CSTR) was integrated to the (ASBR) for waste pre-digestion. The dark fermentation leads to the improvement of organic matter solubilisation and bio-hydrogen productivity, reaching 0.73 L/L/day (H_2content of 49.8%) when pH decreased to 5.8. Therefore, methane productivity increased from 0.6 to 1.86 L/L/day in the methanogenic reactor with a better VS biodegradation (91.1%) at high OLR. Furthermore, the bio-hythane production was performed through a controlled biogas recirculation from the dark fermentation stage into the methaniser to reach 842.4 L/kgVS $_{inlet}$. The produced biogas was composed of 8% H_2, 28.5% CO_2 and 63.5% CH_4.
Conclusion: Therefore, two-stage anaerobic co-digestion with coupled CH_4 and H_2 recuperation may be an important contribution for pollution control and high-rate bioenergy recovery (21.1 kJ/g VS inlet) from organic wastes.
Keywords: Dark fermentation. Anaerobic co-digestion. Organic wastes. Bio-hythane production

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EBO4: *Pseudomonas protegens* with high antifungal activity protects apple fruits against Botrytis cinerea-gray mold

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Background and aim: Certain soil bacteria are able to enhance plant growth through several mechanisms. They are referred to as Plant Growth Promoting Bacteria (PGPB). Using such beneficial bacteria to promote plant growth and to ensure phytopathogen control has proved its efficiency as ecofriendly alternative to the excessive use of chemical fertilizers and fungicides. This work aimed to isolate and select new bacterial strains with high ability to protect apple fruits against *Botrytis cinerea*-gray mold.
Methods: Herein, one bacterial strain (RhiNA) was selected, among others, from an agricultural land in northern Algeria (Bejaia). Selection was based on the strain's ability to produce antifungal and plant growth promoting (PGP)-metabolites. It was then molecularly identified and screened for its antagonistic activity against *Botrytis cinerea*, *Mucor* sp., *Aspergillus niger* and *A. flavus*. Finally, the isolate was tested for its ability to attenuate gray *B. cinerea*-gray mold on apple fruits.
Results: The studied bacterium, identified as *Pseudomonas protegens*, showed high in vitro ability to produce hydrolytic enzymes, IAA, siderophores, HCN, ammonia and efficiently inhibited mycelial growth and spore germination the four tested fungi (PGI: 66, 66, 62 and 58%; SGP: 14.32, 55.10, 28.92 and 10.15% against *A. niger*, *Mucor* sp., *B. cinerea* and *A. flavus*, respectively). Only 172.823 mm^2 of each apple fruits inoculated with *P. protegens* were touched by *B. cinerea*-gray mold, compared to 529.74 mm^2 of rotted zone in absence of the antagonistic strain.
Conclusion: The isolate *P. protegens*-RhiNA proved its high efficiency against virulent strain of B. cinerea. Thus, it could be used as promising agent to control postharvest phytopathogens for sustainable agriculture in future.
Keywords: postharvest diseases, *Aspergillus*, *Mucor*, *Botrytis*, Chitinase, Siderophores.

EBO5: Protective effect of *Zizyphus lotus* jujube fruits against cypermethrininduced oxidative stress and neurotoxicity in freshwater mussel *Unio pictorum*

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Background and aim: Plants are frequently used in phytotherapy for their biological activity. The present study aimed to evaluate the therapeutic potential of *Zizyphus lotus* fruits extracts against toxicity induced by cypermethrin in the freshwater mussel *Uniopictorum*.
Methods: The protective effect of Zizyphus lotus jujube fruits on freshwater mussel was investigated through of against cypermethrin-induced oxidative stress and neurotoxicity in freshwater mus sel Unio pictorum. Mussels were exposed to 1 50µg/L of cypermethrin and/or *Z. lotus* fruits extracts in the following concentrations 0.5, 1, 2.5 and 5g/L during 2 and 4 days.
Results: Cypermethrin increased hydrogen peroxide levels in *Unio pictorum* gill and digestive gland tissues, while a treatment with *Z. lotus* fruits extracts decreased those levels to the normal. It inhibits the catalase activity, which increased in groups treated with *Z. lotus* fruits extracts. Similar for the malondialdehyde and acetylcholinesterase, levels of both enzymes were affected by cypermethrin, and are almost regulated in groups receiving *Z. lotus* fruits extracts; it seems that 1and 2.5 g/L of *Z. lotus* fruits extracts were effective for the restoration of the activities of both enzymes to the normal status.
Conclusion: Our data, prouved the toxic effect of cypermethrin on *Unio pictorum* gill and digestive gland tissues, mussels biomarkers profiles can be corrected by a treatment with *Z. lotus* fruits extracts.
Keywords: *Zizyphus lotus* fruits extracts, *Unio pictorum*, Cypermethrin, Biomarkers, Oxidative stress, Neurotoxicity.

EBO6: Developing of a biocide to fight against Thaumetopoea pityocampa phytophage of Aleppo pine forests

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Background and aim: As part of the operation and enhancement of genetic plant resources of the Mediterranean areas. we chose *Nerium oleander* characterized by its toxicity against phytophagous insects to prepare an insecticide.
Method: The extraction by maceration of the aerial parts of oleander by using two solvents characterized by distinct polarity values.
Results: This work aims to develop a biocide based to bioactive substances of Neium oleander and evaluating its insecticidal effect in order to fight against *Thaumetopoea pityocampa* who is a dangerous phytophagous ofAleppo Pine. This method allowed obtaining extracts which have presented a powerful insecticide that can induce mortality of phytophagous insects. Using a sprayer we applied the extracts obtained from individuals of the track by direct contact. Then we evaluated the mortality rates (Figure 1) which were 88%, 40% and 10% respectively for the filtrate of leaves, petals, chalices macerated with methanol after a contact time equal to 10 minutes. Under the same conditions and with the same method we estimated the mortality of individuals by extracts pulverized leaves, petals and calyx macerated by n-hexane which were of the order of 100%, 80% and 10% respectively. The analysis of variance shows that there is a significant difference between the values of the mortality rate because of the effectiveness of treatments made.
Conclusion: This study shows that the use of biocides from *Nerium oleander* will bring success in the fight against Thaumetopoea pityocampa and consequently great ecological interest.
Keywords: *Neium oleander*, Biocide, valorization, maceration, processionary chenille.

EBO7: Improved real textile wastewater decolourization by fungal bioaugmentation technology: Application process at SITEX pilot-scale plant

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Background and aim: The textile processing is an important sector in the Tunisian industry. However, the persistence and toxicity of used dyes cause serious problems for the natural ecosystem. The aim of this work consists to examine the effect of bioaugmentation of activated sludge process (ASP), treating a real textile wastewater (RTW) at SITEX pilot-scale plant, using a novel fungal strain.
Methods: Several filamentous fungi were previously isolated. The best microorganism was screened on the basis of its potential for soluble COD removal efficiency, color reduction and enzymes production. The selected fungus strain was identified based on the analysis of the nucleotide sequence of the nuclear ribosomal ITS1-5.8-ITS2 region. Bioaugmentation strategy of (ASP), by using *Chaetomium globosum IMA1 KJ472923*, has been proposed at SITEX pilot-scale plant.
Results: The bioaugmented system at pilot scale plant showed a better chemical oxygen demand (COD) removal efficiency (91,3%) compared to system without addition of the strain (93,3%). Further, this application enhanced significantly color removal (OD620) efficiencies from 90, 3% to 98, 4% when the newly fungal strain was added. In addition, results indicated that this method improved the efficiency of the system to reduce the level of pollution parameters such as total suspended solids (TSS) with highest removal yield which reached 99%. Thus, positive effect of the fungal bioaugmentation is noticed on TSS of the colored effluent.
Conclusion: Fungal bioaugmentation could be used as an effective and efficient method to improve a decolourization process facing sudden toxic pollutant shock loading.
Keywords: Dyes, Biodegradation, Real Textile Wastewater, Fungal Bio-augmentation, Activated Sludge System.

TC2-EB/EBO8: Microbial peroxidases as promising tool for lignin and humic acid biodegradation, and other environmental applications

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Background and aim: Peroxidases (EC 1.11.1.7) are the ubiquitous enzymes represent a large family of isoenzymes found in almost all living organisms. In general, peroxidases are heme proteins containing iron(III) protoporphyrin IX or ferriprotoporphyrin IX (four pyrrole rings are coordinated to Fe(III)) as the prosthetic group. Peroxidases may be categorized as mammalian and plant peroxidases ranging in molecular weight from 35 to 100 kDa. Peroxidases or a group of oxidoreductases typically catalyze biological reactions, in which peroxides such as hydrogen peroxide (H2O2) and alkyl hydroperoxide (ROOH) are reduced, while a redox substrate acting as an electron donor is oxidized. It should be noted that the nature of the electron donor is very much dependent on the structure of the enzyme. Through this catalysis, peroxidases can scavenge $_{H2O2,}$ a naturally occurring by product of oxygen metabolism in human body, resulting in the formation of water and oxygen. In this way, peroxidases play an important role as highly efficient antioxidant defense systems to combat complications engendered by reactive oxygen species. In fact, there is a large body of literature showing that a number of naturally-occurring phenolic monomers can undergo oxidative coupling reactions catalyzed by oxidative enzymes (laccases and peroxidases), to produce humic substances-like. Microbial peroxidases have often been reported to constitute a resourceful class of enzymes with promising industrial applications.
Methods: The biochemical characterization of the some pure microbial peroxidases were investigated though physico-chemical determination as well as spectroscopy analysis.
Results: To the author's knowledge, this is the first work describing, the purification and characterization of a novel three peroxidases MnP TP55 from *Trametes pubescens* strain i8; and LiP BA45 and MnP BA30 from *Bjerkandera adusta* strain CX-9 and 4 novel humic acid peroxidases (HaP1-4) from decolorizing actinomycetes strains isolated from Algerian and Tunisian sites. We also explore their promising potential for their useful tool for lignin and humic acid biodegradation, and other environmental applications.
Conclusion: Overall, some peroxidases MnP TP55, LiP BA45, and MnP BA30 as well as HaP1-4 are considered as good candidates for future application in lignin and humic acid biodegradation, and other environmental applications.
Keywords: white-rot fungi; actinomycetes; environmental applications.

EBO9: Elaboration of porous bead based on iron pillared clay and their application in the elimination of a mixture of dyes

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Background and aim: The aim of the present work consists to develop a new adsorbent which is a porous beads based principally on iron modified clay and sodium alginate. This adsorbent was used in the adsorption of two organic pollutants (dyes) Green Malachite (GM) and Rhodamin B (RhB) in discontinuous systems.
Methods: In this work we have firstly prepared the iron pillared clay according to previous reported methods. Alginate acid sodium salt PVA and CaCO3 were used in preparation of the beads at different ration. The experimental tests were carried out in discontinuous system using initials concentration of 100 mg/L for the pollutants. The desired dye concentrations were prepared through dilution by distilled water of a stock solution at a concentration of 1000 mg L^{-1}. The experimental tests were realized in shaker at rate of about 200 rpm in brown flasks. Kinetic study was carried out at pH medium. Adsorption isotherm has done at varied pH values. The residual concentrations of MG and RhB were determined by UV spectrophotometry (6800 UV/VIS, Jenway) at 618 and 554 nm, respectively.
Results: The different adsorption tests of the two dyes in discontinuous system at initial concentration of about C0 = 100 mg.L-1 reveals an important removal percent of RhB and GM (90%) at equilibrium contact times of 3 to 6 h. The modeling of adsorption isotherms show that Freundlich model fit very well the results R^2= 96%) compared to other models. In binary mixture of the couple (GM/RhB), the adsorption capacities were enhanced compared to those obtained in single system. These results can be explained by the strength interaction between green malachite which cationic dye and rhodamine which is zwiterionic one.
Conclusion: The adsorption capacities of the two dyes in single system were observed at acid medium. The equilibrium contact time for the GM and RhB is from 3h to 6h. In binary mixture the presence of GM with RhB enhances its adsorption. The synergetic mechanism between RhB and GM explain the increases in the adsorption capacities onto the prepared porous bead.
Keywords: iron pillared clay, competitive adsorption, green malachite, rhodamine, single system, Freundlich model, sheindorfmodel.

EBO10: Studies on the biodiversity of thermophilic bacteria isolated from hammam Righa hot spring (Algeria) under aerobic conditions

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Background and aim: For several decades, thermophilic bacteria have attracted the interest of many scientists due to their biotechnological potential in addition to scientific curiosity. In particular, phenotypic and genotypic characterization of thermophilic bacteria has been done for many geothermal areas in different regions in the world, including Turkey, Italy, Bulgaria, Greece, China, India, Yellowstone National Park and Iceland. In Algeria, there is no information with regard to indigenous thermophilic aerobic microorganisms inhabiting these extreme environments so far.The main objective of this study was to isolate and characterizethermophilic aerobic bacteria from HammamRigha hot spring(GPS coordinates: 36°22'N; 2°23'E) E), located in AinDefla province northern Algeria.

Methods: The thermophilic isolates were identified by by using phenotypic (morphological, biochemical and physiological features) and phylogenetic approaches (16S rRNA gene sequence analysis). Moreover, hydrolytic activities of these isolates that we obtained were determined

Results: A total of 70 thermophilic bacterial strains were isolated from this Hammam. The optimum temperature of isolates was 60°C. Totally 40 bacterial strains were selected for this study. The phenotypic characterization (morphological, physiological, and biochemical tests) of those isolates was confirmed by genotypic method using 16S rRNA sequence analysis. 16S rRNA gene analysis found them related to *Tepidimonas, Meiothermus, Albidovulum,Hydrogenophilus,Anoxybacillus, Geobacillus*, and*Bacillus*genera.Positive results on several enzymes such as amylases, proteases, cellulases, xylanases,chitinases, esterases and lipases of most isolates are indication of potential applications of these bacterial products in biotechnology.

Conclusion: This is the first investigation of thermophilic aerobic bacteria originating from HammamRigha hot spring. The results showed a clear dominance ofthermophilic*Bacillaceae.* Our results extend our knowledge of microbial diversity existing in hot springs. They (i) suggest that there are biological markers of hot springs (ii) indicate that new thermophilic populations can be found in these hot environments. They provide evidence that the HammamRigha possesses a rich microbial diversity to be explored by further studies.

Keywords*:* Biodiversity, Thermophilic bacteria, HammamRigha, Hydrolytic activities Biotechnology.

EBO11: Eco-friendly degreasing processes of sheep skins and hides: Potential application of a yeast lipase in leather industry

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Background and aim: Global environmental regulations are changing the leather-processing industry. Tanning processes contribute 80-90% of the total pollution in the industry and generate noxious gases, such as hydrogen sulfide, as well as solid wastes, such as lime and chrome sludge. In order to overcome the hazards caused by effluents containing hazardous chemicals and organic solvents, enzymes have often been proposed as viable alternatives to chemicals to improve the efficiency and cost-effectiveness of a wide range of industrial processes. The aim of this work is to substitute chemical leather process with a microbial lipase as a degreaser suggesting more ecofriendly tanning processes in the future.
Methods: Several conditions were studied for maximizing the enzymatic degreasing of sheep skins using lipases. The mechanical properties of the leather have been investigated after enzymatic leather process on universal testing machine. The biochemical oxygen demand (BOD) and chemical oxygen demand (COD) parameters of effluent were analyzed.
Results: The yeast *Yarrowia lipolitica* lipase (called YLLIP2) exhibits very high specific activities and remarkable lipolysis rates on long chain triglycerides than the commercial LipolaseTM from the fungus *Thermomyces lanuginosus* widely used in leather process. Several reaction media were carried out to optimize the degreasing of sheep skins and hides using these lipases as alternatives to chemicals leather processing. Maximal degreasing yield could be obtained at pH value of 8.0, agitation rate 220 rpm and temperature 37 °C with the enzyme amount about 4000 U/mL within only 6 hours. The results between the chemical and enzyme processed leathers were compared. It was found that the YLLIP2 processed leather has a high tensile strength (15.35 N/mm^2) and a percentage elongation (19%) as compared to the chemical processed leather. The enzyme based leather processing showed that natural skin color was preserved along with a drastic reduction of BOD and COD values of effluent.
Conclusion: The YLLIP2 lipase, used as a degreaser for boosting the quality of leather, could be an eco-friendly alternative to the conventional chemicals leather process.
Keywords: Leather, Degreasing, Lipase, Physical properties, Effluent analysis.

EBO12: Development of an environmentally friendly approach of feather degradation using a new keratinase from Actinomycetes

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Background and aim: Currently used chemical treatment of the keratinous material is the preferred method of poultry waste disposal in Tunisia, however it is costly, and environmental pollution problems from the chemical leachates as well as the poor nutritional quality of the end-product render this method undesirable. Keratinases have high activity for keratin substrates. This is mainly due to their high stability and activity under harsh operational conditions as well as their significant abilities to hydrolyze various keratinous substrates. Keratinolytic microorganisms and their enzymes may be used to enhance the digestibility of feather keratin. They may have important applications in processing keratincontaining wastes from poultry and leather industries through the development of nonpolluting methods
Methods: The biochemical characterization of the keratinase KERDZ from strain DZ50 was investigated though physico-chemical determination as well as spectroscopy analysis.
Results: *Actinomadura viridilutea* strain DZ50 was able to grow in medium M containing 10 g/L of feather-meal, chicken feather, goat hair, bovine hair, and sheep wool (as a sole carbon and nitrogen source). Simultaneously to the keratin-degradation, increase of protein level and sulfhdryl groups was observed. Of the seven keratin substrates tested, feather-meal was the most strongly degraded (100%), followed by chicken feather (93%), duck feather (86%), goat hair (75%), bovine hair (5 8%), and rabbit wool (34%) whereas sheep wool showed relatively low degradation rate (15%). Furthermore, when the purified KERDZ was incubated with native chicken feathers, total degradation was observed after 48 h, with a simultaneous increase of protein concentration and sulfhydryl group formation, while no degradation was observed with the control.
Conclusion: These properties make KERDZ a potential, promising and eco-friendly alternative to the conventional chemicals used for industrial applications. The application of this system may improve the rapid disposal of poultry waste as well as the public and environmental health in the region.
Keywords: Keratinases, Actinomycetes, Feather degradation.

EBO13: Feasibility and Performance Study of the Ground-source Heat Pump System For Heating Greenhouse

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Background and aim: The heating system has a strong effect on the time of cultivation, quality and quantity of the products. The auxiliary systems cannot meet alone the heating requirements due to the relatively high cost of energy and big greenhouses surfaces. Therefore, it seems that the use of a suitable heating system, with low cost, is crucial to furnish optimum indoor conditions during cold months. Therefore, the most renewable energy adapted in heating greenhouses seems to be the geothermal energy. The aim of this work is to evaluate the thermal performance of a geothermal heat exchanger as an horizontal form to be used for greenhouse heating.
Methods: The considered system is basically composed of: a horizontal heat exchanger buried at a depth of 1m, storage tank and circulation pumps. The heat pump is coupled with heat exchanger in order to obtain a comfortable climate under greenhouse in the winter conditions. The effect of this heating way was investigated through yield.
Results: To estimate the greenhouse heating needs, a model was developed using Trnsys16. We present a mathematical description as well as the numerical results of the simulation of the system. The exploitation of the geothermal energy has increased nocturnal air temperature inside the greenhouse of 6° C. In fact, the COP reaches 3.18 for a 14.8 m^2 greenhouse. That good performance was proved experimentally. In fact, for a 14.8 m^2 greenhouse a precocity of 7 days was found and a gain of about 3 kg/ plant was also realized for the best category of fruits (category A) which is a significant economic result.
Conclusion: This work was devoted to study the efficiency of a greenhouse solar heating system in order to make it appropriate for agricultural purposes during coldest period of the year (January–April). It is noted that the system is totally sufficient for a small greenhouse. Greenhouse heat pump heating system is a profitable system which does not pollute the atmosphere.
Keywords: Heat pump, heat exchanger, greenhouse, Performance

EBO14: Effect of Solid State fermentation by *Chaetomium globosum* on the anaerobic co-digestion performances of municipal solid waste with the urban and industrial sludge

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Background and aim: The anaerobic co-digestion of urban and industrial sludges with municipal solid waste is a good alternative to improve process performances in terms of methane production, biodegradability and microbial community balance. However, the startup of the process remains slow because of the lignocellulosic nature of this waste. The aim of this work is to overcome the hydrolysis step and to accelerate it to improve the process performances.
Methods: In this work, solid state fermentation (SSF) was carried out using a soft rot filamentous fungus *Chaetomium globosum*. The effect ofthis pre-treatment is revealed by the determination of the ligninolytic and hydrolytic enzymatic activities, the FTIR spectroscopy and the percentage of solubilization. The equilibrium of the microbial communities was revealed by the fluorescence in situ hybridization method (FISH).
Results: Results shows that the limitation of the hydrolysis step has been overcome. The activity assay of ligninolytic (LiP and MnP) and hydrolytic enzymes (Proteases) revealed a maximum production of 1014,8 IU L, 137,5 IU / L and 384 IU / L, respectively. These results associated with FTIR spectroscopy and the percentage of solubilization (55,45%) confirm the partial or total degradation of lignocellulosic complex. The best results from the anaerobic co-digestion were obtained with the co-digestion between industrial sludge and pretreated municipal solid waste. Biogas production was 0,62 $_{l/gVSremoved}$ with 71% CH4 and the percentage of methanogens reach 84 %. The removal percentage of VS was 78%. The process stabilization was obtained after 15 days. The energy produced increases to reach 14,27 kJ/gTS which confirm the efficiency of the anaerobic co-digestion associated to a pretreatment step by SSF.
Conclusion: The achievement of an integrated process for the improvement of biodegradability and methane production through the solid-state fermentation of solid municipal wastes and the co-digestion process made it possible to overcome the limitation of the hydrolysis phase due to the ligninocellulosic nature of municipal solid waste.
Keywords: Anaerobic co-digestion, Urban and industrial sludge, Municipal solid waste, Solid state fermentation, FISH, Energy balance.

EBO15: Potential of Duckwwed (*Lemnagibba*) for removal of Nitrogen and Phosphorus and mater organic from dairy wastewater

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Background and aim: Dairy wastewater is considered as a significant source of organic matter and nutrient contamination of ground water and surface water. Nutrient pollution in water bodies caused by nitrogen and phosphorus has become a recent focus of concern.The aim of this work is to evaluate the potential of *Lemnagibba* (duckweed) for removal nitrogen and phosphorus and mater organic.

Methods: Samples of wastewater were obtained from the dairy. The $P\text{-}PO_4^{3-}$, NTK and COD were measured for each sample.To determine the removal of Nitrogen (Total Kjeldahl Nitrogen NTK), Phosphorus, and matter organic in term of COD (Chemical Oxygen Demand) by duckweed, we cultured *Lemnagibba*, a common species of duckweed, in 250 mL diluate(1/100) dairy wastewater for 24 h and 72 h and 168 h, respectively.

Results: The results showed that in average $P\text{-}PO_4^{3-}$, NTK and COD in the diluate dairy wastewater was 0.13 mg/L, 0.65 mg/L and 127 mg/L, respectively.The removal capacity of duckweed for N and P and COD was higher after 168h of culture(35% P ; 45%NTK et 37% COD). During the first 24 hours removal are quite low and seems to be related to the acclimation of the plant to its new environment.

Conclusion: Our research showed that longer cultivation time helped duckweed to remove more N and P and COD. Our results suggest that L. gibbashould be used to remove N and P and COD from dairy wastewater.This plant, whichis often used for toxicity tests, also appeared to be good candidate for phytoremdediation of dairy wastewater.

Keywords: phytoremediation, Lemna, nitrogen, phosphorus, matter organic.

POSTER PRESENTATIONS
(EBP)

EBP1: Daily monitoring of colonisation, by free-living marine nematodes, of azoic sediments made coarser

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Background and aim: Various studies have investigated the colonization of azoic sediments by free-living marine nematodes. However, the evolution over time of this colonization remains to this day a major unknown for science. To remedy this, an opened microcosm and a daily sampling technique were developed during 25 days.
Methods: The experimental design was made by using plastic pipes divided into several compartments with different sediment qualities. Once inside the pipes, meiobenthic nematodes faced two migratory choices: (1) an azoic sediment,made coarser by broken shells of *Mytilus galloprovincialis* (M), and (2) an azoic sediment, made coarser by minced leaves of *Posidoniaoceanica*(P). These two compartmentswere connected to a third one, noted PM, filled with azoic sediment made coarser by mixing broken mussel shells and minced *Posidonia* leaves. Nematodes sampling was conducted from holes drilled at regular intervals all through the microcosm, thus tracking the progress of colonization day by day.
Results: The results revealed the high exploration capacities of some species, such as *Marylinia punticaudata* and to a lesser extent *Oncholaimus campylocercoides*. These omnipresent species were sampled every daythroughout the whole microcosm. The daily monitoring also revealed that some species like *Daptonama Trabeculosum* and *Prochromadorella longicaudata*, were frequently sampled during the first days of the bioassay but decreased until a complete disappearance at half of the experimental period. Finally, the ultimate compartment PM showed a remarkable presence of three species, *Synonchiellaedax*, *Metoncholaimus pristiurus* and *Sabatieria pulchra*, who reached successfully that last level even they were rarely inventoried before.
Conclusion: The set-up sampling technique throughout the opened microcosm, allowed us to establish a global schema of the differential colonization of azoic sediments by a natural nematode community.
Keywords: free-livingmarine nematodes, migration, monitoring, sampling technique, azoic sediment, opened microcosms.

EBP2: Elimination of organic pollution from dairy wastewater by electrocoagulation: Effect ofthe operating parameters

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Background and aim: Urban or industrial effluents are loaded with pollutants of different types. Dairy wastewater treatment is a serious problem, in particular that they are loaded with organic matter. When spilled without prior treatment; cause a harmful damage to the receiving environment. To overcome the pollution that cause a great deal of damage to the ecosystem and the water resources, many processes for wastewater treatment are implemented. Among the various possible solutions, we examine in this study, the purification of a dairy effluent by the electrocoagulation technique.
Methods: Electrocoagulation is a technique derived from conventional coagulation, it eliminates colloidal particles and dissolved pollution through the in situ production of hydroxide flocks obtained by anodic dissolution of soluble electrodes. It has proved its effectiveness for the elimination of different pollutants in different industrial or urban effluents.
To test the effect of some operating parameters: current density, treatment duration, surface state of the electrodes, a series of runs was conducted in an electrochemical cell in continuous recirculation mode. The evolution of the COD, the turbidity and the concentration of aluminum in solution were analyzed as a function of time.
Results: The obtained results show that the efficiency of the electrocoagulation is proportional to all the parameters tested. The abatement rate of the DC0 and the turbidity reaches 80% and 95 respectively after 30 min with a 150A / m2 applied current, higher processing times and currents increase energy consumption. The repeated use of aluminum electrodes attacks the electrode surface, which increases the concentration of aluminum in the effluent to be treated and decreases the technique efficiency.
Key words: Dairy effluent, organic pollution, electrocoagulation, aluminum electrodes

EBP3: Interaction between AuTiO2 nanoparticles and benzanthracene in the Mediterranean mussels (*Mytilus galloprovincialis*)

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Background and aim: Laboratory experiments studying the interaction between nanoparticles and emerging contaminants in aquatic organisms are still scarce. In addition, to measure the consequences of these interactions at the populations and communities levels, we must be able to determine the effects at individuals. The aim of this work is to evaluate the effect of the AuTiO2 nanoparticles and benzanthracene hydrocarbon on mediterranean mussels *Mytilus galloprovincialis*.
Methods: The response of marine mussels to several contaminants was investigated through filtration rate capacity of mussels and enzyme activities in gills, digestive gland and mantle.
Results: AuTiO2 Results of the single-chemical exposure tests highlighted the relatively low toxicity of AuTiO2 NPs. In contrast, Ba exposure resulted in clear filtration capacity reduction and enzymatic modification in organ dependent manner showing the toxicity level of this hydrocarbon to mus sels. In addition, the gills are the more sensitive to contamination than digestive gland and mantle. Based on biochemical results, we hypothesize that under acute exposure, benzanthracene might interact with cell metabolism leading to oxidative stress (increase of MDA level and catalase activity) and acetylcholine receptors, thereby affecting oxidative and neuromuscular status.
Exposure tests using mixtures ofAuTiO2 NPs and Ba revealed that these two substances were acting in an antagonism manner on the selected biomarkers.
Conclusion: Physiological response and biochemical biomarkers determination in the mussels are strongly recommended to confirm the toxicity of hydrocarbon on marine bivalve. Gills could be used as a target organ of hydrocarbons contamination in marine ecosystem. Finally, interaction between nanoparticles and hydrocarbons could occur in marine organism witch influence their toxicities.
Keywords: AuTiO2 nanoparticles, Benzanthracene, Biomarkers, Antagonism, Biomonitoring

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EBP4: Adsorption of methyl orange dye on CTAB functionalized almond shell powder: Optimization, kinetics, isotherms

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Background and aim: With respect to all methods, adsorption is a common and effective method for heavy metal ions removal because of low cost, high efficiency, and good operational conditions.
Methods: The present investigation focuses on the removal of methyl orange (MO) by cetyltrimethylammonium bromide (CTAB) functionalized almond shell powder. Various process parameters such as effect of adsorbent/chemical dose, reaction temperature and contact time were experimented through batch study and statistical method to study the effect ofthese parameters on dye removal efficiency.
Results: The analysis of various models (Langmuir, Freundlich, Toth, Dubinin– Radushkevich, Elovich and Temkin model) were performed. The Freundlich isotherm model was best fitted (R2 = 0.999) with the experimental data, which confirmed interactions of heterogeneous sites and the formation of multiple layers. In addition, the MO adsorption onto CTAB-modified AS fit the pseudo-second order model, with a correlation coefficient (0.999 ≤) greater than the pseudo-first order model. The qe (calculated) values obtained in the pseudo-second order model were also closer to the qe (experimental), confirming the better fit to the model. This result is indicative of the occurrence of chemisorption, since the pseudosecond order model assumes that adsorption is controlled by processes of chemical interactions. ΔG° values, which were in the range of $-20 < \Delta G^{\circ} < 0$, showed that the adsorption of MO by CTAB-modified AS is a spontaneous physical process. In addition, the negative values of $\Delta H°$ demonstrate the exothermic nature of the adsorption process. In addition, the values of ΔS are negative, suggesting the decreased randomness at the solid/solution interface. To evaluate reusability of new adsorbents, deionized water, absolute ethanol and acetic acid (1M) were used in desorption and the absolute ethanol was used as a regenerating agent during 60 min. The absolute ethanol showed the best desorption efficiency.
Conclusion: These results demonstrate that the CTAB modified almond shell powder is highly efficient in removing MO dye from aqueous solution and suggest that it can be used to treat MO-polluted wastewaters.
Keywords: almond shell, CTAB, methyl orange, functionalization, Adsorption.

EBP5: Screening of marine fungal diversity to identify new oxidases for biotechnology and sustainable development

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Background and aim: Most of the fungi studied to date are isolated from the forest and terrestrial environment, while very few studies have focused on the exploration of marine fungal diversity. The objective of this study is to characterize the fungal adaptation to the halophilic medium by (i) screening the main lignocellolytic enzyme activities, (ii) analyzing the global physiology of a marine fungus model by a differential proteomic analysis and (iii) producing new enzymes of interest for sustainable applications.
Methods: Marine fungal strains were purified by a repetitive subculturing of the isolated strains. They were identified by molecular and morphological analysis. Identified strains were cultured on medium containing cellulose and xylanase and their growth measure daily. In addition, they were grown on medium containing wheat straw or sea gras s to assay their main ligninolytic activities using 2,2'-azino-bis-(3 -ethylbenzthiazoline-6-sulfonic acid (ABTS), azo-CM-cellulose, azo-xylan, and azo-carob-galactomannan.
Results: We collected fungal samples from the harbour of Sfax, Tunisia.The molecular and morphological identification of these samples showed that the isolate fungal strains correspond to *Trichoderma sp., Stemphylium sp.* and *Aspergillus nidulans*. Analyzing their growth on cellulose and xylan, *Stemphylium* demonstrated a better growth in the presence of salt. Testing their enzyme activities of the strains grown on straw and sea grass, we showed that sea salts have a negative effect on laccase and xylanase activities for all the strains, except for *Trichoderma* sp. Isolate 1 and *Stemphylium*sp activities grown on CMC and ABTS, respectively. In addition, activities on ABTS and xylanwere higher for the fungi grown on wheat straw, except for *Stemphylium* grown on algae (factor 4).For the two other substrates, activities were in general higher in sea gras s cultures. Taking into account all the results, *Stemphylium* was selected as a model to study the physiological adaptation to the marine environment.
Conclusion: The next step of my research will consist in establishing a comparative proteomic analysis of *Stemphylium* grown on wheat straw and sea gras s in order to identify the enzyme machinery specific to the terrestrial and the marine substrates
Keywords: *Trichoderma sp., Stemphyliumsp, Aspergillus nidulans*, ABTS, azo-CM-cellulose, azo-xylan, azo-carob-galactomannan.

EBP6: Effective removal of heavy metals and nutrients excess by phytoremediation using *Lemna gibba*

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Background and aim: Heavy metal contamination and eutrophication of aquatic ecosystem are global environmental problems. Phytoremediation is an emergent technology using selected plants to clean up the contaminated environments. *Lemna gibba* is an Algerian autochthonous species can be a suitable candidate for removal of heavy metals from pollutant water bodies and for the reduction of nutrients excess (nitrate and phosphate) that cause eutrophication. The aim of this studyis to evaluate the effectiveness of the aquatic plant *Lemna gibba* in the simultaneous removal of cadmium (Cd), lead (Pb), individually or in combination, nitrate ($NO3^-$) and phosphate ($PO4^{3-}$) from a synthetic medium.
Methods: The metals analyze was carried out by means of atomic absorption spectrophotometry. The phosphate and nitrate concentrations of the growth medium were analyzed, using the ascorbic acid and salicylate methods, respectively.
Results: Overall, in all treatments, orthophosphates, nitrate, Cd and Pb concentrations in the medium, decreased markedly within the two days of initiating experiments.
The maximum phosphate removal efficiencies were recorded on the fourth day at 1 mg Cd/L (90.32%) and at 1 mg Pb/L (97.57%).Whereas, nitrate removal was 47.22-79.31%and 48.57- 80.91% for Cd and Pb containing set-ups respectively. In the binary mixtures, phosphate removal efficiency exceeds 80%. Nevertheless, nitrate removal was weak with time.
In the binary mixture of Cd and 0.1 mg Pb/L, removal percentage ranged between 41- 100% for Cd and 66-100% for Pb, whereas, weak Cd removal efficiencies were recorded when the duckweed was exposed to mixtures of Cd and 1 mgPb/L. The maximum Pb removal efficiencies were 57% and 86.58% respectively at 1 and 10 mg/L of Pb alone.
Conclusion: The present study suggested that *L. gibba* has a high tolerant ability to Cd and Pb. High removal efficiencies were achieved over the exposure period. Phytoremediation using *L. gibba* seems to be a good option for the simultaneous removal of heavy metals and nutrients (nitrate and phosphate)
Keywords: Cadmium, lead, *Lemna gibba*, phosphate, nitrates, phytoremediation.

EBP7: Influence of nanoparticles in the toxicity of pesticides in the Mediterranean mussels (*Mytilus galloprovincialis*)

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Background and aim: Anthropogenic activity can reduce water quality and cause serious harm to aquatic organisms. This can ultimately lead to a decrease in natural resources. Among the most commonly contaminants, pesticides are on the list of substances that have been transferred to the aquatic environment. Additionally, in the last decades, nanoparticles have been largely used in different sectors. These chemicals could also reach marine ecosystem after their use. Little is known about interaction between nanoparticles and pesticides in aquatic system. The aim of this work is to evaluate the influence of AuTiO2 nanoparticles in the toxicity of permethrin on biological model *Mytilus galloprovincialis.*
Methods: The effect of AuTiO2 nanoparticles and permethrin in individual or in combination was investigated by measuring capacity filtration rate and biomarkers responses of mussels exposed to environmental concentration of contaminants.
Results: Permethrin (Per) contamination decrease filtration rate after of mussels and no significant effects ($p > 0.05$) was observed after exposure to AuTiO2 NPs. In addition, compared to control group, no effect was observed in filtration rate after exposure to binary mixture of Per and AuTiO2 NPs showing an antagonist interaction. Catalase activity (CAT) increased in both gills and digestive gland after exposure to Per and AuTiO2 NPs and effects was more pronounced in the digestive gland than gills. Malondialdehyde (MDA) level increased also in both organs after both treatments in organ dependent-manner. The mixture of Per and AuTiO2 NPs induced additive effect in considered organs showing the capacity of NPs to increase the toxicity of pesticides.
Conclusion: oxidative stress and physiological alteration can occur after nanoparticles and pesticides contamination on marine invertebrates. Digestive gland can be used as the target organ of permethrin contamination. In addition, additive effect can occur between nanoparticles and pesticides on marine organisms.Furthermore, contaminants interactions are a complex phenomenon that requires more research to better understand their impact on the biological components of the ecosystem.
Keywords: *Mytilus galloprovincialis*, Biomarkers, Pesticides, Nanoparticles, Biomonitoring

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EBP8: Single and combined effect of Cadmium and UV-C rays on mussels

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Background and aim: Intensive anthropogenic activity along the coast represents the major source of contaminants that can be potentially harmful for the health of aquatic organisms. Besides that, solar UV radiation, and other drivers of global change are undergoing significant changes and models forecast that these changes will continue for the remainder of this century and will eventually affect marine life .Taken in total, it is clear that the future changes in UV radiation coupled with anthropogenic pollution are expected to cause significant impacts on the marine organisms. The aim of this work is to investigate the hazardous effect of UV-C and Cdon mussels.
Methods: Mediterranean mus sels *"Mytilus Galloprovincialis"* were exposed at low and high concentrations of Cd (10µg/L ,100µg/L) for 7 days with and without UV-C rays.Single and combined effect of UV-C and Cd were evaluated by the measurement of biomarkers (CAT,Ache, and MDA).
Results: This experiment showed that all biomarkers'activities were stimulated in gills and digestive gland in a concentration dependent manner.
Conclusion: From the overall results of our study, it appears that mussels is sensitive to the proposed contaminants and can be considered as a bioindicator for this type of contamination. Thus, and even if its present situation is not very alarming precautionary measures must be taken to preserve our natural resources.
Keywords: *UV-Cradiations, Cadmium, Mytilus galloprovincialis, Biomarkers*.

EBP9: Phenotypic and genotypic characterization of the symbiotic rhizobia of the spontaneous legume *Medicago littoralis* of the palm groves of Oued Righ, Touggourt region, Algeria

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Background and aim: Nitrogen-fixing bacteria or rhizobia have low persistence in arid soils due to the salinity and high temperature that characterize the Saharan regions. These major parameters affect the establishment and effectiveness of rhizobia-legume symbiosis. The aim of our work is the isolation and characterization of rhizobia in the arid soils of the palm groves of the Touggourt region of south-eastern Algeria, a spontaneous legume species chosen for this study: *Medicago littoralis*.
Methods: The isolation from the nodules allowed us to purify 15 isolates. They were tested for their ability to nodulate their host plants, the tolerance to salinity (1, 2, 3, 4.5 and 6% of NaCl) and pH (4.5, 5, 6.9, 8 and 9), as well as the sequencing of 16S rRNA genes of some strains.
Results: All isolates showed a high ability to nodulate their host plants with an average of 15 nodules / plant.
Culture of isolates on YMA supplemented with different concentrations of NaCl showed that the majority of isolates supported concentrations up to 3% of salt.
The study of the tolerance of the isolates with the extreme pH showedthat the tested isolates can tolerate a pH range between 4.5 and 9.
The results of the 16S DNA sequencing of three isolates (MD05, MD09 and MD12) showed that the isolates belonging to the group of *Sinorhizobium (Ensifer) meliloti*.
Conclusion: The isolates of *Medicago littoralis*from the palm groves soil of Oued Righ in Algeria were fast-growing rhizobiabelonging to the group of *Sinorhizobium meliloti*. They can persist in arid environment and establish an effective nitrogen-fixing symbiosis.
Keywords: Nitrogen fixation, rhizobia, *Medicago littoralis*, soil ofpalm groves

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EBP10: The "Seybouse", one of the largest rivers in Algeria threatened by various pollutions

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Background and aim: Wadi Seybouse is one of the largest rivers in all of Algeria. It stretches for 225 km and crosses 7 wilayas. 66% of the water of this wadi is intended for the food of more than 1.5 million inhabitants, 8% for the food of the more than 71 factories and 26% for the irrigation of the lands considered as the most fertile. Alongside the 68 communes bordering this wadi, used industrial oils, waste and very dangerous chemical products of all kinds are dumped. It constitutes a particular threat for this ecosystem.
The present work is a contribution to the study of pollution of "oued Seybouse". It was concerned by the final sequence of the river from El Hadjar until Sidi Salem, passing by the upstream of the wadi mouth into the sea.
Methods: The study aimed to evaluate some parameters indicating water pollution during the years 2017 et 2018. As the study is still ongoing, we present in this communication the first results of the period between winter and spring 2017. The temperature, pH, EC, rH, turbidity and instantaneous flow where measured *in situ* in four sampling stations. In addition, nitrate, nitrite, ammonium, phosphate and chlorophyll-a, were analyzed by spectrophotometry.
Results: *In situ* measurements showed a maximum water temperature of 30°C and a neuter to slightly alkaline pH. The EC exceeding 1000µS/cm referred to a highly mineralized water and the rH levels reported reducing mediums. The water of the valley was confirmed turbid (especially in station 2) but low loaded in suspended solids. It streams at a very low speed and a variable instantaneous flow, with a negative gradient from upstream to downstream. Moreover, results ofthe chemical analysis and the chlorophyll-a revealed an eutrophic state of the studied final sequence of Seybouse wadi.
Conclusion: These results denote an alarming situation of the wadi during winter and spring 2017 and incite to take urgent dispositions to fix the pollution problems of this important river.
Keywords: Hydrological parameters, physico-chemical parameters, pollution, Seybouse wadi, Northeast Algeria

EBP11: Spent coffee ground a promising soil amendment: A field study

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Background and aim: The sustainability of production is a major challenge to agriculture. This is especially based on the organic soil fertility which mainly depends on its richness in organic matter. To improve the fertility there is recourse to the use of organic amendments, among other household waste. Hence this project focuses on the valuation of spent coffee grounds as organic amendment. This will improve soil fertility, productivity and sustainable waste management.
Methods: We have studied under field conditions the evolution of the physicochemical parameters of the soil following the application of various organic amendments: manure, compost, olive mill wastewater and spent coffee grounds. The work was done in the field with a random block device. The follow-up was carried out during 52 days with 13 periodic samples
Results: We followed the evolution of some physicochemical parameters namely pH, electrical conductivity, soil respiration, mineral nitrogen content and organic carbon content. The application of organic matter into the soil caused changes from the outset and the soil returned to equilibrium around day 14.We have found that compared to other amendments the spent coffee grounds appear to have effects which give it the character of a good organic soil amendment as it has allowed an improvement of soil respiration, improved electrical conductivity and provided significant levels of organic carbon.
Conclusion: In perspective, we plan to further extend the duration of treatment, to deepen our study by measuring other parameters like the levels of assimilable ions. We also plan to study the evolution and diversity ofmicrobial communities.
Keywords: Spent ground coffee, soil, characterization, amendment

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EBP12: Greener and eco-friendlier approach to leather processing using two keratinases from *Bacilus amylolequefaciens* S13

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Background and aim: The use of biotechnology by leather processing has increased in recent years. The feathers produced in large quantities are probably the biggest abundant source of keratinous materials; therefore, they constitute a real waste problem. Currently, incineration represents the main way to remove the feather waste. Every year, millions of tons of keratinous wastes are generated worldwide. Keratinases have long been used as alternatives to chemicals to improve the efficiency and cost-effectiveness of a wide range of industrial systems and processes. Enzymes can be applied during different steps of the leather production process: soaking, dehairing, bating, dyeing, degreasing or in effluent and solid waste treatment.
Methods: The purification and biochemical characterization of the pure keratinases KERZTA and B from *Bacillus Amylolequefaciens* strain S1 3 were investigated though physicochemical determination as well as spectroscopy analysis.
Results: The current paper reports the purification and biochemical characterization of two keratinolytic enzymes, with moderate elastolytic activity, from *Bacillus amyloliquefaciens* strain S13 newly isolated from the brown alga *Zonaria tournefortii*. The enzymes were purified to homogeneity by precipitation with (NH4)2SO4-dialysis, followed by size exclusion HPLC column, and submitted to biochemical characterization assays. The findings revealed that the pure enzymes designated KERZT-A and B were monomers with molecular masses of 28 and 47 kDa, respectively. Their identified NH2-terminal amino acid displayed high homologies with those of *Bacillus* keratinases. While KERZT-A was optimally active at pH 6.5 and 50 °C, KERZT-B showed optimum activity at pH 8 and 60 °C. Both enzymes were completely inhibited by PMSF and DFP, which suggests their belonging to the serine keratinases family. Interestingly, KERZT-A displayed higher levels of hydrolysis, substrate specificity, and catalytic efficiency than KERUS from *Brevibacillus brevis* strain US575, NUE 12 MG (commercial enzyme), and KERZT-B unhairing keratinases.
Conclusion: The enzymes exhibited powerful keratinolytic activity that made it able to accomplish the entire feather-biodegradation process on its own. Overall, the findings indicated that KERZT-A and B enzymes seems to be an effective and an eco-friendly alternative to the conventional chemicals used for the feather keratin-biodegradation and for the unhairing of hides or skins in the leather processing industry.
Keywords: Eco-friendly approach; Keratinases; *Bacillus Amylolequefaciens; Zonaria tournefortii.*

EBP13: Influence of the toxicity of titanate of barium strontium in the Mediterranean mussels (*Ruditapes decussatus*)

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Background and aim: Anthropogenic activity can reduce water quality and cause serious harm to aquatic organisms. Little is known about interaction between combination metals in aquatic system. The oxides ferroelectric is potentially significant materials for a variety of devices. However, it contains various types of polluting matters like lead, mercury, cadmium and chromium. This work aims to describe a new simple approach for the synthesis of new oxides ferroelectric, the titanate of barium strontium (BST), by reaction in a solid state. For a sure and effective use of the BST in the electronic field, it is critical to include/understand how this material can interact with a biological component. The aim of this work is to evaluate the influence of titanate of barium strontium on biological model *Ruditapes decussatus.*
Methods: The effect of the titanate of barium strontium (BST) was investigated by measuring capacity filtration rate and biomarkers responses of mussels exposed to environmental concentration of contaminants.
Results: The predictions of such an effect are necessary before their use in several applications. On this subject, the present study must underline the possible interaction between BST and the marine organisms. For this purpose, the clams were exposed to various concentrations of BST ([BST] 1 = 0,001 Mg / L, [BST] 2 = 0,01 Mg / L, [BST] 3 = 0,1 Mg / L and [BST] 4 = 1 Mg / L). The results relating to the capacity of filtration and the oxidative biomarker (SOD, CAT) shows that the BST can affect the health of the clam in organ and concentration dependent manner. In the same way, the AChE activity showed that the threshold of disturbance of the cholinergic system is between 0, 01 and 0,1 mg/l of BST.
Conclusion: These results show the level of sensitivity of the clams providing a framework for the comprehension of the toxicological effects of oxides ferroelectric on the filter feeding organisms. Moreover, at low concentrations, the BST represents a good alternative replacing oxides ferroelectric containing lead and could be regarded as a durable candidate in various electronic applications while drawing left the nontoxic level for the environment.
Keywords*:* BST; Ferroelectrics Oxides; Electronics; Biomarkers; biomonitoring

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EBP14: Effects of cadmium on the Mediterranean meiofauna and especially on marine free nematodes

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Background and aim: Anthropogenic activity can reduce water quality and cause serious harm to aquatic organisms. This can ultimately lead to a decrease in natural resources. Among the most commonly contaminants, Heavy metals are on the list of substances that have been transferred to the aquatic environment and can cause health problems. The aim of this work is to evaluate the influence of Cadmium on biological model meiofauna and particularly on marine free nematodes.
Methods: The ecotoxicological effect of cadmium on the meiofauna particularly nematodes in terms of density and diversity.
Results: This work aims to study the individual ecotoxicological effects of two concentrations of Cadmium (C1 = 5 mg/300g and C2 = 10 mg/300g).The quantitative and quantitative studies showed that the individual contamination by Cd, is toxic on the meiofauna and particularly on the free-living nematodes. Based on quantitative and qualitative data of free living nematodes; Oncholaimus campylocercoides,Anticoma acuminate and Marylynnia puncticaudata could be considered as " Cadmium sensitive species ". However, Terschellingia communis is "opportunist" specie, whereas Daptonema tuberculosum, Paramonhystera faber, Terschellingia longicaudata and Metalinhomoeus numidicus could be considered as "Cadmium resistant species ".
Conclusion: This study showed that the meiofauna and in particularly the free living nematodes can be used in the biomonitoring programs of lagoons against metal pollution. These results showed that the individual contamination by Cd, is toxic on the meiofauna and particularly on the free-living nematodes. The different meiofaunistic taxa were differently affected by the two doses of cadimium as well as the diversity of nematodes.
Keywords: Cadmium, bioindicators, meiofauna, free living nematodes.

EBP15: New Efficient Laccase Immobilization Strategy Using Ionic Liquids for Biocatalysis and Microbial Fuel Cells Applications

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Background and aim: Laccases belong to the family of blue multicopper oxidases, which catalyze the four-electron reduction of dioxygen to water concomitantly through the oxidation of phenolic and other aromatic compounds. They are potential enzymes in many applications including biofuel cells to produce electricity through chemical reactions. Several laccase immobilization methods have been elucidated (such as inclusion, covalent link, entrapment, etc) but never the current strategy.
Methods: In the present work, laccase from *Trametes versicolor* has been successfully immobilized for the first time by casting into polymer inclusion membrane (PIM) based on ionic liquids and polyvinylchloride. The immobilization rate was very high in all the cases (99.2 ± 0.6%), then enzyme entrapment strategy can be considered as successful and one from the best used for laccases immobilization up till now. The assayed membranes has been evaluated as performing carrier for laccase by the reaction of ABTS (2,2′-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt) oxidation.
Results: Four screened Ionic Liquids (ILs) has been novelty used for this purpose: [OMIM][NTf_2], [EMIM][NTf_2], [Epy][NTf_2] and [Chol][NTf_2]. We have demonstrated its efficiency regarding the activity rate and the stability of the enzyme versus ILs concentrations. The most suitable ILs regarding activity was the [Chol][NTf_2]. However, the reuse assays showed that the most stable PILM has been the one constituted by 50% [Epy][NTf_2]. For physical resistance and water insolubility, [OMIM][NTf_2] PILM has been selected for the Microbial Fuel Cell (MFC) application.
Conclusion: This is the first report using successfully casting method for laccase immobilization into PILM. The application of new PILM in MFC showed high stability of the laccase immobilized membrane during almost one week and its efficiency to produce bio-energy and remove COD from industrial wastewater.
Keywords: Microbial fuel cell; immobilization; Laccase; Ionic liquids; Polymer inclusion membrane.

EBP16: Genetic Type Relationship with Mortality of Young Rabbits of Algerian Local Population

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Background and aim: In Algeria, rabbit rearing is mainly based on the use of a local rabbit population which needs a better knowledge of its biological politissibies and its adaptability to the rearing conditions as well. Various studies were conducted in the goal to characterize and preserve the genetic wealth. The aim of the present work was to study the relations between the phenotype (limited to the fur coat color) and d the mortality of young rabbits during the birth to weaning period.
Methods: Rabbits were reared in wire mesh cages placed in a building with natural lighting and absence oftemperature regulation.
This study includes a large number of observations: 6254 kits of 896 litters of 209 females in a closed population for a period of four years. The rabbits followed in this study belong to the local population and present two phenotypes: albino and colored. The kits are counted and weighed twice a week for 30 days.
Results: The colored females had a significant higher prolificacy at birth (P <0,01). The prolificacy of the colored rabbits is greater than that of the white females with (P = 0.011) at birth and (P = 0.0019)at weaning. The average value of the mortality rate from birth to weaning is around 3 6.84%. The effect of the phenotype is significant (P = 0.0339).Concerning the phenotype of kits, the results obtained at weaning from the different parental crosses according to the phenotype recall the Mendelian proportions of F1. In litters from a colored female and a white male, 49.7% of white rabbits and 50.3% of colored rabbits were recorded. The litters resulting from the crossing of males and females all white have the phenotype white 100%.
Conclusion: At the end, it could be concluded that within the local population, the "albino" females were less prolific than the colored ones (the mortality of kits is more important). This work suggests that it may be worthwhile to develop breeding programs to provide new and better performing rabbits.
Keywords: mortality, genetique, coat color, kits

EBP17: Purification and characterization of a novel manganese peroxidase from the white-rot fungi *Irpex lacteus* strain SBT16 for lignin degradation

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Background and aim: Peroxidases [E.C. 1.11.1 .x; donor: $_{H2O2}$ oxidoreductase] require H_2O_2 or other peroxides to oxidize various reducing substrates, which confers them an active role in different biological processes. Peroxidases, which are encoded by small or large multigenic families, are involved in several important physiological and developmental processes. They use various peroxides as electron acceptors to catalyse a number of oxidative reactions and are present in almost all living organisms. We have created a peroxidase database (http://peroxibase.isb-sib.ch) that contains all identified peroxidase-encoding sequences (about 6000 sequences in 940 organisms). They are distributed between 11 superfamilies and about 60 subfamilies.
Methods: The biochemical and biochemical characterization of the pure peroxidases were investigated though physico-chemical determination as well as spectroscopy analysis.
Results: A novel extracellular manganese peroxidase-producing (75 U/mL) white-rot fungus was isolated from was isolated from symptomatic wood at the Hamma Botanical Garden (Algeria) and identified as *Irpex lacteus* strain BT16. The pure enzyme (MnP IL45) was purified to apparent electrophoretic homogeneity and biochemically characterized. The specific activity and Reinheitzahl value ofthe purified MnP IL45 were 632.58 U/mg and 2.7 1, respectively. MALDI-TOF/MS analysis revealed that the purified enzyme was a monomer with a molecular mass of 45,211.10 Da. The sequence of its 21 NH2-terminal amino acid residues showed high homology with those ofwhite-rot fungi peroxidases. MnP IL45 showed optimal activity at pH 6 and 80 °C using 2,6-DMP. This peroxidase was completely inhibited by sodium azide and potassium cyanide, suggesting the presence of heme-components in its tertiary structure. Interestingly, MnP IL45 showed higher catalytic efficiency of 5.2; 4.3; 2.5, and 1.7 fold than MnP TP55, MnP BA30, HRP, and MnP PC, respectively.
Conclusion: Data suggest that MnP IL45 may be considered as potential candidate for its useful tool for environmental applications mainly the development of enzyme-based technologies for lignin degradation.
Keywords: White-Root Fungi, Manganese Peroxidases, Lignin Degradation.

EBP18: Combined effect of *Carbamazepine and Clarithromycin* on the cockle *Cerastoderma edule*

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Background and aim: Pharmaceuticals are used throughout the world being not only applied in human medicine but also in livestock breeding and aquaculture and they are not completely metabolized by the organism. Among pharmaceutical drugs commonly detected in the aquatic environment,carbamazepine (CBZ) and clarithromycin (CLR).The objective of this study is to evaluate the toxicity of the simultaneous exposition to these two emergent pollutants.
Methods: The cockle *Cerastoderma edule* collected from Ghar El Melh lagoon (Tunisia), was used as a biological model to assess the toxicity of this pollutant. Bivalves were exposed to three nominal mixture concentrations (5+5, 10+10, 20 +20 µg / L) of CBZ and CLR for two periods 48 and 96 hours.
Results: After 48h of exposure, a significant change was observed in the concentrations of (H_2O_2) with C1, C2and C3 of mixture compared with the controls. After 96h the H_2O_2 level increased non-significantly by comparing it with the controls 0.122 ± 0.019 nmol / mg prot.SOD activity, decrease significantly compared to controls with C1 (0.093 ± 0.0 10 U SOD / min / mg prot) to reach (0.079 ± 0.007 U SOD / min / mg prot) with C3. After 96 h it decreased significantly by 53.06% with the lowest concentration. The GSTactivity decreased significantly by 36.14%with C1 (0.126 ± 0.007 nmol / min / mg prot) and 57.83% with C3 (0.083 ± 0.003 nmol / min / mg prot). After 96 h it recorded a decrease, it was significant with C1by 43.27% (0.108 ± 0.01 nmol / min / mg prot) compared to the controls (0.191 ± 0.008 nmol / min / mg prot).After 48 h of exposure to the mixture, a highly significant inhibition of AChE activity by 77.94% was observed for the mixture compared to the controls (0.016 ± 0.002 nmol / min / mg prot). The same result was observed after 96 hours ofexposure.
Conclusion: Carbamazepine and clarythromycine are two persistent molecules. Their combined effect impaired the health status and induce oxidative stress to *Cerastoderma edule*.
Keywords: toxicity, *Cerastoderma edule*, carbamazepine, clarithromycin.

EBP19: Study of the biosorption of an industrial dye (Crystal Violet) by sludge from urban water treatment

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Background and aim: Many industrial wastes (plastics, cosmetics, paper, and especially textiles) are heavily laden with dyes which cause aesthetic problem but also dangerous toxicological impacts to the environment. Crystal violet is a cationic dye widely used in painting, printing, and in veterinary drugs .... However, this dye has been reported to be toxic and remains in the environment for a long time because of its stable and non-biodegradable structure. Various physical, chemical and biological techniques have been developed and tested for the treatment of dyed effluents; adsorption is one of the easy-to-implement technologies and is widely used for water treatment.
The main objective of our work is to evaluate the adsorption capacity of sewage sludge from the crystal violet dye.
Methods: This study is carried out by a raw and treated sludge, examining the effect of several factors on the bleaching power of material used, in particular, temperature, contact time, adsorption mass and pH. The results obtained will be processed by applying some models of equilibrium and kinetics. The characterization is done by IRTF and XDR spectroscopy.
Results: According to the variation of the final pH as a function of the initial pH, it is found that the sludge possesses a certain buffering capacity. The trace of the adsorption isotherms shows that the langmuir (L) and Redlich-Peterson models represent perfectly with a maximum adsorption capacity of about191 mg/g. The biosorption kinetics of the dye follows perfectly the pseudo-second order model with R^2=0,98 and ERM= 0,35% and intraparticular diffusion with R^2= 0,99 and ERM= 1,304%. The thermodynamic parameters obtained indicate that the adsorption of CV is a spontaneous and exitherlic physisorption process.
Conclusion: the sludge can be considered as a major biosorbent with an elimination rate of CV of the order of 95,22% and therefore this material can be used in the treatment of industrial wastewater loaded with dyes.
Keywords: Sewage sludge, hydrochloric acid, The Crystal Violet, Biosorption.

EBP20: Production, partial purification, characterization, and application as laundry detergent additive of an alkalophilic protease from *Bacillus velezensis* strain F35

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Background and aim: Proteases represents one of the major groups of industrial enzymes and a number of detergent stable proteases have been isolated and characterized because of its widespread use in detergents. It is worthwhile to screen microbes from new habitats for proteases with novel properties to meet the needs of rapidly growing detergent industry. High-alkaline serine proteases have been successfully applied as protein degrading components of detergent formulations and are subject to extensive protein engineering efforts to improve their stability and performance. Protein engineering has been extremely used to study the structure-function relationship in proteases and led to deeper understanding of the factors influencing the cleaning performance of detergent proteases. The aim of this work is to produce an alkaline protease having characteristics allowing its potential incorporation and application in the detergence industry.
Methods: Enzyme purification was conducted using the sulfate ammonium precipitation. The amplifications of the ARNr 1 6S and the gene encoding for the studied protease were performed by PCR.
Results: A collection of 88 strains was subjected to the screening of proteolytic activity in solid medium. Thirty-six strains showed a halo of inhibition which correspond to the protease activity. The strain F-35, isolated from wastewater at the EJM Company, which exhibited the largest halo of inhibition, was retained for further studies and assigned as *Bacillus velezensis* based on physiological and biochemical properties and 1 6S rRNA gene sequencing. The study of the effect source of carbon and azote on the activity of the studied protease showed that the best activity of 7500 U/ml was obtained by combining the Modilac (powdered milk) and the yeast extract. The characterization of the physico-chemical properties showed that the partial protease has an optimum activity at 60 °C and pH 10. The partial enzyme exhibited excellent stability and compatibility with surfactants and commercial detergents, revealing 95% stability with 2% LAS and 100% stability with Class and Arial commercial laundry detergents. The gene encoding for the studied protease was amplified by PCR and showed a size of 1.5 kb.
Conclusion: Accordingly, such a protease could be considered as a good detergent-additive in detergent industry.
Keywords: *Bacillus velezensis*, alkaline protease, detergence formulations.

EBP21: Response of benthic prokaryotes to anthropogenic contamination by total petroleum hydrocarbons in sediment of Bizerte lagoon

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Background and aim: Petroleum hydrocarbons are ubiquitous pollutants in marine sediment as a consequence of industrial discharges, accidental spills, shipping activities, and marine oil and gas explorations, causing severe risk to marine ecosystems. The aim of this work is to determine the concentration of petroleum hydrocarbons and its impact on response of benthic prokaryotic communities.
Methods: TPH concentration with GC/MS analysis and ARISA data ofbacterial communities were investigated in Bizerte lagoon sediments during two sampling period (August 2011 and February 2012).
Results: The sediment TPH concentrations were significantly different in terms of both temporal and spatial variations (p 0.0 1). The temporal variations in TPH content showed the lowest values in August 2011, ranged from 0ppm and 352.3ppm with an average of 85.5ppm. But, it was significantly increased in February 2012 (118 - 1 160.7ppm) with an average of 409.7ppm. The highest levels were detected at the level of the ships 'journey (channel and in front of Socomena). The mean of prokaryotic density showed important spatio-temporal fluctuations, ranging across all stations, between $1.03\ 10^9 \pm 6.02\ 10^8$ cell g^{-1} dry sed. (August 2011) and $2.26\ 10^9 \pm 1.3\ 10^9$ cell g^{-1} dry sed. (February 2012). ARISA analysis revealed that bacterial communities' composition is influenced by TPH. 161 OTUs were identified in August 2011, among them 400, 451 and 448bp showed the highest abundances and contributed respectively for 19.71%, 6.50% and 5.69% whereas reduced its importance in February 2012 respectively for 5.50%, 0.87% and 4.54%. Only 156 bacterial OTUs were identified in February 2012.
Conclusion: the result of the present study indicates that TPH has a significant impact on the microbial components, suggests that anthropogenic contamination by TPH might be responsible for the potential risk of adverse biological effects along the coastal ecosystems represents a priority for conservation.
Keywords: Benthic prokaryotes, ARISA, Bacterial OTUs, Total Petroleum Hydrocarbons, Bizerte lagoon.

EBP22: Improvement of textile wastewater treatment plant performance for reducing environmental impact of produced sludge

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Background and aim: Textile wastewater treatment using activated sludge process (ASP) is a very effective solution. However, such a process has a lot of drawbacks especially on the environment. An investigation of an industrial textile wastewater treatment plant (KsarHellal, Tunisia) was carried out. Results showed that the plant produces high amount of sludge containing 304.5 kg of adsorbed dye per day. An improvement of dye biodegradation to decrease ASP environmental impact was performed.
Methods: A multi-scale study was performed to determine the best operating conditions for a maximum dye biodegradation.
Results: The lab-scale batch cultures helped to determine dyes degradation specific kinetic and stoichiometric parameters. The Y_{Hind}, μ_{Hind} and K_{Sind} were in the range of 0.55-0.7 g cell COD formed/ g COD_S oxidized, 2.85-3.1 1/d and 20.07-40.01 g/m^3, respectively. These values were used to establish a model for dyes biodegradation calibrated on indigo dye biodegradation. The best substrate/biomass ratio selected in batch biodegradation tests was applied at a pilot scale with different operating conditions. Several hydraulic retention times (HRT) and sludge recycling rates (SRR) were tested. The best selected operating conditions were 5 days and 0.22 for the HRT and SRR, respectively. Under the best working conditions, the amount of adsorbed dye on the waste sludge was reduced from 304.5 kg/d to 33kg/d.
Conclusion: ASP proved to be a very efficient process even with a very toxic wastewater. Working with both Numerical and experimental tools helped to determine the ultimate working conditions to have a well-adapted activated sludge able to biodegrade complex dye molecules.
Keywords: Textile wastewater, sludge, adsorption, biodegradation, toxicity.

EBP23: Climatic factors effects on the morpho-physiological behavior of some durum wheat varieties (*Triticum durum* Desf.) in the eastern high plateaus

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Background and aim: This work was conducted at the agricultural experimental station of the technical field institute (ITGC) of Sétif during the 2016/17 crop year. This study, through the use of some morpho-physiological tests, aims to evaluate the selection of durum wheat varieties (*Triticum durum* Desf.) That can tolerate water stress under the eastern high plateaus conditions.
Methods: The tests were conducted in a randomized block design with 4 repetitions; the elementary plot is 6 rows x 5m long x 0, 20 m interrow spacing. The ratings and measurements were carried out by elementary plot and focused on the determination of aboveground biomass (BIO, g/m^2), harvest or seed yield (SY, g/m^2, leaf area (cm^2), relative content water (RCW %), cell integrity (CI %), and canopy temperature (CT°) or highest cover vegetation (HCV) temperature.
Results: The results show that the variety named (V2) exhibits a greater reduction in biomass (BIO) and harvest or seed yield (SY) compared to the variety named (V3) and the control, while the highest mean Chlorophyll (Ch) and cell integrity (CI %) values are found in the variety (V2), and the highest cover vegetation (HCV) temperature is noted in the control with little deference between the two varieties, however, the leafarea is reduced in all varieties.
Conclusion: The study carried out showed a very pronounced unequal effect of eastern high plateaus factors such as drought stress on the morphological and physiological behavior of different varieties.
Keywords: Durum wheat, variety, Climatic factors, morpho-physiological behavior, eastern high plateaus.

EBP24: Larvicide activity of essential oils of *Artemisia Herba-Alba* and *Artememisia Absinthium* on the larves of *Culex pipiens*

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Background and aim: The massive application of insecticides has several drawbacks, essential oils seem to be a better alternative for biological control. In this context, our work aims to test the larvicide activity of essential oils of two species of *Artemisia* on the fourth larval stage of mosquitoes of genus *Culex*, vectors of parasitic diseases for humans.
Methods: Four prepared concentrations of the essential oils of *Artemisia Herba-Alba* and A. *Absinthium* (S1 = 1%, S2 = 2%, S3 = 5% and S4 = 10%) were tested. 3replications were performed for each concentration as well as for the witness. The number of dead individuals was calculated for each concentration and for each replication after 1 hour, 2 hours, 4 hours, 20 hours, 22 hours and 24 hours. Mortality and LC50 lethal concentrations were determined and compared.
Results: The toxicity test showed that the larvicidal activity of the essential oils depends on the plant species, the concentration and the duration of exposure. Indeed, after 24h of exposure, the essential oil of *A. Herba-Alba* showed a high mortality rate of 63.33 and 73.33% for S3 and S4 respectively. The larvicide effect of the essential oil of *A. Absinthium*, was important with mortality rate of 63.67% for S3 and 66.67% for S4. The toxic effect of essential oils was also highlighted by the LC50 values which were in the order of 0.1% for *A.Herba-Alba* and 0.6% for *A. Absinthium*.
Conclusion: Results have shown also a good larvicidal activity of the essential oils of the twospecies studied on stage 4 larvae of *Culex pipiens*. Particularly, *Artemisia Herba-Alba* exhibited better larvicide toxicity. It will be advisable to complete this work by studying the larvicide activities of aqueous extracts and floral water against larvae of *Culex pipiens*.
Keywords: Larvicide activity, *Artemisia Herba-Alba* and *Artemisia Absinthium*, *Culex pipiens*.

EBP25: Bacteriophages of *Erwinia amylovora* from Tunisia:potential application in biological control of fire blight

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Background and aim: Fire blight is a complex disease where the infective agent is continually associated with plant tissue. In Tunisia, fire blight symptoms was observed for the first time in the spring of 2012 in pear trees and has spread rapidly through the most important pear growing regions causing the destruction of several hundred hectares of pear plantations in tunisia. Several reviews have been previously published, highlighting the possibilities and limitations ofphage therapy in plant disease control.
Methods: Bacteriophage isolates were distributed on UBA top agar layers supplemented with 1 % (w/v) sucrose and containing the test bacterium (*E. amylovora*). The thermostability of the phages was observed at different temperatures of 50 ° C to 100 ° C as a function of time.
Results: Twenty bacteriophages isolates of *Erwinia amylovora*, the causal agent of fire blight, were isolated from blighted apple and pear trees from different sites in Tunisia (Manouba, Tebourba). The majority of the phages in the collection were isolated from the soil surrounding trees exhibiting fire blight symptoms. Our phage isolates formed plaques of different sizes, with a diameter of 0.5–2 mm on the soft agar layer containing the test bacterium. The phages thermostability tests have showed that all these viruses resist at 80 ° C for 45 min.
Conclusion: Some of the bacteriophages withstand a heat treatment of 100 ° C. So, we have decided to continue the identification and classification of these phages and the study of their mecanisms of persistence in the environment**.**
Keywords: *Erwinia amylovora*, bacteriophages, plant desease, thermostability resistance.

EBP26: Antioxidant capacity of *Oxalis pes-caprae* aerial part methanolic extract

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Background and aim: Scientific studies of natural products as an interesting source of new therapeutics used in ethnic medicine have led to the discovery of many valuable drugs especially for cancer therapy. In folk prescription Oxalis is widely used to cure diarrhea, sorethroat, in clinical practice oxalis has significant effects on curing inflammation. The evaluation of antioxidant activity of *Oxalis pes-caprae* aerial part methanolic extract was the main aim ofthe present study.
Methods: *In vitro* antioxidant activity was evaluated by ABTS radical-scavenging activity (Re *et* al, 1999), ferrous ion chelating assay emploing (Decker et Welch, 1990) and Bcarotene test according to (Barrier et al.,2001 ;Aslan et al., 2006)
Results: The results obtained showed that the methanolic extract exhibit a scavenging activity against ABTS radical with $_{EC50}$ values of 0.01± 0.00 mg/ml, the ferrous ion chelating assay of the extract had an $_{IC50}$ of 0.16±0.00 mg/ml while the inhibition in Betacarotene was 8 8.44%. Polyphenols are derived from natural food product, they are often considered safer and more easily integrated into life style changes than convential pharmaceutical drugs, however, the antioxidant activity does not depend solely on phenolic content but also on other phytoconstituants such as triterpenoids, or the combined effect of them.
Conclusion: Overall, these findings provide scientific basis to the traditional use ofthis plant as source of natural anti-oxidant compounds, and anti-inflammatory remedy based on their crucial antioxidant properties, however, further studies need to be carried out to unveil the structure-activity relationship and to estimate the effectiveness of the substances.
Keywords: Antioxidants, *Oxalis pes-caprae, ABTS* scavenging activity, ferrous ion chelating, Beta-carotene.

EBP27: Development of an innovative new environmentally-friendly approach for enzymatic soaking, unhairing-liming, and degreasing of sheep skins and hides

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Background and aim: Leather making is an important socio-economic activity for several countries throughout the world. Leather processing involves a complex set of steps, from skin to finished product, including soaking, dehairing, bating, tanning, and degreasing. The conventional methods of leather processing involve the application of various hazardous chemicals and organic solvents, which generates several environmental and waste disposal problems. In order to overcome the hazards caused by these effluents, enzymes have often been proposed as viable alternatives. In fact, enzymes have long been used as alternatives to chemicals to improve the efficiency and cost-effectiveness of a wide range of industrial systems and processes.
Methods: The biochemical characterization of the target hydrolytic enzymes (subtilisins, keratinases, and lipases) were investigated though physico-chemical and kinetic determination as well as spectroscopy analysis.
Results: Considering the attractive properties of microbial enzymes and the promising opportunities that they might open for the development of efficient and eco-friendly leather manufacturing processes, the present national project aimed to the production, purification, and biochemical characterization of proteases and lipases isolated from Tunisian strains for leather interest. Two extracellular keratinases: KERUS from *Brevibacillus brevis* US575 and KERQ from *Bacillus tequilensis* Q7 are potential candidates for soaking and dehairing-liming processes, respectively. KERUS displayed higher levels of hydrolysis, substrate specificity, and catalytic efficiency than NUE 12 MG and KOROPON® MK EG keratinases. KERQ7 displayed higher levels of hydrolysis and catalytic efficiency than Basozym® CS 10, Koropon® SC 5K, and Pyrase® 250 MP. *kerUS* and *kerQ7* genes encoding KERUS and KERQ7, respectively were cloned, sequenced, and expressed in *E. coli* BL21 (DE3)pLysS. For the enzymatic degreasing process, the potential lipases candidates are YLLIP2 from the yeast *Yarrowia lipolitica* and ROL from the filamentous fungus *Rhizopus oryzae* which they are particularly important because they exhibit very high specific activities and remarkable lipolysis rates on long chain triglycerides than the commercial lipolase™ from the fungus *Thermomyces lanuginosus*.
Conclusion: These properties make the first selected enzymes a potential promising and eco-friendly alternative to the conventional chemicals used for the soaking, dehairing-liming, and degreasing process of sheep skins and hides in the leather processing industry.
Keywords: Leather; Soaking; Dehairing-liming; Degreasing; Proteases; Keratinases; Lipases.

EBP28: The possibility of proatherogenic effects of permethrin mediated by arterial accumulation of native and oxidized LDL in rats: The beneficial role of vitamin E

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Background and aim: Nowadays, research findings have strengthened the link between exposure to pesticides and the increase risk of cardiovascular diseases. Therefore, the aim of the present study was to examine the effect of chronic exposure to permethrin, one of the effective types II synthetic pyrethroid, on plasmatic and arterial native and oxLDL levels, as well as arterial LDL receptors and scavenger receptors in rats, as predictors of atherosclerosis. In addition, the underlying molecular mechanisms of Vit E supplements as a mitigation strategy were also targeted.
Methods: Rats were treated by gastric gavage with Vit E (100 mg/kg/bw) in alone and cotreated groups for 90 days. Apart from control and Vit E groups, all the groups were subjected to permethrin (3 mg/kg, via gavage) toxicity.
Results: Results showed that the permethrin is able to cause long-term increase in plasmatic and arterial lipid parameters such as total cholesterol, low density lipoproteins (LDL) linked to cholesterol, apolipoprotein B 100. A dramatic increase in levels of oxidized LDL in plasma and aorta has also been demonstrated, compared to controls. Moreover, permethrin treatment significantly increased the plasmatic levels ofthe proinflammatory cytokines TNF-α, IL-2 and IL-6. In addition, the densitometric quantification of protein bands showed that the amount of hepatic native LDL-receptor protein decreased significantly in the intoxicated rats compared to the control group. Whereas, the expression of arterial LDL receptors (LDLR) and scavenger receptors (CD36) was amplified owing to permethrin toxicity. This harmful effect was confirmed by histological study using oil-red-o staining. Owing to their antioxidant effects, Vit E has restored all the changes in plasma and aorta lipids, and prevented the proatherogenic effect observed in permethrin -treated animals.
Conclusion: Our study showed that, chronic exposure to permethrin induces arterial accumulation of native and oxidized LDL in rats, a hallmark in early atherosclerosis. Vit E supplementation has been shown to ameliorate all the plasmatic and arterial lipid disturbances.
Keyword : Permethrin, vitamin E, western blot, native LDL, oxidized LDL

EBP29: Biodiversity of filamentous fungi isolated from Olive mill waste water and olive cake from different mills of Tunisia

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Background and aim: The increase in olive production and olive oil will generate much larger quantities of by-product that threaten the environment. Current trends should focus on the integration of various technologies to treat and valorize these effluents, but at a low cost. Therefore, it would be interesting to study the biodiversity of the filamentous fungi of olive by-products as well as their physiological and biochemical mechanisms. In this context, the aim of this work is to study biodiversity of filamentous fungi isolated from olive mill waste water and olive cake from different mills of Tunisia.
Methods: The study of biodiversity of filamentous fungi was investigated through the isolation, purification and identification of new strains; and the study of the physiological and biochemical mechanisms of the selected strains by describing the nutritional needs and the metabolic potentialities of these microorganisms with a view to selecting strains capable of producing enzymes.
Results: A total of 47 strains of filamentous fungi were isolated and purified from samples obtained from the OMW and olive cake of different mills of Tunisia. The isolates thus obtained mainly belonged to the genera *Aspergillus*, *Penicillium*, *Alternaria*, *Talaromyces*, *Geotrichum Cladosporium*, *Mucorale*, *Fusarium*, *Aureobasidium*, *Curvularia*, *Trichoderma*, *Purpureocillium*, *Rhizopus*, *Myceliophthora*. The percentage distribution of different species showed a predominance of *Aspergillus* (33%) and *Penicillium* (26%). A physiological characterization of the strain was performed by studying the apical growth rate and the sporulation index. From the results found, all isolated strains show relatively fast apical growth and produce abundant amounts of spores. A biochemical characterization of the strain showed that starch, sucrose, glucose, casein and carboxymethylcellulose have been used by all the strains. The low development of the strains was observed for the medium containing phytic acid and tannic acid.
Conclusion: The results obtained show that our fungi collection shows an important biotechnological potential, thereby the isolated strains can produce several extracellular enzymes of great interest for biotechnology and industry such as lipase, phytases and tannases which represent high added value products.
Keyword: Olive mill wastewater; Olive cake, Biodiversity, Fungi, Extracellular enzymes.

EBP30: Statistical optimization of production, and study of the stability for application in detergency of a new thermostable bacterial α-amylase

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Background and aim: In this study, a thermophilic and starch-degrading bacterial strain (named HB23), newly isolated from the thermal spring of Hammam Righa (Aïn Defla, Algeria), was the subject of a statistical optimization aimed to improve its a-amylase production. The potential use of the enzyme as a detergent additive has thereafter been investigated.
Methods: Screening of factors that significantly influenced the production was carried by the use of L27 Taguchi design. Then, the response surface methodology (RSM) using a BoxBehnken design allowed the study of the interactions between selected factors and the determination of their optimum levels. Analysis of the residual activity of the enzyme when pre-incubated with detergent additives and commercial detergents, allowed the assessment of its stability and compatibility as a detergent additive. A washing test was performed in order to evaluate enzymatic performance in removing chocolate stains on cotton fabrics.
Results: Optimization allowed reaching an activity of 320 U/mL, which represented an increase of 21.3 folds. Wheat bran (among soluble starch, tryptone, and inoculum size) was a factor that positively influenced production. Molecular identification of the bacterial strain showed that it was affiliated to *Tepidimonas fonticaldi*. Compared to the commercial a- amylase Termamyl®300L, the a-amylase HB23 crude extract showed outstanding stability towards detergent additives and remarkable compatibility with wide range of commercial detergents. Washing performance analysis revealed that the a-amylase HB23 could effectively improve the removal of chocolate stains when added to the iSiS detergent.
Conclusion: *Tepidimonasfonticaldi* HB23 strain turns out to be an efficient crude a-amylase producer when grown in adequate conditions. According to the stability, compatibility and washing tests results, a-amylase HB23 stands as a potential candidate in detergent formulations.
Keywords: Optimization, a-amylase, *Tepidimonasfonticaldi*, Detergent formulations.

EBP31: Enhancement of biogas production from food waste and cattle manure co- digestion

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Background and aim: In many countries great amounts of food wastes(FW) are currently land filled or incinerated. The world targets to reduce the environmental burdens and to create new resources of bioenergy. FW is indeed an untapped resource with great energetic potential. Anaerobic Digestion (AD) is the most suitable method for its valorization. The aim of this work is to evaluate its feasibility and to identify the key factors governing its performance.
Methods: Laboratory experiments of AD were performed in a continuous reactor and in batch reactors under mesophilic conditions. Effects of hydraulic retention time, organic loading rate, co-digestion of (FW) and cattle manure (CM) were examined in the continuous reactor. Effects of substrate pre-treatment were examined in the batch reactors.
Results: Results showed that co-digestion of CM+FW at different ratios improved biogas yield and methane yield. Comparing to mono-digestion, for FW: CM=2, biogas yield and methane yield increased by 169.8% and 154% respectively and by 169.1% and 64.4% for FW: CM=3. However, the best biogas yield and methane yield were produced by the liquid fraction of FW (LFW). For LFW: CM=2, it increased by 132.62% and 133.13% respectively comparing to raw FW (RFW) at the same OLR and the same ratio. Simultaneously, thermophysical pretreatments were evaluated to identify its effect on biogas production. RFW showed a better biogas production comparing to dried one at T=105°C with a reduced size of 1 0mm and 1 5mm respectively.
Conclusion: The high biodegradability of FW makes it a promising organic substrate for AD. However, mono-digestion of FW often leads to digester instability and failure at high organic loading rates also, some pretreatments should be applied in order to improve biogas and methane yield. For this reasons, the choice of suitable types of pretreatment and substrates for co-digestion with optimized operating conditionsmay significantly improve the running of the process and reduce its financial cost.
Keywords: Food waste, cattle manure, co-digestion, pretreatment, bioenergy.

EBP32: Isolation, Characterization of ichtyopathogenic bacterial strain in a lake ecosystem in Algeria

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Background and aim: Algerian fish production remains below the world production average, particuly continental production. According to the distinctive appearance of fish and the obvious pathological nature (parasitic, viral or bacterial), fish diseases have been recognized for centuries and have an impact on the economy of national aquaculture industry. The aim of our work is to characterize the diversity of ichtyopathogenic bacteria present in an Algerien lake ecosystem (dam of Guenira,Skikda).
Methods: The sampling was carried out aseptically, than cultured on specific and selective media. The ichtyopathogenic bacterial strains were identified firstly by the Api gallerys followed by molecular identification using PCR technique and sequenciong of 1 6S rDNA, also the resistance of bacteria to 20 different antibiotics was tested.
Results: The presence of an important ichtyopathogens species has been noted (*Providencia vermicola*, *Yersinia enterocolitica*, *Vibrio sp*, *Aeromonas hydrophila*, Psychrobacter sp, Microbacterium sp ... etc), belonging to the group of negative gram bacteria, as well as other strains not mentioed as ichtyopathogens (Alcaligenes sp, Staphylococcus sp, Comamonas sp, Exiguobacterium sp, Jeotgalicoccus halotolerans, Leucobacter sp, Vagococcus sp, ... etc) somme of this bacterial species is know b its resistance to antibiotic also its tolerance to metal, whereof, it can transfer resistance genes.
The isolates presente a multiple resistance to antibiotic especially to the β-lactams family how have a large spectrum of activity and are often prescribed in various infections.
Conclusion: The presence of a diversity of ichtyopathogenic bacteria how have an importante antibiotic resistance that can conributing to the spread of resistance genes in the Algeriens lakes ecosystems could have disastrous consequences for the health of fish fauna. As a result, the disruption of the trophic chain. For that, a special attention should be paid to this bioope.
Keywords: Ichtyopathogenic bacteria, Diversity, lake ecosystem, antibiotic resistance.

EBP33: Effect of biopesticide treatments on apple fungal necrosis under field conditions

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Background and aim: The main objective of this study is to evaluate the antagonistic effect of yeast and bacterial strains against fungal pathogens on apple trees under field conditions.
Methods: The effect of biopecticide on apple trees was investigated through spraying bacterial and yeasts suspensions under field conditions.
Results: Apple growing is economically important in Tunisia. It represents the third fruit crop after dates and citrus. Despite its progress remarkably, this culture is characterized by seasonal and anual fluctuations depending on climatic factors and losses due to pathogens attacks can adversely affect reproduction in quantity and quality. In order to investigate the appropriate formula, different nutrients were used : three sugars three amino acids and three salts each, were adjusted to three various concentrations. The evaluations of the reduction of Apple necrosis in fields revealed the appropriate basic formulas for yeast strains adjusted to the concentration of 10^8CFU/ml. The most protective strains belong to *Cryptococcus albidus (Or3), Pichia guielliermondii* (Pe1 1) and *Aureobasidium pullulans* (Prj3) with the percentages of reduction necrosis ranging from 77% to 92 %. Notable resistance of yeast strains has been reported against the main chemical pesticides used in the field.
Conclusion: individual treatments of yeast and bacterial strains reduced significantly the progress of apple necrosis under field conditions.
Keywords: biological control; apples; fungal necrosis; formulation; yeasts; bacteria.

EBP34: Effects of Ca and EGTA on antioxidative enzyme activities and their related gene expression in cadmium-treated chickpea (*Cicerarietinum* L.)

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Background and aim: Exposure of chickpea seeds (*Cicerarietinum* L.) to cadmium (Cd) stress for 6 days resulted in growth reduction. Numerous strategies have been adopted with the aim of improving plant responses to heavy metal intoxication, namely the selection of resistant cultivars, or exogenous treatment with chemical compounds. In this context, the enrichment of the seed germinating medium with calcium (Ca) and ethylene glycol tetraacetic acid (EGTA) relieved the detrimental effect of Cd on root growth (Sakouhi et al. 2016).This study was undertaken to provide deeper insights into the mechanisms induced by Ca and EGTA to protect plant cell against Cd stress. Impacts of Ca and EGTA on antioxidative enzymes activities and their related gene expression were examined in roots.
Methods: Chickpea seeds (*Cicerarietinum* L.) were germinated for three days in H_2O or 200 µM $CdCl_2$, then treated or not for three additional days with 10 mM $CaCl_2$or 100 µM EGTA.The content of hydrogen peroxide (H_2O_2) was evaluated. The activities of total superoxide dismutase (SOD; EC 1.15.1.1), SOD isoenzymes, catalase (CAT; EC 1.11.1.6), and ascorbate peroxidase (APX; EC 1.11.1.11) were measured spectrophotometrically at 25 °C, and their related gene expression was evaluated by reverse transcription and real-time quantitative (RT-PCR) method. The actin *C. arietinum* gene (ACT) was used as an internal control gene.
Results: Calcium and EGTA application to Cd-treated seedlings led to oxidative stress alleviation as evidenced by H_2O_2 content decrease and the restoration of superoxide dismutase,catalase and ascorbate peroxidase activities at a level similar to control roots. Moreover, the analysis of the transcriptional system relating to the up-cited enzymes revealed a decreased gene expression subsequent to the enrichment of germination medium with Ca or EGTA.
Conclusion: Ca and EGTA supply alleviate Cd-inducedoxidative stress in chickpea root cells. Furthermore, these effectors seem to mitigate Cd effect on antioxidative stress enzymes activities. This enzymatic modulation would be essentially the consequence of a transcriptional regulation of related genes. Therefore, amendment of Cd-polluted soil with Ca or EGTA, even after seed sowing, could reduce the toxicity of Cd and increase the crop production.
Keywords: Cadmium, Calcium, Gene expression, Oxidative stress.

EBP35: Abundance and diversity of prokaryotes in ephemeral hypersaline lakeChott El Jerid

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Background and aim: Chott El Jerid is the largest hypersaline ephemeral lake in southern Tunisian Sahara desert and is one of the biggest depressions at the North of Africa. The aim of this study was to investigate the diversity and abundance of microbial communities inhabiting ChottEl Jerid during wet season.
Methods: The abundance and the diversity of prokaryotes were investigatedusing molecular approaches:QPCR, sequencing by Illumina Miseq, and DGGE.
Results: 16S rRNA gene analyses revealed that bacterial community was dominated by *Proteobacteria* followed by *Firmicutes*, *Bacteroidetes*, *Cyanobacteria*, *Actinobacteria* and *Verrucomicrobia*. The results obtained using prokaryotic universal primers showed low relative abundance of Archaea dominated by few OTUs related to *Methanosarcinaceae* and *Methanomassiliicoccaceae* families and the presence of sulfate-reducing Archaea affiliated with Archaeoglobus. However, the results obtained using Archaea-specific primers showed that archaeal community was mainly composed of aerobic *Halobacteria* and anaerobic members of *Methanomicrobia*. QPCR results revealed that Archaea were more abundant in studied samples than Bacteria. The sulfate-reducing Bacteria were also found abundant (~ one-third of the bacterial community) and outnumbered methanogens.
Conclusion: The present study provides valuable information about the prokaryotic communities inhabiting the hypersaline ephemeral lake Chott El Jerid. Betaproteobacterial group (*Ralstonia* species) and *halobacterial* group (*Halorubrum* species) dominated Bacteria and Archaea, respectively, during wet season. Methanogenic populations and sulfate-reducing Bacteria were also detected during flooding period, suggesting their potential important role in this sulfate-rich and hypersaline ecosystem.
Keywords: DGGE; Hypersaline lake; Illumina Miseq sequencing; Methanogens; Prokaryotic diversity; Quantitative PCR; Sulfate-reducing Bacteria.

EBP36: Phytochmical screening, chemical composition, physical and chemical properties, and anti-feedant effect of essential oils extracted from medicinal plant, *lavandula angustifolia* against stooredpruducts pest, *Rhyzopertha dom inica*(F.) (Coleoptera: Bostrichidae)

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Background and aim: Losses Post-harvest of cereals and pulses are a major problem in Algeria. Insect pests of stored commodities, mostly Coleoptera can cause the total loss of a stock. The most common way to limit their activities is the use of pesticides whose side effects are unfortunately very numerous. In the last two decades, many works were carried for the purpose research for more selective, more specific methods of protecting stored products that respect human health and the environment. The use of chemicals of botanical origin for their repellent effects, fumigant and antifeedant propertiesas the best alternative of clean fight against these pests.
Methods: Our work is part of this theme and aims to evaluate éfficacityof the powder and essential oils of *Lavandula angustifolia*medicinal plant against adults of *Rhyzopertha dominica*, beetle pest of stored products.
Results:
-Phytochemical screening of the plant: the following compounds: flavonoid flavonoids, tannins, leucoanthocyanans, saponins, terpenoids and steroids are detected.
- Physicochemical properties of essential oils : isolated with hydro-distillation method by Clevenger apparatus, the results showed that the parameters tested are in agreement with those mentioned by the standards.
-Chemical compositionof essential oils was determined by gas chromatography-mass spectrometry (GC-MS).Results showed that EO of *l angustifolia* contains 56 compounds with Linalool (20.48%), Linalyl acetate (13.24%)Camphor (13.15%), and1,8 Cineole (12.96%). as the majority compounds With 3.2 ± 0.15 %. in yield relative to the dry matter.
- Effect of essential oils on the amount of food ingested and growth of insect's individuals: the results obtained showed decrease of weight and the amount food intake against the targeted insects
Conclusion: This study provide an interesting opportunity to develop bioinsecticides based on the extracts from plants medicinal. This finding showed that L. angustifolia EO induced toxicity and physiological disruption affecting the ability of the insect to digest food. This antifeedancy provide a new strategy for integrated pest management programs. Also, we need some additional studies for formulating and improving methods of application.
Keywords: *Rhyzopertha dominica*, *Lavandula angustifolia*, antifeedant activity, Phytochemical screening.

EBP37: Study of the competitive adsorption in a ternary system of the three organo-inorganic micropollutants on granulated activated carbon (GAC)

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Background and aim: The objective of this study consists firstly in the adsorption of singlecomponent systems GM Green Malachite, RhB, and cadmium (Cd) as inorganic pollutant model onto granulated activated carbon (CAG).Adsorption studies in binary mixtures of several pairs adsorbates (RhB/VM);(RhB/Cd); (VM/Cd) as in ternary mixtures (VM/RhB/Cd) were realized by examining the effects ofthe mass ratio adsorbat/co-adsorbate
Methods: For the purpose of investigating and investigating the capacity and affinity of granulated activated carbon in the adsorption of organo-inorganic micropollutants, on the one hand, in static mode (adsorption in single-component system and multicomponent system) and on the other hand, in continuous mode in fixed bed (dynamic adsorption). We chose two water-soluble commercial colorants widely used in the textile and tanning industries. These products are Rhodamine B (RhB) and Malachite Green (VM). For the inorganic pollutant, we studied cadmium (Cd), which is considered one of the most serious pollutants of concern for health and the environment
Results: Kinetics adsorption results in the single system reveal that the molecules of VM are adsorbed better than the molecules of RhB and cadmium. Results analyzing of adsorption isotherms of the three micropolluants, show that, the adsorbed quantities of RhB, VM and Cd in binary systems and ternary is lower than those obtained in simple systems monocomposés for the whole of the mass ratio, which reveals the antagonistic effect of the competitive adsorption ofthe three micropolluants onto activated cabon.
Conclusion: Obtained results in single and binary system confirm that, the application of CAG in dynamic adsorption in fixed bed confirm that the activated carbon has the same order of affinity presented towards the organic micropolluants in batch.
Keywords: Granulated activated carbon (CAG), VM, RHB, competitive adsorption, fixed bed.

EBP38: Influence of season and phenotype on ovarian steroids secretion in Algerian local rabbit does

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Background and aim: The global study was carried out on the reproductive performances of an Algerian local population rabbit does that have a white dress with red eyes (albino) or a colored dress with black eyes, raised in natural conditions, natural mating and semi-intensive rhythm. The aim of the present study is to determine under these conditions, the serum concentration of 17β- œstradiol (E2) and progesterone (P4) with their seasonal variations according to their phenotypes (albino *vs* colored).
Methods: Blood was collected over one year, from 92 does and 76 does (for the analysis of E2 and P4 respectively), separated into equal groups between the four seasons and the two phenotypes. In the middle of each season, blood samples were collected immediately after mating (E2) or at 12 days after mating with proved pregnancy by palpation (P4). Samples were centrifuged and analyzed by the ECLIA process. The results were analyzed using analysis of the variance (ANOVA on STATISTICA 6.0).
Results: The average concentration of E2 at mating was 18.7± 5.9 pg/ml and at 12 days of pregnancy the concentration of P4 was 18.04 ± 5.85 ng/ml. Significant seasonal variations in the production of steroids have been observed principally between winter and summer with 22.9±5.2 pg/ml *vs* 17.6±5.7 pg/ml ($P<0.05$) and 14.89±4.73 ng/ml *vs* 20.93±5.42 ng/ml ($P=0.01$), for E2 and P4 respectively. The two phenotypes gave averages of these steroids without significant difference, both for E2 and P4. In spite of the absence of phenotype effect, interactions season-phenotype showed a low significant variation in the production of E2 ($P=0.05$) in the colored rabbit does and in P4 ($P=0.05$) production in the white ones.
Conclusion: This study has shown environmental conditions effect on steroid production by low averages and seasonal variations. The inversion observed in the seasonal hormonal production between E2 and P4 is probably due to the antagonism of these steroids in their functions and regulations. The significant interactions season-phenotype can be explained by the different adaptation abilities of the rabbit (like other animal species) to temperature or photoperiod changes.
Keywords: local rabbit does, ovarian steroids, seasonal variations, phenotype.

EBP39: Effect of biosorption parameters on the removal of anionic azo dye Direct Red 28 (Congo Red) from aqueous solutions by Algerian CMPpS, Kinetics, Thermodynamics, and Application in desalination of natural seawater

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Background and aim: Congo Red (CR) that is an anionic azo dye adsorption was carried out in batch experiments on Calcined Mussels *Pernaperna* Shells (CMPpS) from aqueous and saline solutions.
Methods: The characteristics of the adsorbent were determined by using X-ray fluorescence (XRF), X-ray Diffraction (XRD), Fourier Transform Infrared (FTIR), Scanning Electron Microscopy (SEM), pH_{PZC} and BET. The adsorption process was strongly influenced by pH, contact time, particle size, adsorbent dose, dye concentration, agitation speed, NaCl salt and temperature.
Results: It was found that the adsorption of Direct Red 28 on CMPpS follows the Freundlich adsorption isotherm (r^2= 0.979), while the pseudo second order rate equation described the kinetic adsorption data quite well (R^2> 0.99). Free energy of adsorption (ΔG^0), enthalpy (ΔH^0) and entropy (ΔS^0) changes are calculated to know the nature of adsorption. The calculated values ofΔG^0 at 303K, 313K and 323K indicate that the adsorption process is spontaneous. The estimated values of ΔH^0 and ΔS^0 both show the positive sign, which indicate that the adsorption process is endothermic and the affinity between CMPpS adsorbent and RC. In the other hand, the CMPpS was employed to desalinate CR dye from saline solutions: NaCl solution (concentration similar to artificial seawater), synthesized and real seawater. The results, showed removal efficiency, 74.08%, 69.09% and 63.95% from (CR/NaCl mixture), (CR/ artificial seawater mixture) and (CR/ natural sea water) respectively, when compared to (CR/ distilled water mixture) (99.68%).
Conclusion: The results indicate that the possibility to use CMPpS in pretreatment of seawater doped with anionic azo dye by adsorption and readily available materials is very important for guaranteeing the viability of desalination by reverse osmosis.
Keywords: Dye removal; Adsorption; Calcined mussel shells; *Pernaperna*, Water treatment.

BM Session
Bioactive Molecules and Applications

ORAL COMMUNICATIONS
(BMO)

TC1-BM/BMO1: Investigation of the physicochemical and antioxidant properties of gelatin edible film mixed with blood orange (*Citrus sinensis*) peels extract

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Background and aim: The present study aims to develop gelatin-based films containing phenolic extracts from blood orange (*Citrus sinensis*) peel, as an alternative for the existing non-natural packaging.
Methods: The effect of drying pretreatment on antioxidant and antibacterial activities as well as phenolic compounds' profiles of the extracts was investigated.
Results: Results showed that the fresh orange peel extract (FOPE) was more effective against all bacteria tested and exhibited higher antioxidant effect than that thermally-dried (DOPE). In addition, the LC-ESI-MS analysis showed that the quinic acid was the major compound among the total poly-phenols followed by rutin, trans-ferulic acid, naringenin and 4,5-di-O-caffeoylquinate. FOPE modified also the mechanical and thermal properties of gelatin films, where the FOPE enriched ones were more deformable than the control gelatin film. This effect is may be caused by the interactions between phenolic compounds of extract and gelatin chains, as assessed by the FTIR analysis. The FOPE-added films presented higher antioxidants properties than those of the control. Finally, the release kinetics of FOPE showed a typical shape of non-time dependent diffusion.
Conclusion: The overall results of this investigation contribute to the valorization of a high value-added orange and fish by-products to provide further progress in the development of food packaging. These films could be applied to the life extension of food products. Additionally, this strategy can be used for utilization of orange and any other citrus peel as bioactive ingredient in gelatin films. Further work should explore the potency of the elaborated films as an effective active packaging for food applications.
Keywords: Grey triggerfish skin gelatin; Blood orange (*Citrus sinensis*) peel, Release kinetics of phenolic extract

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BMO2: Anti-tumor effect of native and recombinant phospholipases A2 from *Scorpio maurus* venom glands

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Background and aim: Integrins are a large family of cell surface receptors mediating the interaction of cells with their microenvironment and they play an important role in glioma biology. In a previous study, we purified Sm-PLGV an heterodimeric phospholipase A2, from the venom glands of the Tunisian scorpion Scorpio maurus. This enzyme contains a Long chain, a penta-peptide insertion, which is cut out during the maturation process, followed by a short chain. A disulfide bridge links the two chains. Three recombinant forms of this enzyme were produced in Escherichia coli: rPLA2(þ5) with a penta-peptide insert, rPLA2(-5) without the penta-peptide, and the Long chain alone without the short one.
Methods: The anti-tumor effect of the native phospholipase A2 Sm-PLGV and its recombinant constructs was tested using Human glioblastoma cells U87 (ATCC).
Results: All phospholipases inhibited in a dose dependent manner the U87 cells adhesion, migration and invasion onto fibrinogen and fibronectin without any cytotoxicity. Sm-PLGV and its recombinant constructs blocked U87 migration by reducing their velocity and directional persistence. The inhibitory effect was related to a blockage of the integrins αvβ3 and α5β1 function. Inactivation of the enzymatic activity of Sm-PLGV by chemical modification with p-bromophenacyl bromide did not affect its anti-tumor properties, suggesting the presence of 'pharmacological sites' distinct from the catalytic site in scorpion venom phospholipases A2.
Conclusion: The anti-tumor effect for recombinant scorpion venom enzymes may serve as starting point for structure–function relationship studies leading to design a new generation of specific anti-cancer drugs.
Keywords: Adhesion, Anti-tumor effect, Invasion, Heterodimeric phospholipase A2 Migration, Scorpion venom glands.

BMO3: Expression, purification and biochemical characterization of a lipase from *Serratia* sp. *W3* Tunisian cultivar

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Background and aim: Lipases from microorganisms have attracted great attention since they find many biotechnological applications; recombination process is the most appropriate approach that allows for both an adequate fundamental study of the protein and a high-level production. In this study, we describe the cloning, expression, refolding and purification of a gene coding for a novel *Serratia* sp. w3 lipase (Sml).
Methods: The gene part, encoding the mature lipase was cloned and sequenced. The mature lipase from *Serratia* sp. *W3* (SmL) was expressed in *Escherichia coli* BL21 and purified to homogeneity using Sephadex G-75 gel filtration and a Ni-NTA affinity chromatography. The expression levels and some biochemical and kinetic properties of the recombinant Sml were determined and compared to the wild type one.
Results: The gene encoding the mature lipase was cloned and sequenced. The deduced amino acid sequence showed a significant similarity with various *Serratia* lipases. The recombinant lipase was purified by affinity chromatography with 35% recovery and the specific activity was 1400 U/mg using tributyrin as substrate, at pH 9 and 45 ◦C. The recombinant lipase presented quite interesting pH stability in a large wide of pH from 5 to 9 and activity is enhanced with Ca2 + and Mg^{2+} while Zn^{2+} and cu^{2+} are two inhibitors of Sml activity. Kinetic study revealed that Sml may hydrolyze both positions (*sn* 1 and *sn* 3) but exhibit a clearly regio-preference toward the *sn*-3 position with significant increase in the specific activity of the rSml in comparison to the native form.
Conclusion: Recombination process allowed a high-level production and an improvement of specific activity that elect Sml as a potential candidate for biotechnological application.
Keywords: *Serratia* sp. W3; lipase; Purification; Sequencing; Expression; Characterization.

BMO4: Chemical composition, antibacterial and antibiofilm activities of selected essential oils against *Staphylococcus epidermidis* biofilm

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Background and aim: Bacterial biofilms cause major health problems because of their increased resistance to antibiotics. The current trend is the use of natural molecules from plants to combat microbial biofilm. Spices are widely used in Tunisia and their essential oils are rich with bioactive molecules and are well known for their medicinal properties.
Methods: The chemical composition of essential oils was analyzed by GC-MS. The antibacterial activity was investigated using microdilution method and the antibiofilm effect was evaluated by crystal violet test.
Results: Four spices essential oils *C.v.*, *C.l.*, *C.c.* and *P.n.*were used in this study. The chemical analysis of essential oils by GC-MS showed the abundance of Eugenol (57.87%) in *C.v.*EO. Whereas, *C.l.* (EO) was mainly composed of f3-turmerone (43.98%) and a-curcumene (30.34%). The main component in *C.c.* (EO) was Cuminaldehyde (28.22%), while *P.n.* EO was mainly characterized with the abundance of Limonene (19.99%), f3-Caryophyllene (16, 8%), a-Pinene (15.88%) and -3-carene (13.05%) as major components. The antibacterial activity showed that *C.v.*EO exhibited the most important antibacterial activity against several tested bacterial strains. Antibiofilm activities evaluated against *Staphylococcus epidermidis* S61 showed important antibiofilm effects for all testedspice oils, whose, *C.v.*EO exhibited the strongest anti-adherence and eradication activities. In fact, the concentration of 5 jtl ml^{-1} of *C.v.*EO was able to inhibit the formation of *S. epidermidis* biofilm (87.3%) and to eradicate 68.6 % of *Staphylococcus epidermidis* mature biofilm. The mixture of these oils improved the anti-adherent activity and the biofilm eradication of *Staphylococcus epidermidis* S61. Fluorescence and scanning electron microscopy observations confirmed these results. On the other hand, a statistical analysis through the Pearson correlation matrix and the Bayesian networks modeling (BN) agreedwith these results and confirmed the importance of *C.v.*EO in the inhibition and the eradication of *Staphylococcus epidermidis* S61 even at low concentration.
Conclusion: Essential oil from selected spices is promising source of anti-biofilm activities against *S. epidermidis* biofilm. A remarkable pronounced activity was recorded with The *C.v.* EO compared to the other EOs.
Keywords: Essential oil; GC-MS; *Staphylococcus epidermidis*; antibiofilm.

BMO5: Effect of lipopeptides from *Bacillus methylotrophicus* DCS1 strain on the stability of sunflower oil in water emulsion and on the conservation of raw beef patties quality during chilled storage

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Background and aim: The present study aims to investigate the effect of lipopeptides DCS1 on the stability of sunflower oil in water emulsion and on the conservation of raw beef patties quality during chilled storage.
Methods: An oil-in-water emulsion was prepared by dissolvingtween-20 (1%, w/w of emulsion) as emulsifier in Milli Q water (89%, w/w) and adding sunflower oil treated (10%, w/w) by drop wise addition to the water phase with continuous sonication, then addition of lipopeptides DCS 1 The primary oxidation products were measured using the peroxide value according to the ferric thiocyanate method and the secondary oxidation products were determined by thiobarbituric acid reactive substances (TBARS) method. The effect of lipopeptides DCS1,incorporated in ground beef patties (0.5%, w/w of meat) and in type Agelatin film (2.5%, w/w of gelatin) used as a coating, on lipidoxidation of the chilled raw beef patties was evaluated during 14 days of storage at 4 °C by the determination of TBARS, pH and color changes.
Results: Lipopeptides DCS1 showed good antioxidant properties in sunflower oil-in-water emulsions during prolonged storage (28 days) at 30 °C. They were effective at concentrations of 0.00416% and 0.0125% (w/w of emulsion) in slowing down the formation of hydroperoxides and secondary oxidation products. Lipopeptides DCS1 at a concentration of 0.0125% (w/w) are able to preserve the nutritional and organoleptic properties of the emulsion during 23 days of storage. The direct incorporation of lipopeptides DCS1 in ground beef patties was found to be more effective than the application of gelatin film enriched with lipopeptides as a coating, in inhibiting lipid oxidation and deterioration of beef patty quality during chilled storage (14 days). Furthermore, the lipopeptides DCS1 were not toxic to humankidneycellsHEK293 up to a concentration of 250 µg/ml.
Conclusion: The results of this study indicate that lipopeptides DCS1 could be appropriate for use in food models as inhibitors of lipid oxidation and substitutes for synthetic antioxidants.
Keywords: Lipopeptides DCS1; Oil-in-water emulsions; Groundbeefpatties; Lipidoxidation; HEK293 cells line.

BMO6: Antioxidative, antimicrobial and healing activity of a prodigiosin of *Serratia marcesens* microbiota of a traditional Algerian fermented food

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Background and aim: The objective of this work is the evaluation of prodigiosin from an isolated microbial operator of a traditional Algerian fermented cereal food. Antioxidative, antimicrobial and healing activities were determined to develop a natural galenic formulation for external use.
Methods: The bacterial strain was identified by Pcr, using a RNA16S probe. After extraction and purification ofthe red pigment, the infrared spectrum (FTIR) was determined. Antioxidative activity was performed by measuring scavenging radical with DPPH, the bleaching of beta-carotene. Antimicrobial tests were performed against bacteria and fungi pathogenics references strains. *Escherichia coli* ATCC25922, *Enterococcus faecalis* ATCC 10541, *klebsiella oxytoca*ATCC 13182, *Staphylococcus aureus* CC1 0541, *Helicobacter pylori*, *Candida albicans* ATCC 10231. Healing activity was measured in vivo on white rats using as a reference the commercial formulation Madécasol.
Results: The bacterium identified is a strain of Serratia marcesens, a strong producer of prodigiosin, whose IR spectrum is characteristic. The DPPH test shows a trapping power of 80% at 1 mg / ml and an IC 50 equal to 0.54 mg / ml. The discoloration of β-carotene is 50%. The anti-candida power is the most remarkable with diameters of inhibition greater than 20mm followed by staphylococcus. We have noticed that Helicobacter pylori is also sensitive with an inhibition diameter of 1 5mm. Prodigiosin ointment at 0.1 % in vaselin is used to measure healing activity in vivo. The results showed a fast and effective healing potential, better than the standard (Madécasol). The cicatrization is total without any trace of lesions. We noticed the absence of the redness phase.
Conclusion: This formulation, based on prodigiosin is very promising as a natural replacement for the synthetic drug, having powerful anti-microbial, healing and anti-i.
Keywords: Antioxidative, antimicrobial, healing activity, prodigiosin,*Serratia marcesens*

BMO7: Novel strategies for functional beef sausages productions

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Background and aim: In recent years, functional food has received special attention concerning not only nutritional and safe products, but also healthy and natural ones. Indeed, biological substances are most favored in functional food industry. Intensive research is currently run for developing functional meat products that reduce contents of salt, cholesterol fraction, nitrites which is obviously unhealthy due to the possible induction of carcinogenic nitrosamines, and fat, by incorporating functional ingredients such as vegetable proteins, antioxidants, probiotics, prebiotics, and synbiotics.
Methods: The effects of incorporation of probiotc strains, *L. plantarum* TN8, with reducing of nitrite content and/or *P. acidilactici* MA 18/5M as biological preservatives and fiber, namely barley beta-glucan concentrate (BBC), in beef sausages were evaluated during storage period at 4 °C. In fact, pysico-chemical, microbiological and textural properties were checked. Moreover, the effect of substitution of synthetic antioxidant in sausage by a biologically active component which is a polysaccharide extracted from *Sorghum bicolor* (L.) (SWSP) seeds were also evaluated as well as its activity against oxymyoglobin and lipid oxidation in beef sausage during storage.
Results: Results showed that the substitution ofpartial nitrite by *L. plantarum* TN8 improves texture profile in terms of a reduction of hardness, springiness and chewiness. Also, TN8 strain addition can be used as a biopreservative for beef sausage by decreasing the growth of *Salmonella enterica* and *Listeria monocytogenes* and maintaining lipid oxidation. The combination of probiotic strains namely, TN8 and MA 1 8/5M, and dietary fiber decrease the *Enterobacteriaceae* in the product. Reformulated sausages recorded good textural attributes and improved sensory features and showed a decrease b* color parameter and cooking loss. Sensorial and color parameters could be used for constructing regression models to predict overall acceptability. On the other hand, the free fatty acid profile was significantly affected by probiotic strains. Finally, SWSP presented high rate of oxymyoglobin and low lipid oxidation.
Conclusion: The incorporation of probiotic strains with fiber into beef sausage formulation could be an interesting strategy to produce healthier sausage. Moreover, SWSP can efficiently substitute synthetic antioxidants in meat industry.
Keywords: functional meat, probiotic strains, fiber, polysaccharides, beef sausages

TC2-BM/BMO8: Concentration and purification of*Porphyridium cruentum* exopolysaccharides by membrane filtration at various cross-flow velocities

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Background and aim: Currently, there is an increasing market demand for natural polysaccharides for the food, cosmetics and pharmaceutical industries. In particular, microalga polysaccharides receive much attention. Those coming from *Porphyridium* have important techno-functional and biological properties. Few studies suggest using tangential ultra- or microfiltration to concentrate, desalt and pre-purify soluble exopolysaccharides (EPS) whereas high degree of purity can be obtained. So, the operating conditions of the membrane filtration, notably hydrodynamic conditions inside the membrane lumen should influence the fouling intensity. Consequently, it was chosen to study the effect of the cross- flow velocity on the filtration performances. The aim of this study was also to set-up a simple refining chain capable of concentrating and purifying the EPS solutions without the addition of chemicals.
Methods: EPS from cell-free *Porphyridium cruentum* media were concentrated then purified on a 0.14 µm ceramic membrane. The influence of cross-flow velocities on filtration performances was investigated.
Results: Mean permeate fluxes equal to 49.8, 68.9 and 81.9 $L.h^{-1}.m^{-2}$ were obtained during the concentration at 4 bars for respectively cross-flow velocities inside the membrane lumen equal to 2.5, 3.3 and 4.2 $m.s^{-1}$; 49.7 $L.h^{-1}.m^{-2}$ for the diafiltration at 3.3 $m.s^{-1}$. Volume reduction factors higher than 7.8 were reached. Rejection rates of polysaccharides and proteins varied according to the cross-flow velocities. Thus, the EPS recovery rate or time of filtration could be modulated following the cross-flow velocity. Rheological behavior of filtered solutions changed following the concentration progress. More than 80 % (w/w) of polysaccharides were recovered while 49 % and 99 % of proteins and salts were removed respectively. EPS had molecular weight equal to 2.4 10^6 Da and contained xylose, galactose, glucose and glucuronic acids in the molar ratios of 1.5/1.3/0.6/0.5.
Conclusion: *Porphyridium cruentum* exopolysaccharides were concentrated more than 6 folds, and purified by ceramic membrane filtration. High yields of EPS recovery and good permeate fluxes were obtained. Final product was poorly salted, low in proteins but with nearly 2 $g.L^{-1}$ of exopolysaccharides. Thus, significant amounts of exopolysaccharides could be obtained in the case of important volumes of culture medium.
Keywords: Membrane filtration, Exopolysaccharides, Microalga, Concentration, Diafiltration.

BMO9: Water-souble polysaccharides and hemicelluloses from Ephedra alata: angiotensin-I-converting enzyme inhibitory and biological activities

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Background and aim: *E. alata* presents one of the most famous Traditional Medicine. In fact, this medicinal herb has the ability to promote diuresis and sweat, and to relieve asthma. In the modern Traditional Chinese Medicine, *E. alata* was used for the treatment of antipyretic, asthma and rheumatoid arthritis. The aim of this study was to investigate the angiotensin-I-converting enzyme (ACE) inhibitory and biological activities of two polysaccharides extracted from *E. alata*.
Methods: Water-soluble polysaccharides (EAP) and hemicelluloses (EAH) were extracted from *E.alata* by hot water and alkaline solution, respectively. These extracted polymers were characterized and their monosaccharide compositions were determined using the GC-MS method. EAP and EAH were then evaluated for their angiotensin-I-converting enzyme (ACE) inhibitory effect as well as their antioxidant activities using the DPPH, ABTS radical scavenging activity and reducing power tests. Afterward, the extracted polysaccharides and hemicelluloses were tested for their functional properties and their antibacterial activities against a panel of 10 pathogenic bacteria.
Results: The extraction yield of EAP and EAH was 4% and 17%, respectively. The chemical composition of EAP and EAH indicated that the contents of total sugars and uronic acid were 71.75%, 6.82%, and 75.68%, 6.96%, respectively. The GC-MS results showed that these two polysaccharides were essentially composed from Glc, Gal and Ara. These both extracted polymers displayed high ACE inhibitory activities (CI50 = 0.21 mg/ml and 0.20 mg/ml respectively). The test of antioxidant activities showed that these extracted polysaccharides exhibited strong DPPH radical scavenging activities (CI50 = 0.32 mg/ml and 0.54 mg/ml, respectively) and high reducing powers (RP0.5AU = 0.28 mg/ml and 0.30 mg/ml, respectively). The evaluation of EAP and EAH functional properties demonstrated that these polymers presented important emulsifying and foaming properties. Finally, the results of antibacterial test showed that these polymers presented interesting antibacterial activities against *Enterobacter sp*, *Salmonella typhimurium*, *Listeria monocytogenes*, *Micrococcus luteus* and *Bacillus subtilis* when applied at 50 mg/ml.
Conclusion: Our results demonstrated that these polymers could offer promising sources of bioactive molecules for future applications as dietary ingredients in food and pharmaceutical or cosmetic industries.
Keywords: *E. alata*, Polysaccharide, Hemicelluloses, Angiotensin-I-Converting Enzyme and biological activities.

BMO10: Structural characterization, rheological investigations and antioxidant activity of water-soluble polysaccharides from the Tunisian brown seaweed *Cystoseira compressa*

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Background and aim: Over the past decade, there has been a growing interest in the search for new bioactive compounds extracted from marine algae, in part because of their many beneficial effects on human health. Particular attention is given to the polysaccharides of brown algae, which have diverse and marked biological activities and interesting structural richness. Numerous studies highlighting the anti-cancer, anti -thrombotic, anticoagulant or anti-inflammatory activities of polysaccharides extracted from these macro -algae can be enumerated in the literature. Due to its geographical location, Tunisia and more particularly the island of KERKENNAH has a set of geographical and environmental specificities that confer a particularly varied marine biodiversity. *Cystoseira compressa*, an abundant brown algae along the Tunisian coast, has been selected as a candidate to investigate its content of potentially bioactive polysaccharides.
Methods: Biochemical assays, chromatographic (HPAEC, GC/MS and SEC/MALLS) and spectroscopic (FT-IR, NMR) analyzes led to the characterization ofpolysaccharides.
Results: A fucoidan (CCF) and a sodium alginate (CCSA) were extracted and purified from the Tunisian brown seaweed *Cystoseira compressa*. CCF was a highly sulfated heterogalactofucan composed of *a*-(1—*3), *a*-(1—*4)-linked L-Fuc*p* as main backbone which could be highly branched (31.84%) at *O*-3 and *O*-4 positions of *a*-(1 —*4)-L-Fuc*p* and *a*(1—*3)-L-Fuc*p* by terminal monosaccharides and side chains such as terminal *a*-L-Fuc*p*, terminal *ß*-D-Gal*p*, *ß*-D-Gal*p*-(1—*3)-*a*-L-Fuc*p* and *ß*-D-Gal*p*-(1—*4)-*a*-L-Fuc*p*. The ratio of *a*(1—*3)/*a*-(1—*4) linkages was estimated at 3.86:1. CCSA was characterized by HPAEC-PAD, GC/MS-EI, ATR-FTIR, and ^{1}H-NMR. The M/G ratio was M/ G=0.77, indicating that CCSA respectively contained 44% and 56% of mannuronic and guluronic acids. The values of FGG, FMM, FGM (or FMG) blocks as well as the parameter η were estimated. The two polysaccharides exhibited effective antioxidant activities by ferrous ion chelation, ferric ion reduction and DPPH radical scavenging. Their rheological behaviors were also evaluated, outlining their potentials as natural additives.
Conclusion: These structural analyses showed that CCF and CCSA presented a new and different structures compared to the fucans and alginates ofothers brown seaweeds.
Keywords: Brown seaweed, Sulfated polysaccharide, Fucoidan, Alginates, Antioxidant activity.

BMO11: Exopolysaccharides from a new isolated bacteria: Purification Structural Analysis and Antioxidant activities

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Background and aim: Exopolysaccharides (EPSs) are defined as macro molecular weight, consisting mainly of carbohydrates, biodegradable polymers which biosynthesized by a wide range of organisms as bacteria, fungi, and blue-green algae. The main of this work is to isolate and identify a bacterial strain capable of producing appreciable amount of EPS, to characterize and to purified the produced EPS and to assess its antioxidant activities.
Methods: 16S rDNA, Purification,GC-MS,NMR
Results: This bacteria appointed KS1 identified as *Bacillus subtulis* based on morphological and physiological characteristics and phylogenetic analysis of 1 6S rDNA sequences. The polymer purified by chromatography gel filtration S400. The new EPS was determined as homogeneous,as determined by High-Performance Size-Exclusion Chromatography(HPLCSEC).Structure of EPS was investigated and elucidated using gas chromatography–mass spectrometry (GC–MS), methylation, nuclear magnetic resonance (NMR). This study also aimed to evaluate the antioxidant activity of EPS. Indeed the evaluated activities included the total antioxidant activity of purified EPS were 70,27±3,315-tocopherol (µmol/mL) ,DPPH radical-scavenging capacity (IC50 EPS = 2mg/mL), reducing power (OD: 0.64±0.532 at a concentration of 2 mg/mL.
Conclusion: These findings indicate that the EPS have potent antioxidant activities, outlining their potentials as natural additives.
Keywords: exopolysaccharides, purification , characterization ,antioxidant activity.

BMO12: Biocidal effect of *Melia azedarach* seed oil encapsulated in B-cyclodextrin on devastating insect species and environmental protection

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Background and aim: Synthetic chemical pesticides are often found as residues in food and pose risks to human health and the environment. In order to reduce the harmful effects of synthetic products, it is interesting to move towards control by the use of active substances and non-polluting. *Melia azedarach* seed oil can act as an effective and natural pesticide that has strong anti-insect pest and regulate the growth substance
Methods: The aim of this study is to validate the use of *Melia azedarach* seed oil of Algerian origin extracted by soxhlet and encapsulated by lyophilization's method "freeze-drying". The manufacture of the capsules is obtained in two stages (emulsification and lyophilization) in order to characterize their physicochemical properties. In vitro controlled release of encapsulated *Melia azedarach* seed oil was achieved for the first time. We also evaluated the insecticidal potential of encapsulated *Melia azedarach* seed oil on two devastating insect species: Rice weevil (*Sitophilus oryzae*) and Grain capuchin (*Rhyzopertha dominica*).
Results: The results of the insecticidal activity of the encapsulated *Melia azedarach* oil indicate that this oil exhibits an effective toxicity against *Rhyzopertha dominica (F.)* and *Sitophilus Oryzae* which reach 100 % for the following doses 0.05, 0.1, 0.3 and 1g of capsule.
Conclusion: The results of this study show that the use of β-cyclodextrin-epichlorohydrin polymer as a matrix can create a protective barrier around the oil to protect the most active compounds of the oil from environmental damage and ensure a long life. The duration of the conversation, as well as this study shows that the seed oil of Melia azedarach has very good insecticidal properties against Rice weevil (*Sitophilus oryzae*) and Grain capuchin (*Rhyzopertha dominica*) and could serve as an alternative to synthetic chemical pesticides.
Keywords: encapsulated *Melia azedarach* seed oil, *in vitro* release, insecticide, *Rhyzopertha dominica (F), Sitophilus Oryzae.*

BMO13: Physicochemical, structural and sensory properties of smooth hound autolysates-sugar conjugates formed using Maillard reaction

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Background and aim: Among the numerous existing protein modification techniques, the glycation of proteins induced by sugar isomerisation, via naturally-occurring Maillard reaction (MR), is commonly known as the best process on improving functional properties of food proteins. In fact, the amine-carbonyl condensation occurring during cooking, results in numerous transformations giving rise to protein-sugar conjugates and then the formation of brown and polymeric products, named Maillard reaction products (MRP). Thus, the aim of this study is to asses the conjugation of smooth hound viscera autolysates (SHVA) and glucose (Glu), through the Maillard reaction (MR).
Methods: The conjugation between autolysates and glucose was performed by mixing the selected SHVA powder and glucose at an equal weight ratio (protein / glucose; 1:1) followed by their incubation at 60 °C during 4 days to induce the MR. The degrees of glycation, free amino groups, color parameters, structural (infrared and scanning electron microscopy analyses) and antioxidant properties of the resulting SHVA-based MRP were studied.
Results: At the end of the experience, the degrees of glycation reach 87% after 4 days of heat treatment, proving the effectiveness of the MR throughout heating in both systems. In addition, the MR improved the sensory attributes of the MRP, particularly, by reducing the unfavorable bitter taste. Moreover, structural changes were observed in the infra-red spectra and the SEM micrographs of the final conjugates, compared to the un-conjugated ones. These changes were consistent with the antioxidant activities of the different samples. In fact, it was found that the antioxidant capacity of the MRP derived from both autolysates increased with heating time, as assessed by their reducing power and metal chelating activities.
Conclusion: The present study demonstrates that the combination of SHVA and glucose induced the production of bioactive products with different structural and enhanced biological characteristics, and they may be, therefore, potentially used in food applications.
Keywords: Smooth hound shark; Autolysates; Glucose; Maillard reaction products.

TC3-BM/BMO14: Characterization and anticoagulant activity of a novel fucosylated chondroitin sulfate

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Background: Sulfated polysaccharides have attracted considerable interest in recently years due to their potential therapeutic application particularly in the medical field as an anticoagulant and antithrombotic agent. Indeed we investigated structure and anticoagulant effect of fucosylated chondroitin sulfate (FuCS) from sea cucumber *Holothuria polii*.
Methods: The FuCS isolated from the sea cucumber body wall was purified by anion exchange chromatography and the structure was characterized by FT-IR and NMR spectroscopy. The molecular weight measurements were performed by HPSEC-MALLS-dRI. Anticoagulant activity was measured *in vitro* in plasma by classical anticoagulation tests and in a purified system by thrombin inhibition in the presence of antithrombin and heparin cofactor II. The effect on thrombin generation with and without CTI was investigated using calibrated automated thrombography (CAT).
Results: FuCS with high sulfate content 43% and with a relatively low average molecular mass of 45.8 kDa was isolated from *H. polii* at 4.66.±.0.5 mg/g dry body wall. Structural analysis of this polysaccharide revealed the presence of alternating units of chondroitin sulfate branched at position 3 of the D-glucuronic acid by two fucose 2,4-O-di and 3,4-O-disulfated residues in the proportion of 46 and 54%, respectively. The FuCS exhibited a high anticoagulant effect mediated by heparin cofactor II and to les ser extent by antithrombin with IC50 value of 0.035 µg/mL and 0.125 µg/mL, respectively. Moreover, the CAT assay depicted a pro- and anticoagulant effect of FuCS.
Conclusions: The high anticoagulant effect and the particular physicochemical characteristics of *H. polii* FuCS open clearly new perspectives for the development of new anticoagulants drug replacing those of the first-generation.
Keywords: Fucosylated chondroitin sulfate, Sea cucumber, Thrombin generation, anticoagulant effect, *Holothuria polii*.

BMO15: Study of the effect of leaves's aqueous and methanolic extracts of *Olea europaea var sylvestris* on germs responsible for urinary tract infections

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Background and aim: Infectious and parasitic diseases are a public health problem in Algeria because of their frequency. Medical consultations and antibiotic prescriptions for urinary tract infections are common. The aim of this study is to verify the effectiveness of various extracts "aqueous, methanol, dimethy sulfoxide" of wild olive leaves on bacteria and yeasts responsible for urinary tract infections.
Methods: the leaves of *Olea europaea var sylvestris* are harvested in the Chréa forest, the bacteriological material consists of reference bacterial and fungal strains, and strains from ECBU. The "aqueous, methanolic, and dimethyl sulfoxide (DMSO)" extracts are tested for phytochemical screening, and the phenolic compounds are assayed by high performance liquid chromatography (HPLC).
Results: Phytochemical analyzes revealed the presence of secondary metabolites. HPLC identified Oleuropein as the major phenolic compound of the crude extract of the species.
Antimicrobial activity tests carried out in vitro have made it possible to evaluate the potency of the extracts of the oyster oil on pathogenic germs. At concentrations between 62.5 mg / ml and 250 mg / ml, the organisms proved to be sensitive to all three extracts, only the *S. aureus* strain showed resistance to the different doses. The zones of inhibition (ZI) were between 06-12mm for the aqueous extract, 07-1 1mm for the methanolic extract, and 09-1 5mm for the DMSO.
Conclusion: In the present research, we revealed the secondary metabolites and phenolic compounds of the *Olea europaea var sylvestris* extracts and that the three extracts tested have an inhibitory effect on the growth of the studied germs.
Keywords: *Olea europaea ver sylvestris,aqueous extract, methanolic extract, urinary infections*.

BMO16: Effect of pollen source and pollination time on yield and fruit characteristics of "Deglet Nour" date palm (*Phoenix dactylifera* L.) in the Oasis of Djerid (Tunisia)

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Background and aim: In Tunisia, date palm cultivation is the mainstay of agricultural activity in the regions South West. Oases are unique sources of greenery and life in the desert. The oasis represents a pillar industry that affects economic growth and added value for farmers. On the other hand, the date palm growers have been able to deduce the importance of the origin ofthe pollen used on the quantity and the quality of the date production. As a result, they selectively use pollinating male varieties. The study was undertaken during tow years 2017 and 2018; it aimed to determine the influence of pollen sources and pollination time on fruit set, yield and quality of date palm cv. 'Deglet Nour'.
Methods: Tow 'Deglet Nour" female trees and ten different male trees (M-1 to M-10) were selected for the experiment. On each female tree three spathes were chosen and were pollinated each with pollens collected. Out of six, two spathes were pollinated with an interval of three days until one month after opening spathe. Data were collected for physico-chemical characteristics of the fruit.
Results: The results indicated that males were significantly differed from one another in all these characters namely for fruit set (%), fruit weight (g), fruit length (cm), fruit breadth (cm), fruit and yield (kg). Pollination at different day had significant effect on the fruit setting and other quality parameters. The pollen variety had significant effects on fruit characteristics as total sugar, acidity, bricx and mineral elements. Finally the pollen variety and pollination time interaction effects showed that, application of pollen from 2-4 days after spathe opening with the most production date palm yield, so this treatment is the best and are recommended.
Conclusion: In conclusion the nature of the pollen has an important effect on the quality of the fruits obtained; the farmers must choose their pollen based specifics criteria. To ensure a good rate of setting the pollination must be done between the 2nd and the 4th day after opening spathe.
Keywords: Date palm, pollen, pollination time, metaxenia, yields, for physico-chemical characteristics, fruit quality.

BMO17: Insecticidal toxicity of *Laurus nobiis* essential oil on *Tribolium castaneum* adults and impact on semolina characteristics

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Background and aim: Historically, cereal production has always been an important component of Tunisian agriculture. Durum wheat is the major crop and the most widely cultivated cereal. Insect pests caused yearly great economic losses both in fields and in storage and are considered as a serious biosecurity threat to Tunisian cereals' production and industry. Thus, several methods based on techniques such as fumigant action and repellent activity of a broad-spectrum of insecticides have been always used in storage. Nevertheless, the use ofthese chemicals is controverted due to human and environmental worries.
Methods: Trials were conducted using laurel *Laurus nobilis* (L.) essential oils for the control of the red flour beetle *Tribolium castaneum* (Herbst). Adults were treated with *L. nobilis* essential oil at the $_{LC50}$ concentration (208 µl/l air) and mortalities were assessed after 10 and 60 days of storage under an 100% spaces occupied with semolina. Moreover, we investigated the impact of the essential oil on semolina characteristics (protein content, wet gluten content, gluten elasticity, the falling number and ash contentdetermined according to International Standards) and rheological dough analysis (tenacity of the dough (P), extensibility of the dough (L), elasticity-to-extensibility ratio (P/L), strength of the dough (W) and elasticity resistance of the dough (Ie). These analyses were determinedusing a Chopin alveograph according to ISO Standard.
Results: Results indicated that the oil exhibited strong fumigant potentialities. High mortalities were obtained after 60 days of exposure and with spaces 100% occupied with semolina (75.5% mortality). Additionally, results revealed that essential oil treatment slightly modified semolina characteristic but did not negatively affect rheological proprieties of dough that remains conform to international ISO standards.
Conclusion: We conclude that *L. nobilis* essential oil constitute a promising biocontrol agent for the control of *T. castaneum*.
Keywords: *Tribolium castaneum*, essential oil, fumigation, semolina, ISO standards.

BMO18: *Syzygium aromaticum* aqueous extract inhibits human neutrophils myeloperoxidase and protects mice from LPS-induced lung inflammation

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Background and aim: *Syzygium aromaticum* (L.) Merr. & Perry (Myrtaceae) is widely used as a spice and has numerous medicinal properties. This study investigated the antioxidant potential of *Syzygium aromaticum* aqueous extract (SAAE) *in vitro* and its protective effects on LPS-induced lung inflammation in mice.
Methods: Neutrophils were isolated from healthy donors and reactive oxygen species (ROS) generation was measured by luminol-amplified chemiluminescence in resting cells and stimulated with N-formyl-methionyl-leucyl-phenylalanine (FMLF) or phorbol myristate acetate (PMA). Superoxide anion generation was detected by cytochrome c reduction assay. H2O2 was detected by DCFH fluorescence assay. Myeloperoxidase (MPO) activity was mesured by tetramethyl benzidine oxidation method. To study the anti-inflammatory activity of SAAE, Lung inflammation was induced in mice (BALB/c) by intra-tracheal instillation of lypopolysaccharide (5 µg/mouse), and SAAE (200 mg/Kg body weight) was injected intraperitoneally prior to LPS administration. Bronchoalveolar lavage and lung tissue were collected to assess inflammatory cells count and total protein content. Metalloproteinases activity was detected by zymography.
Results: In this study we show that SAAE inhibits luminol-amplified chemiluminescence of resting neutrophils and stimulated with FMLF or PMA, with an inhibitory effect starting at a concentration as low as 0.5 µg/mL. However, SAAE had no direct effect on NADPH oxidase activity and does not react with intracellular $_{H2O2.}$ Moreover, SAAE reduced significantly and dose-dependently myeloperoxidase (MPO) activity $_{(IC50}$ value: 0.5 µg/mL) *in vitro*. On the other hand, *in vivo* studies showed that SAAE (200 mg/Kg body weight) decreased markedly neutrophils count (From 61% to 15%) and proteins leakage into bronchoalveolar lavage fluid (BALF) after LPS administration. Gelatin zymography assay showed that *Syzygium aromaticum* inhibited MMP-2 (15%) and MMP-9 (18%) activity in lung homogenates.
Conclusion: Our findings suggest that the anti-inflammatory activity of SAAE, *in vivo*, is due to the inhibition of ROS production and metalloproteinases activity via its action on myeloperoxidase. Thus, SAAE possesses promising anti-inflammatory therapeutic potential.
Keywords: *Syzygium aromaticum*, neutrophils, ROS, myeloperoxidase, Bronchoalveolar lavage fluid, metalloproteinases.

BMO19: Evaluation of antioxidant andbiological activities of bioactive compounds from the rhizomes of wild *Asparagus albus* L.

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Background and aim: *Asparagus albus* L. possesses various health-promoting functions. However, the available information regarding biological activity of their bioactive components is still limited. Studies have shown that among different plant parts of *A. albus*, the rhizomes exhibit bioactivities which include antimicrobial, antioxidant and cytotoxic activities. In this work, chemical characterization, pancreatic lipase inhibitory, antioxidant, and cytotoxic activities of the crude saponins and polysaccharides fractions purified from the rhizomes of *A. albus*,were evaluated.
Methods: After hot water extraction, the saponins and water soluble polysaccharidesfractions were partially purified from the aqueous extracts, using an adsorbent polymeric resin. The high (HMW) and low (LMW) molecular weight polysaccharide was separated from the water soluble polysaccharides fraction by using alcohol precipitation method. The chemical characterizations of these bioactive compounds were determined by LC-MS, GC, GC-MS, and methylation analysis. Moreover, antioxidant (DPPH and FRAP assays), pancreatic lipase inhibitory activities and cytotoxic effect against human colon carcinoma HCT-1 16 cells were evaluated in vitro.
Results: The results revealed that LMW and HMW polysaccharides mainly contained respectively; carbohydrates (83% and 64%), uronic acids (13% and 3 1%), proteins (2% and 3%) and phenolics (2% and 2%). These polysaccharides are pectic in nature, with a predominantly unbranched galacturonan domain and with a domain bearing side chains that consist of highly branched arabinan, galactan, and/or arabinogalactan. In addition, based on methylation analysis, both polysaccharides fractions contain (1→2,1)- and (2→6)-linked β-D-fructofuranosyl units suggesting the presence of low percentage of fructans. The saponin fraction was characterized by the presence of sarsasapogenin as an aglycone and glucose, arabinose and rhamnose are the main monosaccharides linked to the aglycone. Additionally, the crude saponins and the two polysaccharides fractions showed a potent antioxidant activity. As well as they exhibited favorable pancreatic lipase inhibitory activity and significantly inhibit the growth of HCT- 116 human colon cancer cell line.
Conclusion: The preliminary results showed that the crude saponins and water soluble polysaccharides obtained from the rhizomes of *A. albus* are of interest for their biological activity and are suitable for being used as functional ingredients due to their potent antioxidant capacity.
Keywords: *Asparagus albus* rhizome, water soluble polysaccharides, saponins, antioxidant, cytotoxic activity, pancreatic lipase inhibitory activity.

BMO20: New Schiff bases derived from Benzimidazole and their Zn(II) complexes: synthesis, characterization and biological evaluation

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Background and aim: Schiff bases are an important class of ligands in coordination chemistry due to their ease of synthesis and their ability to be readily modified both electronically and sterically. These ligands possess potential applications in diverse fields such as optical materials, catalysis, chemical sensor and biological probes. Metal complexes of Schiff base ligands have been extensively studied because of their potency as active sites of metalloenzymes, antibacterial, antivirus, antimicrobial and antifungal agent.
Benzimidazole and its derivatives are reported to be physiologically and pharmacologically active and some applications are found in thetreatment of several diseases including epilepsy, diabetes and infertility. These compounds show also widerange of biological activities.
Methods: The structures of ligands were determined by nuclear magnetic resonance and single-crystal X-ray diffraction. Crystallographic data were collected with a Bruker APEX2 diffractometer using Mo-Kα radiation. The *in vitro* antibacterial screening of synthesized compounds was performed using the disc diffusion method. The antioxidant activity was investigated using DPPH radical.
Results: Using 1-methylbenzimidazole-2-carbaldehyde and 2-acetylbenzimidazole as starting materials, some new Schiff bases were prepared and their structures were determined. Synthetized benzimidazole shiff bases were subjected to complexation reaction with zinc (II), the results of the biological activity obtained indicates moderate to interesting activities, for both antibacterial activity and DPPH scavenging.
Conclusion: In this work, new Zn(II) coordination compounds have been synthesized, characterized by X-ray analysis, IR and UV-Vis spectroscopy, and screened for theirpotential as antimicrobial agents and as DPPH radical scavengers.
Keywords: benzimidazole, shiff bases, Zinc(II), antimicrobial activity, X-ray crystallography, DPPH essay.

BMO21: Synthesis and Biological Activities of 1,2,4-Triazol-3-yl-Thiazolidin-4-Ones

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Background and aim: A new 3-(5-alkyl-2-phenyl-2H-1,2,4-triazol-3-yl) thiazolidin-4-ones derivatives were obtained by condensation of 5-amino-1,2,4-triazoles, mercaptoacetic acid with aromatic aldehydes and catalyzed by Sm(SO3CF3)3 using microwave irradiation. The prepared compounds were tested for their antioxidant, antibacterial and antifungal proprieties. Some of these compounds displayed significant activities. Among them, compound **2e** exhibited remarkable activity against a broad spectrum of Gram positive, negative bacteria and pathogenic fungal strains with low MIC values. The investigation of the mode of action of the most potent antifungal compounds on the fungus *Pythium phanidermatum* showed a membrane alteration and distortions of hyphal morphology. The newly synthesized compounds exhibited also promising radical scavenging activity.

Methods: Microwave assisted combining with the use of DCC (dicyclohexylcarbodiimide) for synthesizing 3 -(5-alkyl-2-phenyl-2*H*- 1 ,2,4-triazol-3 -yl)thiazolidin-4-ones.

The *in vitro* antibacterial activity of compounds were carried out against Gram-positive and three Gram-negative bacteria. I*n vitro* antifungal activity against various phytopathogenic was tested on Potato Dextrose Agar (PDA) medium. For antioxidant activity, radical scavenging activity of synthesized compounds was determined using DPPH.

Results: The developed synthetic protocol used to access to a series of triazol-3ylthiazolidinones **2a–p**, which were obtained in moderate yields ranging from 45% to 56%. The compound **2e** exhibited the strongest antibacterial activity with inhibition zone ranged from 11 to 23 mm and very low minimum inhibitory concentration (MIC) values. It was also noticed that the Gram-positive bacteria *Bacillus cereus* and *Staphylococcus aureus* are the most sensitive bacteria to all synthesized compounds, whereas Gram negative ones are resistant to these compounds except for **2e.**

All thiazolidinones derivatives exhibit broad-spectrum antifungal activity towards several phytopathogenic fungi. Compound **2e** showed the greatest antifungal activity against all tested fungi with low MIC values ranging from 0.172 to 1.375 mg/ml.

Among the tested compounds, the most potent radical scavenger effect was obtained with **2f** which showed an inhibition of 91.57% compared to 98% using ascorbic acid.

Conclusion: Thiazolidinones are synthesized in moderate yields using microwave assisted combining with DCC. The biological profiles ofthese new compounds would represent a fruit full matrix for further development ofbetter antifungal, antibacterial and antioxidant agents.

Keywords: Aminotriazoles; Triazolothiazolidinones; Antimicrobial activity; Antioxidant activity.

POSTER PRESENTATIONS
(BMP)

BMP1: Study of the antifungal activity in vitro of the essential oil of rosemary (*Rosmarinus officinalis*) on fusarium (*F. oxysporum*) parasitizing tomato (*Lycopersicum esculentum*)

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Background and aim: In Algeria, cereals are particularly wheat, are the predominant crops and require continuous improvement, to meet agri-food needs. Obtaining high yielding cultivars, with resistance to various diseases and insects and good technological quality is a requirement in basic research The search for new molecules taking into account criteria other than efficiency, has become indispensable. Biological control through the use of natural antifungal substances can be an alternative to chemicals. Among these natural substances are essential oils extracted from aromatic and medicinal plants. *Fusarium Oxysporum* is one of the most destructive fungi of tomato (*Lycopersicum esculentum*) that can lead to wilting and death of the infected plant at an advanced stage. Pesticides, used in the preservation of tomatoes against deterioration agents, are often harmful to the environment. Biological control using biopesticides is currently required.
Our work consists in testing the effect of a biofungicide in this case the essential oil of an aromatic and medicinal plant Rosemary (*Rosmarinus officinalis*) on a microscopic mushroom (*Fusaruim oxysporum* sp.) affecting the tomato.
Methods: Extraction of the essential oil from the leaves of rosemary was done by hydro-distillation. The application of the oil extract was carried out according to two methods: well diffusion and remote diffusion.
Results: For this test, the two diffusion methods made it possible to demonstrate the antifungal power of the essential oil of the rosemary leaves on the fungal strain tested. The reducing effect of rosemary is also important with a percentage inhibition that varies from 55 to 100% (depending on the method used) while the control shows a rapid growth of the mushroom over the entire surface of the petri dish.
Conclusion: in vitro antifungal tests with the essential oil of oregano have shown that this natural fungicide has fungicidal properties very interesting and very effective
The essential oils of oregano, have shown a good antifungal effect at laboratory scale " in vitro " from where they can be used as alternative chemicals for the protection of wheat, the study must be carried out on fields " in vivo.
Key words: Essential oils, Biofungicides, *Rosmarinus officinalis*, *F. oxysporum*, Tomato, Yield.

BMP2: Evaluation of protective effects of *opuntiaficus-indica* cladodes on colorectal cancer

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Background and aim: Recent epidemiological studies on patients with obesity or type 2diabetes have revealed higher risks for several kinds of cancer, including breast, colorectal, pancreatic, and liver. In addition, oxidative stress may be a linking factor between obesity and cancer. Recently, functional foods are the subject of numerous studies because in addition to their high nutritional value, they offer an additional health benefit. The cactus and cladodes of *Opuntiaficus-indica* are widely consumed in the Mexican diet. It has a low glycemic index and is considered a functional food due to its high content of dietary fiber and polyphenols concentration. Phenolics are an important class of secondary plant metabolites possessing an impressive array of pharmacological activity. In this study, we investigated the protective effect of dietary cactus *Opuntiaficus-indica* cladodes (OFIC) on the oxidative stress and the incidence of colorectal cancer, induced by N-Methyl-Nitrosurea (NMU) in diet-induced obesity.
Methods: Phenolic compounds were extracted, and total polyphenols, flavonoids and tannins were assayed. Antioxidant parameters were evaluated by colorimetric methods. Wistar rats were fed for 4 weeks with a control or a high-fat diet, enriched or not with OFIC (50%), then colorectal cancer was induced by intraperitoneal (.i.p) injection of NMU (50 mg/kg body weight).The oxidative stress was monitored in plasma by measuring the MDA level, the carbonyls protein generation, CAT, SOD, GSH-Px and GSSG-Red activities and evaluation of antioxidant status by ORAC. BALB/c mice were subcutaneously implanted with CT26 colon carcinoma cells, and then were .i.p injected with (OFIC) polyphenols (50 mg/kg/d) during 7days.
Results: Our results clearly showed that obesity and NMU induced significant alterations in oxidative stress markers. Conversely, the cactus cladodes diet improved the oxidative stress and reduced the number of colon polyps induced by NMU/obesity. Furthermore .i.p. administration of OFIC polyphenols reduced CT26 tumor growth.
Conclusion: Dietary *Opuntiaficus-indica* cladodes (OFIC), by their content ofpolyphenols, may improve oxidant stress and decrease the risk of colorectal cancer, associated with obesity.
Keywords: *Opuntiaficus-indica*, cladodes, polyphenols, NMU, oxidative stress.

BMP3: Protective effect of Zizyphus lotus jujube fruits against cypermethrininduced oxidative stress and neurotoxicity in mice

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Background and aim: Cypermethrin (CYP) is a synthetic pyrethroid insecticide used worldwide in agriculture, home pest control. Cypermethrin (CYP), the α-cyano-3- phenoxybenzyl ester of 2, 2-dimethyl-3 -(2, 2-dichlorovinyl) 2-2-dimethyl cyclopropane carboxylate, is the most commonly used type-II pyrethroid insecticide. CYP exerts its neurotoxic effects through acting on voltage-dependent sodium channel and integral protein ATPase in the neuronal membrane The toxicity of CYP is well studied in many organisms.
Methods: Hence, the present study was undertaken to explore the toxic effect of the insecticide CYP on mice. This work also aimed to investigate the protective effect of Zizyp fruits against oxidative stress and neurotoxicity induced by CYP in heart, liver and kidneys of mice.. Mice were divided into four groups of six each: groups I and II were used as control and CYP control (20 mg/kg body weight). While, groups III was orally treated with Zizyphus lotus fruit (5 g/kg body weight) plus CYP (20mg/kg body weight) for 18 days. Furthermore, HPLC–ESI–MS–MS (Q-Tof) and GC–MS were used to identify the compounds fraction.
Results: Antioxidant enzyme catalase (CAT), neurotoxicity enzyme acetylcholinesterase (AChE) activities and hydrogen peroxide (H2O2), malondialdehyde (MDA) levels were determined in the liver. CYP caused decreased CAT activity, inhibition of AChE activity and increased the levels of H2O2 and MDA in liver.
Conclusion: Our results indicate that Zizyp fruit is markedly effective in protecting mice against CYPinduced biochemical changes. This protection may be due to its antioxidant property and scavenging ability against active free radicals. In addition to post-harvest investigations, further studies may determine the bioavailability and the physiological relevance of the elucidated constituents found in Zizyphus lotus fruits.
Keywords: Cypermethrin; Zizyphus lotus jujube fruits; HPLC–ESI–MS–MS; mice; oxidative stress.

BMP4: Cardioprotective effect of Hibiscus sabdariffa L. Extract (HSE), on Cypermethrine-induced oxidative stress in healthy mice

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Background and aim: Cypermethrin (CYP) is a synthetic pyrethroid insecticide used worldwide in agriculture, home pest control. The toxicity of CYP is well studied in many organisms. The aim of present study was to investigate the protective effect of Hibiscus sabdariffa L. Extract (HSE), against cardiotoxicity induced by CYP in mice.
Materials and methods: Thirty-six male mice (32–38g). Mice were randomly divided into six groups of six animals each and received daily intraperitoneal (IP) injection as follows; Group 1 (Control): mice administered with 10% ethanol during 21 days (n = 6). Group 2 (CYP): mice receiving a single dose ofCYP (20 mg/kg) at day 12. Group 3 (CYP + HSE200): mice treated both with 200mg/kg bw of HSE during 21 days and a single dose of CYP at day 12. Group 4 (HSE200): mice treated with HSE (200mg/kg bw) during 21days. Group 5 (CYP + HSE500): mice treated both with 500mg/kg bw ofHSE during 21days and a single dose of CYP at day 12. Group 6 (HSE500): mice treated with HSE (500mg/kg bw) during 21days.
Results: Antioxidant enzyme catalase (CAT), specific enzyme acetylcholinesterase (AChE) activities and hydrogen peroxide (H_2O_2), malondialdehyde (MDA) levels were determined in the heart. CYP caused a decrease of CAT activity, inhibition of AChE activity and increased the levels of H_2O_2 and MDA.
Conclusion: Our results indicate that HSE is markedly effective in protecting mice against CYP-induced biochemical changes. This protection may be due to its antioxidant property and scavenging ability against active free radicals.
Keywords: Cypermethrin, Hibiscus sabdariffa L. Extract, mice, oxidative stress.

BMP5: LC-ESI-MS analysis, inhibition of pancreatic lipase and anti-proliferative activity of *Diplotaxis Simplex*

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Background and aim: Many plant extracts have shown interesting pharmacological properties and they have become very popular as potential agents for therapeutic and or cosmetic purposes. *Diplotaxis simplex* (Viv.) Spreng (Brassicaceae), is a good source of health-promoting phytochemicals.
Methods: The current study evaluates the anti-proliferative activity of ethyl acetate and ethanol extracts from *D. simplex* flowers using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) against human breast cancer cells (MCF7) and human blood cancer cells (K562). Furthermore, the anti-obesity potential was evaluated by the inhibitory effect of extracts on recombinant human pancreatic lipase (rHPL). The chemical composition of the organic extract was analyzed by chromatography-electrospray ionization-tandem (LC- ESI-MS).
Results: The LC- ESI-MS analysis of organic flowers extracts allowed the identification of 7 phenolic acids and 9 flavonoids. Hyeroside and quercetrin present the major identified flavonoids and coumaric acid was found to be the major phenolic acid. Flowers extracts were effective in inhibiting cancer cell growth (IC_{50} lower 100 µg/mL). Besides, these extracts inhibited 50% of rHPL activity at low concentrations (0.14 and 0.32 mg/ml).
Conclusion: These data suggest that *D. simplex* may be useful as a candidate in the treatment of obesity and the breast and leukemia cancer.
Keywords: *Diplotaxis simplex*; LC-ESI-MS; anti-cancer effect; rHPL inhibition.

BMP6: Improvement of *Aeribacillus pallidus* lipase production using Box behnken design and its application in detergents formulations

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Background and aim: Lipases are derived from plants, animals and microorganisms which microbial liapses are receiving more attention, since they have a great variety of catalytic activities, are produced at high yields and do not require cofactors. The aim of this work is to improve the yield of lipase production through optimization of the fermentation medium composition for a newly thermostable lipase of Aeribacillus pallidus. Additionally, the enzyme showed excellent stability and compatibility with various commercial detergents suggesting its potential as an additive in detergent formulations.
Methods: Box-Behnken optimization, is one of the RSM designs which is more effective one that creates empirical model equations that correlate the relationship between variables and responses
Results: Lipase from *Aeribacillus pallidus* strain was produced without induction and it is a novel halophilous, thermo-alkaline and detergent- tolerant lipase (GPL).
Considerable interest has been given to this lipase by the improvement of its production. Indeed, lipase activity was tremendously improved and reached 68 U/ml by using a boxbehnkhen design. An appropriate medium was formulated for GPL production and was composed of: 1 .5g/l Glucose, 0.5g/l yeast extract, 23g/l NH_4Cl corresponding to C/N ratio of 1, 30 g/l NaCl, 1 g/l KH_2PO_4, 1 g/l $MgSO_4.6H_2O$, 1g/l $CaCl_2$ at 55°C. . In the present study, it is indicated that the carbon to nitrogen (C/N) ratio which was about 1, the pH value corresponding to 10 and the inoculums size was 0,3 were able to increase the lipase production by 6,68-fold and predict the optimal values of the selected factors. GPL exhibited excellent stability to detergents and wash performance analysis revealed that it could remove blood-stains effectively. Thus, it is a potential candidate for applications in detergent formulations.
Conclusion: GPL was reached under aerobic conditions by glucose at high concentrations, a combination between organic and inorganic nitrogen sources, pH 10 and inoculums size equal to 0,3.It could have a good capacity for removing fatt y stains in alkalin environment, suggesting its potential use as an additive in detergent formulations
Keywords: Box-B ehnken, lipase production, detergent formulation.

BMP7: Biopolymer-based polyelectrolyte complexas a novel excipient for sustained-releasematrix tablets

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Background and aim: The polyelectrolyte complexes (PECs) formed by the combination of opposing charge biopolymers proves to be a widely used method for the development of excipients for pharmaceutical use. The aim of this work is to formulate a matrix tablet based on a PEC formed from carboxymethyl-kappa-carrageenan (CMKC) and chitosan (CTS), with a view to the prolonged release ofValsartan (VAL),a principle active antihypertensive agent.
Methods: Interpolyelectrolyte complexation between CMKC and CTS was studied and characterized. The obtained polyelectrolyte complex (PEC) was used to develop matrix tablets aimed to sustain drug delivery. Valsartan matrix tablets were prepared by direct compression and characterized according to standard procedures (USP30). Drug release in phosphate buffer solution medium (PBS, pH7.4) were reported. The dissolution data were fitted to different dissolution models.
Results: The overall results indicate that, the powder beds of all formulations are freely flowable and easily compressible. The release profiles of batches shows that the release rate was greatly influenced by the concentration of PEC in the matrix. Drug release rate decreased as the PEC content increased because of increase in gel disentanglement. Indeed, the drug release rate depends on the strength of the gel barrier which depends on the proportion of the PEC, rate of hydration and viscosity. A more concentrated gel provides more tortuous and resistant barrier to diffusion. The drug release from matrix systems approaches zero order kinetics. A PEC's optimal proportion of 50 % in the tablet formula provides a dissolution profile characterized by Korsmeyer–Peppas exponent value (n) exceeding 0.99.Drug release mechanism is controlled by the superposition of diffusion and erosion. The relaxation phenomenon appears to increase along the release process and even overcomes diffusion for some systems.
Conclusion: The CTS-CMKC polyelectrolyte complex has demonstrated a high potential as an excipient for the production of swellable matrix systems with sustained drug release properties. A prepared PEC could be a possible potential hydrophilic carrier in the design and development of future oral controlled drug delivery systems.
Keywords: Carboxymethyl-kappa-carrageenan, Chitosan, Polyelectrolyte complexe, Sustained-release, matrix tablets.

BMP8: A review on chemistry and biological activities of essential oil of *Laurus Nobilis* L. wild-growing in Algeria

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Background and aim: The food industry is constantly challenged by consumers seeking for increasingly healthy foods and less use of synthetic chemical additives such as pesticides or preservatives. In this context the aim of the present work was to study the antifungal activity of Laurel essential oil in different treatments against two phytopathogenic fungus *Aspergillus niger* and *Fusarium oxypouim*. These strains are among the most contaminating genera of mycotoxin dry and mainstream producers.
Methods: The extraction was carried out by hydrodistillation and molecular characterization by GC / MS. The direct contact method has been applied to test the sensitivity of molds to essential oil.The potential antioxidant activity of the essential oil from *Laurus Nobilis* L. were determined by employing two complementary tests, namely DPPH free radical scavenging and b-carotene/linoleic acid test systems.
Results: The average yield of essential oil extracted from the plant is studied in the range of 0.8%. The chromatographic analyses resulted in the identification of 25 compounds, representing 97.49 % of the oil, 1,8-Cineole (49.43 %), Terpinyl acetate (14.90 %)and Sabinene (09,88 %) were the major components.
Conclusion: The essential oil extracted from the leaves of *Laurus Nobilis* L. grown in the region of Azzaba, Skikda city (Algeria), showed, *in vitro*, an antifungal activity against both fungi; *Aspergillus niger* and *Fusarium oxypouim* tested. This bioactive power observed in oil is attributed mainly to their contents of 1,8-Cineole, Terpinyl acetate and Sabinene known for their inhibitory properties of mycelial growth.
The Results obtained confirm that the good antioxidant potential of the essential oil of this plant according to others. These results keep an open perspective for research of formulations on the basis of essences of the *LaurusNobilis* L. in place of other synthesis preservatives or antioxidant on the basis of plant used in the field of food industry, pharmaceutical and cosmetics industry.
Keywords: *Laurus Nobilis* L., essential oil, chemical composition, antifungal activity, antioxidant activity.

BMP9: *In vitro* inhibitory potency of the essential oil of *Pelargonium graveolens* L. from Skikda region (northeastern Algeria) with respect to: *Fusarium oxysporum* f.sp. and *Alternaria* sp.

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Background and aim: The food industry is constantly challenged by consumers seeking for increasingly healthy foods and less use of synthetic chemical additives such as pesticides or preservatives. In this context the aim of the present work was to study the antifungal activity of Geranium Rosat (*Pelargonium graveolens* L.) essential oil in different treatments against two phytopathogenic fungus; *Fusarium oxysporum* f. sp. and *Alternaria* sp. These strains are among the most contaminating genera of mycotoxin dry and mainstream producers.
Methods: The extraction was carried out by hydrodistillation and molecular characterization by GC/MS. The direct contact method has been applied to test the sensitivity of molds to essential oil.
Results: *Pelargonium graveolens* L. essential oil was analyzed by (GC/MS), the major components were citronellol (29.2 %), citronellylformate (10.3 %), geraniol (6.6 %) geranyl formate (5.2 %) and linalool (4.6 %) they constituted 55.9 % of total essential oil. The essential oil extracted from the leaves of *Pelargonium graveolens* L. showed in vitro, an antifungal activity against both fungi; *Fusarium oxysporum* f.sp. and *Alternaria* sp. tested.
Conclusion: The bioactive power observed in oil is attributed mainly to their contents of citronellol and citronellylformate known for their inhibitory properties of mycelial growth. These results keep an open perspective for research of formulations on the basis of essences
of the *Pelargonium graveolens* L. in place of other synthesis preservatives or antioxidant on the basis of plant used in the field of food industry, pharmaceutical and cosmetics industry.
Keywords: Geranium Rosat, *Pelargonium graveolens* L., essential oil, chemical composition, antifungal activity.

BMP10: Rheological properties of EPS of LAB

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Background and aim: Lactic acid bacteria (LAB) are known through ages for their wide applications in food, pharmaceutical and chemical industries. But recently, LAB has aroused interest for their ability to secrete extracellular polysaccharides or "exopolysaccharide" (EPS). This substance is economically important because it can impart functional effects on rheological and sensory properties of fermented products and conferred beneficial health effects. The aims of this study is to isolate and screen EPS producing strains of CLAB obtained from camel's milk and fresh red meat of Algeria, in order to evaluate their capacity to produce these EPS and select the most performing strains.
Methods: All the isolates used in this study were evaluated for EPS production on certain solid medium and ruthenium red milk agar plate. Quantitative estimation of EPS was realized by measurement of apparent viscosity and determination of sugar and protein total in these EPS, previously, extracted and purified from the various culture strains.
Results: Based on their EPS-producing colony phenotype in agar plates, five strains were chosen giving an important white-color with different capacities of production and a glistening and slimy appearance on sucrose-based media which being the best for detecting the EPS. The amount of sugar in the polymer rendered more than 400 mg/L and the apparent viscosity ranged from 2.1 to 2.9mPa.s. Therefore, no clear-cut relationship between the slimy phenotype, the medium viscosity occurs and amount of EPS produced by a ropy strain when different strains were compared. For protein assay, a low content of protein was obtained on crude polymer revealing the quality of EPS extracts.
Conclusion: Many CLAB stains were screened for the ability to produce EPS using the qualitative and quantitative analysis. Three strains were selected for their significant production of EPS, yielded an important amount of polysaccharide.
Keywords: CLAB- EPS- sucrose-slimy appearance - apparent viscosity- amount of EPS.

BMP11: Quality by Design approach for encapsulation of natural drugs: optimization of polydispersity and volume/surface parameters

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Background and aim: Due to their multitude bioactivities and applications from food flavor to pharmaceutical and cosmeceutical industry, the formulation of natural drugs such as essential oils (EO) has been highly studied. In aim to facilitate the semiindustrial scaling-up, Design & Development of EO micro-carriers by Ionic Gelation, recommended for hydrophobic bioactive molecules, has been applied and optimized to ensure a high encapsulation yield with response stability.
Methods: EO was microencapsulated by cross-linking a biodegradable polymer under several experimental conditions (*Graphic below*) and the process optimization was carried out using the **RSM** to investigate the 4 experimental parameters. To compare Polydispersity Indexe (***PI***) mean, volume-weighted (***d*43**) and surface weighted (***d*32**) means and particle diameters, ***QbD*** approach based on statistical analysis was performed for the triplicate using **one-way analysis of variance (ANOVA)** and ***T-test***.
Results: Both the loading capacity and the particles size of the obtained microcapsules were evaluated to optimize the ionic gelation process. Laser diffractometry was used to assess the physical characteristics of the developed microcarriers. The selected optimal conditions allow obtaining microparticles with loading capacity of 5 to 15 % with a size range from 0.19 µm to 377 µm, ***PI*** range from 0.852 to 5.695, a ***d32*** of 0.69 to 470 µm and a ***d43*** from 7.39 to 715 µm. According to RSM results combined with the statistical analysis and the range on significant or not significant effects (*P-value*), the most influencing parameter for SPAN is the mixing velocity.
Conclusion: The objective ofthis study was to apply the Quality by Design approach to the development of microcarriers for EO encapsulation using alginate microspheres allowing an interesting loading capacity, acceptable particles size, desired polydispersity and suitable volume/surface characteristics. The designed microencapsulation process is statistically validated and can be easily scaling-up to a semi-industrial level.
Keywords: Natural drugs encapsulation, Quality by Design approach, Polydispersity Indexe, Hydrophobic bioactive molecules, Statistical analysis.

BMP12: Formulation of pharmaceutical lipidique microparticles for encapsulating a nonsteroidal anti-inflammatory drug

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Background and aim: Diclofenac is a nonsteroidal anti-inflammatory and antipyretic drug, because ofits short half-life andits gastrointestinal disturbances due to its repeated administration we have considered sodium diclofenac as a good candidate for microencapsulation.Microencapsulation offers industrial technological solutions when we need to protect the active ingredient or improve its presentation and control its release profile.Stearic acid has neuro-protective effects on brain due to retarded oxidative stress. Hence, it will be interesting to use stearic acid as the matrix material for preparing on solid lipid particles for the delivery of Sodium Diclofenac.
Methods: The aim of the current work is the development of a microparticule system based on solid lipid particlesto encapsulate Diclofenac with a maximum encapsulation yield and a minimum particles size. Various formulations were prepared using Box-Behnkendesign to find the best conditions. The studied parameters are the stirring speed, the amount ofthe oily phase and the concentration of surfactant.
Results: The observation of the particles by a microscopedemonstrate that the particles have an irregular and almost a sphericalshape which confirms the encapsulation of diclofenac. The best encapsulation yield that we can reach is 83.6%with the smallest particle size of 0.427µmwere obtained with a quantity of 2g of oily phase, 4% w/v for the concentration tween 80 and a stirring speed of 800 rpm.We have also noticed that the studied factors have a significate influence on the yield and the size.
Conclusion: The results obtained from the statistical analysis allow us to distinguish the zone of the experimental domain that make possible the optimization the microencapsulation process and predict the results.
Keywords: Encapsulation, Sodium Diclofenac, Optimization,Box-Behnken design.

BMP13: Synthesis of hydroxytyrosol and tyrosol derivatives a high added-value compounds through 2-phenylethanol conversion by catalytic synthesis

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Background and aim: Productions and valorizations of bioactive molecules by catalytic oxidations constitute an original approach compared with chemical productions. Due to KSF ability to catalyze a variety of complex oxidations, it was likely to convert 2-phenylethanol to hydroxytyrosol and tyrosol derivatives. The best time to obtain the highest content of HT was 8h.
Methods: All aromatics compounds were determined and quantified by high performance liquid chromatography HPLC then the identification by mass spectrophotometry using LCMS apparatus and using standards.
Results: Optimization of the main experimental parameters to obtain the best conversion was with an initial concentration of 2-phenylethanol 10^{-2}M, a concentration of $_{H2O2}$ 5.05 10^{-2}M and a quantity of catalyst 0.6 g / L. All compounds in reaction mixture l'*ortho*-tyrosol, *méta*tyrosol and *para*-tyrosol as well as l'hydroxytyrol were determined by spectroscopic techniques such as (UV- HPLC and LC-MS). In the following section the kinetic of conversion revealed that 2-PhEt oxidation by hydroxyl radicals is of a first order pertaining to the 2-PhEt concentration. The antioxidant activity was realized by DPPH method revealed that catalyst solution mixture exhibited a higher antioxidant activity (PI= 81%) at a similar level to that of BHT (PI=82%), which clearly indicates the presence of phenolic mixture rich in monohydroxy compounds, such as TY and dihydroxy compounds such as hydroxytyrosol. This result is also confirmed by the concentration of ortho-diphenol which reveals that after 8 hours of conversion reaction, the higher level of the orthodiphenol was caused by the presence of hydroxytyrosol.
Conclusion: The synthesis of tyrosol and hydroxytyrosol by hydrogen peroxide catalytic oxidation of 2-PhEt using acid-activated clays offering a promising alternative to other previously published methods dealing with unnatural antioxidant invention. This process is very simple, cheap and fast.
Keywords: Catalytic wet oxidation, hydrogen peroxide, 2-phenylethanol, tyrosol, hydroxytyrosol.

BMP14: Evaluation of the synergistic effect of *Daucus carota* honey and essential oils against *Candida albicans*

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Background and aim: The resistance of pathogenic fungal strains to the commonly used antifungal has necessitated a search for novel types of antifungal agents. The main objective of this study was to investigate the synergistic effect of *Daucus carota* honey and essential oils of *Eugenia caryophyllata*, *Mentha pelugium* and *Geranium* against *candida albicans.*
Methods: The aerial plant parts (leaves and flowers, 30 g) of *Menthapelugium* and the flower buds of clove (*Eugenia caryophyllata*) were dried at room temperature, hydro distilled for 3 h using a Clevenger type apparatus (British Pharmacopoeia, 1998) *Geranium* EO which was purchased from Algeria market. In the first step the antifungal activities of essential oils (EOs) of medicinal plants and wild carrot *(Daucus carota*) honey has been determined by the measurement of Minimum Inhibitory Concentration (MIC) using an agar incorporation technique method and in the second step the Minimum Synergistic inhibitory Concentration (M SIC) of honey and essential oils has been determined using the same method.
Results: The results of our study indicate that the EOs and honey are efficient against the tested strain. Honey MIC value was 6% (vol/vol), whereas the MIC values of EOs were 0.4µl/ml for both *Geranium* and *Mentha peligium* EOs (vol/vol) and 0.33 µl/ml for *Eugenia caryophyllata* EO. When honey and EOs are used jointly, we noticed a decrease of the MIC values which is may be due to their synergistic effect.
Conclusion: These preliminary results suggest that honey and EOs could be used together to manage superficial fungal infections.
Keywords: Honey, Essential oils, Antifungal activity, Synergy.

BMP15: Opposite-action of *Opuntiaficus-indica* L. juice and seeds aqueous extract on gastric emptying and small-bowel motility in rat

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Background and aim: *Opuntia ficus indica* L., generally used in folk medicine in the treatment of gastrointestinal (GI) disorders, is known for its richness in important nutrients for functioning of the digestive tract such as high content of fibers, minerals, vitamins, fatty acids, sugars, polyphenols, and flavonoids.
Methods: Separate-groups of rats were randomized to receive NaCl, increasing doses of juice or seeds aqueous extract. Simultaneously, two other groups were received, the reference drugs; clonidine and yohimbine. The charcoal meal was used as a suspension for gastrointestinal motility test. The purgative action of juice was confirmed using the loperamide-induced constipation. To evaluate the antisecretory effect, we were used as a hypersecretion agent, the castor oil. Bioactive components were analyzed using biochemical methods.
Results: Compared to the control and reference-groups, we were showed that the prickly pear (juice and seeds) has a reverse effect on small bowel motility and gastric-emptying. Indeed, the juice at various doses (5, 10, and 20mL/kg, b.w.) has a laxative effect of gastrointestinal transit in healthy and constipated-rats. However, the aqueous extract of the seeds (100, 200, and 400mg/kg, b.w.) leads to a decrease of motility in normal rats which gives it a remarkable antidiarrhoeal-activity, a notable intestinal fluid accumulation decline and electrolyte concentrations reestablishment. Moreover, orally juice administered at different doses accelerated the stomach emptying time in contrast to the seeds aqueous extract. More importantly, a significant variation in the phytochemical compounds levels between juice and seeds was found.
Conclusion: Bioactive components of prickly pear may be used in phytomedicine as nutraceuticals and functional foods to regulate and to attenuate the gastrointestinal physiological activities and disturbances.
Keywords: *Opuntia ficus-indica* L., Juice, Seeds aqueous extract, Gastric emptying, Smallbowel motility, Functional-foods, Rat.

BMP16: Study of the prevalence and expression of efflux pumps in *Pseudomonas aeruginosa* and *Acinetobacter baumannii* isolated from diabetic foot infection

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Background and aim: In front to polymorphic bacterial ecology and antibiotic resistance in diabetic patients with foot infections and good patient care, collaboration between clinicians and microbiologists is needed to improve assessment and management of patients with this pathology. This study was conducted in order to determine the bacterial resistance profile and the mechanism of resistance (pump efflux) from patients with this infection.
Methods: In this work, we determined the bacterial ecology responsible for diabetic foot infection in 117 patients (irrespective of age, sex, and type of diabetes) and the resistance profile of these isolates to antibiotics; we also looked for the expression of efflux pump with the Q-rt PCR method in non-fermenting Gram negative bacteria as a mechanism involved in this resistance.
Results: This study has shown a high prevalence of Gram-negative (6 1%) generally presented by species *E.coli*, *P.aeruginosa*, *A.baumannii*, and other *Enterobacteriaceae*, and (3 9%) Gram positive mainly presented by *S.aureus*. The resistance profile reveals that 93.47% of Enterobacteriaceae are resistant to β-lactams, and 93.33% of *Staphylococci* to penicillin G, and 71.66% to tetracycline, and the majority of non-fermenting Gram negative bacteria are resistant to fluoroquinolones. However, 70% of A*cinetobacter baumannii* and 41.66% of *Pseudomonas aeruginosa* so that it appeared with a positive phenotype ofthe efflux pump expression.
Conclusion: Our study found that from diabetic foot infections, a high level of multidrug-resistant non-fermenting Gram-negative bacilli by phenotypic overexpression of efflux transporters has been observed, which increases the risk of infection and causes the therapeutic failure. These results should be taken into account by the clinician in the prescription of probabilistic antibiotic therapy in this context.
Keywords: antibiotic, mechanisms of resistance, efflux pumps, diabetic foot infection.

BMP17: Quantitative phytochemical analysis of *Medicago ciliaris* L. populations over two years

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Background and aim: Plant secondary metabolites are highly and naturally diverse in their chemical structures and abundances. The analysis of these compounds in forage and pasture species is a key factor determining both animal health and products. The present study through some light on the possible medicinal values of *Medicago ciliaris* L. as it is widely distributed forage and pastoral legume. Its curative proprieties can be attributed to the presence of secondary metabolites such as saponins, tannins, phenolic compounds etc.
Methods: The quantitative phytochemical analysis was carried out on dry biomass of eight local populations of *M. ciliaris* L. to determine the secondary metabolites (total phenols, saponins and tannins) during two years.
Results: Analysis of variance applied on secondary metabolites revealed a significant effect of the year of characterization on total phenols, tannins and saponins contents and significant effects of populations and year*populations interaction on tannins content. A strong and positive correlation was found between tannins and saponins content (r=0.72; n= 46; P<.0001) The principal component analysis showed five groups of populations: on PC1, the population of Ghar El Melh which is the richest one for secondary metabolites (5.1 µg ac tannic of total phenols, 2.83 mg/ac tanic of tannins and 7.37g dios/kg DM of saponins) opposed the poorest population of Zana (3.84 of total phenols, 1.70 mg/ac tanic of tannins and 5.28 g dios/kg DM of saponins) while on PC2, the population of Kalaat Andalous with high saponin content (6,13 g dios/kg DM) opposed the populations of Raoued and Dougga with low saponin values (5.43 g dios/kg DM, 6.25 g dios/kg DM respectively). The rest of populations expressed intermediate values for all the analyzed secondary metabolite. Cluster analysis gathered populations of *Medicago ciliaris* into three main classes and confirm the divergence of population of Ghar El Melh from the rest of the populations thanks to its richness in secondary metabolites.
Conclusion: Since *M. ciliaris* contains secondary metabolites at varying quantities it can be used as medicinal plants. Furthermore, the analyzed secondary metabolites seemed to be highly affected by environmental conditions.
Keywords: *Medicago ciliaris,* phytochemical analysis, secondary metabolites, PCA.

BMP18: Probiotic *Leuconostoc mesenteroides*: *in vitro* and *in vivo* assessments

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Background and aim: To fight pathogenic bacteria, fishes have been protected by vaccination or chemotherapy treatments. However, aggressive use of chemotherapeutic agents may leads to progressive pathogen resistance (Balcázar et al. 2008, Merrifield et al. 2010). As an alternative to antibiotics, the use of lactic acid bacteria probiotics (LAB) in aquaculture has shown promise to overcome issues related to disease control. Recently, we isolated and identified a LAB strain as *Leuconostoc mesenteroides HG93 7700 (El Jeni et al., 2015; 2018).* The aim of this study was dual: (i) evaluating the resistance potential of the LAB we isolated to human GI tract conditions and (ii) assessing the *in vivo* probiotic effect on Nile tilapia (*Oreochromis niloticus HG93 7700).*
Methods: The in vitro probiotic potential was carried throughout assays of: (i) acid tolerance (ii) bile salt hydrolase and bile salt tolerance (iii) Bacterial resistance to pepsin, pancreatin, and heat resistance (iv) Hemolytic activity (v) Antimicrobial agents assay (vi) Bacterial adhesion to stainless steel plates according to the previously well described methods (Tuomola et al. 2001; Ouwehand et al. 2002; Da Cruz et al. 2007). For assessing *in vivo* probiotic effect on Nile tilapia (*Oreochromis niloticus HG93 7700),* fishes were fed 20 days with food, in presence or absence of *L. mesenteroides* isolated strain *HG93 7700*
Results: Interesting, data obtained showed that *Leuconostoc mesenteroides HG93 7700* have significant probiotic potential, since it survived at pH 3.0 and in the presence of bile salts, pancreatin, and pepsin, without any detectable haemolytic activity. Further, moderate heat resistance, adhesion ability to steel surfaces, and sensitivity to clinically relevant antimicrobial agents were revealed for this LAB isolate.
More interestingly, the strain *Leuconostoc mesenteroides HG93 7700* supplementation improved the fish growth and weight. Thus, protein content has been positively increased and remarquable lysozyme activity was observed for the farmed fish and demonstrated positive adaptation of *Oreochromis niloticus HG93 7700* to experimental diets.
Conclusion: These interestingly results highlight the specific probiotic properties of *Leuconostoc mesenteroides HG93 7700* and give evidence for eventual potential application farmed fish feeding aim. Further investigation should be done in order to improve such probiotic use in moderate heat processed food.
Keywords: Freshwater fish, lactic acid bacteria, probiotic, *in vitro* and *in vivo* assays.

BMP19: Effect of olive mill wastewaters on lipid and antioxidant molecules production by *Scenedesmus* sp.

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Background and aim: The conventional olive mill wastewater (OMW) treatment methods adopted in Tunisia are either costly or ineffective, causing environmental pollution. The aim of this work is the treatment of olive wastewaters by the green microalga *Scenedesmus* sp. and the investigation of OMW, OMUF and tyrosol (Tyr) effects on growth, biochemical and physiological composition of *Scenedesmus* sp. during single and two stages cultivation.
Methods: Our investigation evaluated the lipid, protein, carbohydrates, polyphenols and pigment contents and antioxidant activity of ethanol extract using both DPPH and ABTS assays.
Results: During two stage cultivation of *Scenedesmus* cells, highest lipid and polyphenol content were obtained under 40% OMW and 40% OMUF. During single stage cultivation, the protein and carbohydrate content were the highest in 40% OMUF, 80% OMW and 200 mg/L Tyr grown culture, along with higher biomass compared to two stages, making it an efficient strategy to enhance biofuel production potential of *Scenedesmus* sp. This study allows us to obtain high antioxidant activity using two tests; DPPH and ABTS. The phenolic extracts of *Scenedesmus* sp., has an important antioxidant capacity. Oxidative stress was induced in *Scenedesmus* strains when exposed to increasing T, OMUF and OMW concentrations, as evidenced by increased malondialdehyde content. Activities of superoxide dismutase and catalase were mainly inhibited while GPx increased, at all concentrations, indicating that OMW, OMUF and Tyr-induced oxidative damage after short and long exposure. *Scenedesmus* sp. exhibited high polyphenol removal efficiency, reaching 54, 58 and 95% at 200 mg/L of Tyr, 80% OMUF and 80% OMW. BPA removal was mostly attributed to biodegradation. These results were confirmed by HPLC analysis of polyphenols.
Conclusion: These results demonstrate that *Scenedesmus* sp., can be used as a promising renewable feedstock for biodiesel production and antioxidant molecules production.
Keywords: *Scenedesmus* sp., lipids, polyphenols, OMW, OMUF.

BMP20: Optimisation of C-phycocyanin synthesis by response surface methodology from *Phormidium versicolor* (NCC466) and their antioxidant and cytoprotective activities

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Background and aim: *Phormidium versicolor* is unicellular cyanobacteria isolated from Sfax solar saltern (Sfax) rich in phycocyanin (C-PC). This pigment (C-PC) is the association between phycobiliprotein and water-soluble pigments of photosynthesis. Recent studies have shown that phycocyanin have an excellent therapeutic properties, including antioxidant, anti-inflammatory, hepatoprotective activities, nephroprotective activities and anti-cancer activities. The present study was assessed to evaluate the effects of sodium nitrate, calcium chloride and citric acid on C-PC production by Phormidium versicolor. Moreover, this work aimed to determine the antioxidant activities and cytoprotective potential of Phormidium versicolor phycocyanin (C-PC) against cadmium toxicity in renal cells HEK293.
Methods: The antioxidant activity of C-PC was evaluated against DPPH **(**2,2-diphenyl-1- picrylhydrazil), ABTS (2,2'-azino-bis(3 -éthylbenzothiazoline-6-sulphonique) and NO radicals. The cytoprotective potential of C-PC isolated from *P. versicolor* against cadmium induced oxidative damage and alteration in renal cells HEK293. The cells viability was determined using MTT test (3 -(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium).
Results: Our results showed that the optimum C-PC production (0.768 $mg.mL^{-1}$) was recorded on 5 $mg.mL^{-1}$ sodium nitrate, 0.5 $mg.mL^{-1}$ calcium chloride and 30 $mg.mL^{-1}$ citric acid, respectively. The P-PC exhibited a strong antioxidant activity. The protective effect of C-PC on cadmium (Cd)-induced damage was investigated in HEK293 cells. The C-PC did not cause any effect on HEK293 cell line within the range concentration tested (0-150 $mg.mL^{-1}$). Thus treatment with 25 $mg.mL^{-1}$ of C-PC protects HEK293 cells against toxicity induced by cadmium intoxication.
Conclusion: The phycocyanin fraction isolated from *P. versicolor* showed significant antioxidant and cytoprotective potentials which could be related to active compounds present in this pigment.
Keywords: Antioxidant activity, Cadmium, HEK293 cells, Optimisation, *Phormidium versicolor* phycocyanin.

BMP21: Screening of Antibacterial and Antifungal Activities of Ten Extracts of *Stipa parviflora*

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Background and aim: Treatment failures and the higher cost of treatment of infections caused by resistant bacteria constitute a major medical problem worldwide and poses a big threat to human society. Our work is inscribed in the frame of finding of new natural antimicrobials agents and to determine other care alternatives.
Methods: Antimicrobial Activity of Hexane, ethyl acetate, ethanol and methanol extracts of *Stipa parviflora* has been tested against 12 pathogenic multidrug resistant microbial strains: three Gram$^-$(*Escherichia coli, Salmonella enterica, Pseudomonas aeruginosa*) three Gram$^+$(*Bacillus anthracis, Listeria ivanovii, Staphylococcus aureus*) and six fungi (*Aspergilus niger, Candida albicans, Fusarium aluminium, Fusarium ox, Fusarium phylophylom* and *phytium sp.)*. The susceptibility of microbial strains against different extracts was determined using the disc diffusion method. Zone of inhibition produced by different extracts against the test bacteria was measured and compared with standard antibiotic disc.MIC (minimum inhibitory concentrations) and MBC (minimum bactericidal concentrations) were determined for the extract that presented the highest activity in the agar disc-diffusion test.
Results: In the exception of aqueous extracts, all other extracts have an interesting activity, with inhibition zone diameters between 5 and 35 mm. The minimum inhibitory concentrations (MIC) of organic extracts against sensitive microbial strains ranged from 25 mg/mL and 100mg/mL. The study of minimum bactericidal concentrations (MBC) has highlighted the bacteriostatic effect of all extracts.
Conclusion: Among the different extracts, ethanolic extracts were shown to be themost, active on the different tests. *Stipa parviflora* could be a potential candidate for future development of novel broad spectrum antimicrobial herbal formulation.
Keywords: *Stipa parviflora,* extracts, Antibacterial activity, antifungal activity**,** *MIC, MBC.*

BMP22: Antioxidant and hepatoprotective effects of flavonoids-rich fractions from *Fumaria officinalis* in permethrin induced damage liver "*ex vivo*"

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Background and aim: Permethrin (PER), the most frequently used synthetic Type I pyrethroid insecticide in the world might have a variety of toxic effects such as neurotoxicity, immunotoxicity, cardiotoxicity, cytotoxicity as well as hepatotoxicity. A various number of works indicate that oxidative stress played important roles in the various toxicities associated with PER. The aim of this work is to evaluate the antioxidant and hepatoprotective effects of flavonoids-rich fractions from *F. officinalis* against permethrin induced liver damage *ex vivo*.
Methods: Flavonoids-rich fractions of F. officinalis was tested for total phenolic and total flavonoid content, *in vitro* antioxidant activity by DPPH and FRAP assay, and these hepatoprotective effects against permethrin incubation of liver after 60 and 90 min.
Results: Flavonoid rich fractions were isolated by solvent fractionation from plant: diethyl ether fraction (DEF), ethyl acetate fraction (EAF), n-butanol fraction (NBF) and aqueous fraction (AF) were examined. These fractions possessed potent DPPH and FRAP antioxidant power. Total phenolic and flavonoids content of n-butanol fraction were significantly higher than other fractions.
Incubation of the liver with permethrin induces hepatic dysfunction demonstrated by a decrease in transaminase (AST and ALT) and TBARS levels and SOD and GPx increase. Co- incubation of F. *officinalis* fractions with permethrin treated liver showed that ethyl acetate (EAF) and n-butanol fractions (NBF) possessed the most potent hepatoprotective effects against insecticide induced damage.
Conclusion: This study provided scientific evidence on the hepatoprotective potential of fractions of F. *officinalis*, which may be due to their high flavonoid content.
Keywords: Oxidative stress, Permethrin, Flavonoid, *Fumaria officinalis*, liver.

BMP23: Implication of oxidative stress in deleterious nitrite- induced alterations in an experimental rabbit model

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Background and aim: Nitrite is a widespread anion in the environment, also found in the body as a result of endogenous production, consumption from dietary sources such as cured meat and vegetables, or exposure to environmental toxicants. Previously, nitrite was believed to be an end product of NO without any biological effects. However, it has been shown to act as an endocrine reserve of NO that, under certain conditions, exerts potent biological effects.*In vivo* investigations of pathophysiological effects of sodium nitrite supplementation showed that they seem to be dependent on the used animal models and the employed experimental conditions. The aim of our study is to investigate the effect of high exogenous nitrite intake in rabbits with the possible protective effect of Gingko biloba extract (EGb761).
Methods: In this context, NaNO2 (100 mg/kg/day) was orally administrated to New Zealand white rabbits alone or concomitantly with EGb761 for 6 weeks. Total "oxidant/antioxidant" balance as well as Liver and renal functions were analysed by measurement of biochemical parameters and histopathological assessment.
Results: $NaNO_2$ treatment induced a significant increase inserum aspartate aminotransferase and alanine aminotransferase. Total bilirubin levels were not altered, and neither were uric acid, urea or creatinine. Histopathological changes included hydropic degeneration and inflammatory infiltration localized to portal areas associated with moderate portal-portal bridging fibrosis in liver, and focal interstitial inflammation and interstitial fibrosis but no alteration in glomeruli in kidney. The supplementation of EGb761 improved symptoms in both liver and kidney.Nitrite group showed a significant increase of the oxidant markers (AOPP, oxidized albumin, MDA). EGb76 1 supplementation reduced the levels of the three markers and increased significantly the total antioxidant capacity either alone or in association with nitrite in comparison to control group.
Conclusion: In conclusion, the results showed deleterious effects of sodium nitrite on both liver and kidney, without functional impairment of the kidney. These effects were mediated by the induction of oxidative stress witch are attenuated by the administration of Gingko biloba extract in the experimental rabbit model.
Keywords: Nitrite, nitric oxide, liver, kidney, oxidative stress.

BMP24: Comparison of Effects of Simvastatin Versus Atorvastatin on Oxidative Stress in Patients With Acute Coronary Syndrome

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Background and aim: Several studies showed that plasma markers of oxidative stress are increased in coronary heart disease or in the presence of its classical risk factors (Stephens et al., 2008). In vitro, many adverse effects on the vascular system are associated with an increase in oxidative stress (Stephens et al., 2006). However, few studies have evaluated the effects of the therapeutic statin on oxidative stress. The aim of our study was to compare the effects of simvastatin versus atorvastatin on oxidative stress in patients with acute coronary syndrome (SCA) in the long-term.
Methods: Fifty-four coronary patients were treated with atorvastatin 40 mg for 18 months and 42 coronary patients were treated with simvastatin 40 mg for 18 months. Blood samples were taken at admission, after 6, 12 and 18 months. Lipoperoxidation was measured by the malondialdehyde (MDA) assay and the total activity of superoxide dismutase (total SOD) was determined according to the method of Mirsa and Fridovich (1972).
Results: Compared to simvastatin, atorvastatin reduced significantly serum MDA levels ($p < 0.01$) with a decrease of -70.05% vs -40.55%. However, atorvastatin showed a more dramatic increase in total SOD activity (82.08%) compared with simvastatin (67.68%). Percentage changes in MDA and SOD along 18 months of study showed a significant difference between the effect of atorvastatin and simvastatin on both markers.
Conclusion: In conclusion, the comparison of the impact of atorvastatin and simvastatin showed that these two statins exert an antioxidant protection or they are not equipotent with respect to the oxidative status of patients with acute coronary syndrome.
Keywords: Acute coronary syndrome, Atorvastatin, Oxidative stress, Simvastatin.

BMP25: Analysis FTIR of kidney stone and *in vitro*study the effect of the aqueous extracts of *Paronychia argentea l.* on dissolution of the kidney stone (Uric acid anhydrous, calcium oxalate)

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Background and aim: The urolithiasis is a common pathology that effects of 4-20% of the population in world. In this study, we have evaluated the *in vitro* effect of aqueous extracts of the plant *Paronychia argentea* L. on the dissolution two type of kidney stones.
Methods: Our work is based on kidney stones weighed 1.80±5 and 3.44±3 g which differ by their morphological type; this morpho-constitutional is evaluated by a qualitative chemical analysis and spectroscopic IR analysis for each kidney stone placed in the dissolution test. These two stones put in contact with aqueous extracts (120g/l) of this plant for a period for eight weeks; after each week, we evaluated: weight of stones, the pH of extract, the rate of dissolution (%) and analysis of some ions: calcium and magnesium, then we was performed to characterization of saponins in aqueous extracts of this plant.
Results: The results showed that the plant is characterized by a high content of saponins type oleanane. This plant has an important effect and a very significant dissolution the kidney stone of mineral-organic type (anhydrous uric acid) compared the kidney stones of mineral type (oxalate /calcium phosphate).
Conclusion: Our results show that *Paronychia argentea L.* has a significant effect dissolution the kidney stones of anhydrous uric acid by loss of mass the 40 % compared the kidney stone of calcium oxalate. This effect is due to the presence of high levels of saponins the type of oleanane in this plant by the formation of complex soluble oleanane-uric acid anhydrous.
Keywords: Kidney stone, aqueous extract, *Paronychia argentea L.*, *In vitro*, Saponoside oleanane, Uric acid anhydrous, Oxalate calcium.

BMP26: Application of YLLIP2 in leather degreasing

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Background and aim: Skins and hides contain regions of natural fat. In fact, excess fat needs to be reduced during the leather manufacturing process in order to achieve a satisfactory finish of the final leather product. Degreasing of hides and skins is currently accomplished by use of organic solvents and surfactants. Detergents used in degreasing process have a good effect but they present a pollution problem due to its difficult degradation. However, enzyme is a green, efficient and easy degradable substance that can be applied in leather manufacturing. In this context, a lipase from *Yarrowia lipolytica* has been applied in sheepskins and hides degreasing.
Methods: The sheepskins and hides were incubated with YLLIP2 in an aqueous solution under pH 8 during one hour.
To evaluate enzymatic degreasing, two parameters were studied:
The difference in fresh mass which is determined by the percentage of weight loss of degreasing was measured after one hour of incubation ofthe sample with YLLIP2.
Content of residual lipids: Fat content was determined by using the Soxhlet extraction method.
Results: Our results showed a reduction in the content of residual lipids in the sheep skin and hides, from 9% to 5% and a 50% weight loss after one hour enzymatic treatment by YLLIP2 in an aqueous solution under pH 8. This enzymatic process improves an efficient degreasing of ovine skins and reduction in the use of other chemicals and surfactants in a maximum way.
Conclusion: In the current study, an extracellular lipase YLLIP2 from *Yarrowia lipolytica* demonstrated an efficient sheepskins degreasing. This enzymatic process of sheepskins can be used in order to substitute conventional one with detergents and surface-active agents. Overall, the findings indicate that YLLIP2 is bestowed with a number of promising properties that may be considered as potential candidate for ecological degreasing of skins and hides in leather manufacturing.
Keywords: Enzyme, Lipase, *Yarrowia lipolytica*, Degreasing, Leather, Lipid, Detergent.

BMP27: Antioxidant, Haemolytic activities and HPLC-MSN characterization of phenolic compounds from needles of cade

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Background: Phenolic compounds are bioactive molecules exhibiting a lot of scientific attention due to their multiple biological activities. This study aims investigating the antioxidant and the phenolic content in four different extracts (methanol, water, hexane and dichloromethane) of needles of *Juniperus oxycedrus* subsp. *oxycedrus*.
Methods: The extraction was carried out by successive exhaustions of the part of the cade, by three solvents of increasing polarity: hexane, dichloromethane and methanol. A quantification of the phenolic compounds by a spectrophotometer. Evaluation of antioxidant activity is recommended by two tests: DPPH and FRAP (Chaouche et al., 2015). All these tests are determined by parameter IC50. A low IC 50 value corresponds to an activity of interest. The haemolytic activity of the various extracts was measured.
Results: The results showed that the methanol extract was the most concentrated in total phenolics (58.8 ± 1.08 mg GAE.g^{-1} DW), flavonoids (16.2 ± 1.20 mg CE.g^{-1} DW) and tannins (19.05 ± 1.81 mg CE.g^{-1} DW). HPLC-DAD-ESI-MS^n analysis of this extract led to the identification of 27 molecules, among them hydrolizable tannins (proanthocyanidin oligomers), glycosylated flavonoids, biflavones and a furanone glucoside (psydrin). Moreover, the methanol extract exhibited remarkable antioxidant activity. Incubation of the extracts (20 mg/ml) with human erythrocytes for one hour led to haemolytic activities between 1.95% and 4.71%.
Conclusion: Our findings identified the appropriate solvent for extracting phenolics which might provide a rich source of natural antioxidants as food additives replacing synthetic ones in food industry.
Keywords: *Juniperus oxycedrus*, needles, antioxidant activity, quantification of phenolics, HPLC-SM.

BMP28: Screening for antimicrobial metabolites in the extract of the mushroom *Armillaira* sp.

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Background and aim: The spread of antibiotic-resistant pathogenic microorganisms continues to threaten public health. To fight against this phenomenon, the discovery of new antibiotic molecules is necessary. According to the bibliography, mushrooms are reservoirs of antimicrobial and antibiotic molecules. Our aim is a screening for antimicrobial molecules in the mushrooms of the genus *Armillaria*. At our knowledge, little work on their antimicrobial activity has been reported.
Methods: In the practical part of our study, basidiocarps of *Armillaria* are harvested from tree trunks called "false pepper trees", in Oran (western Algeria).
The identification of the mushroom is carried out by classical, morphological and microscopic methods.
The crude extract is obteined from the dried sporophores by Soxhlet. This extract is tested *in vitro*, in Petri dishes, by the disc diffusion method, on the growth of 4 pathogenic microbial strains : *Escherichia coli* ATCC 8739 (Gram negative bacteria), *Staphylococcus aureus* subsp. *aureus* ATCC 6538 (Gram positive bacteria), *Candida albicans* ATCC 10231 (yeast), *Aspergillus niger* ATCC 16404 (filamentous fungi).
Results: In the results, the morphological description of the *Armillaria* showed that the basidiocarps has a smooth cap of 3 to 15 cm in diameter, convex at first but becoming flattened with age. The margins of the cap are often arched at maturity and the surface is sticky when wet. The gills are white at first, sometimes becoming pinkish-yellow or discoloured with age, broad and fairly distant, attached to the stipe at right angles or are slightly decurrent. The stipe is of variable length, up to about 20 cm long and 3.5 cm in diameter.
The results of antimicrobial activity showed a remarkable inhibitory effect of the armillary extract (max. 3cm inhibitions zones) on the growth of the different pathogenic microbial strains tested.
Conclusion: In conclusion, the mushrooms of the genus *Armillaria* are therefore interesting candidates for obtaining antimicrobial molecules of therapeutic interest.
Keywords: *Armillaria*, mushroom, extract, Soxhlet, basidiocarpss, antimicrobial activity.

BMP29: Essential Oil, Hydrosol Extract profile andsome phytochemicals of Algerian *Rhuspentaphylla* Desf. Evaluation of its Antioxidant Activity

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Background and aim: *Rhuspentaphylla*Desf. (Anacardiaceae) widely grown in Algeria, is an underutilized plant and remains poorly studied. The aim of this work is to investigate for the first time, the chemical composition of essential oil and hydrosol extract. Besides total phenolic (TPC), flavonoid (TFC) and condensed tannins contents (CTC) of fruits, leaves and roots, the fatty acid composition was assessed. The radical scavenging activity of *R. pentaphylla*.extracts was then evaluated.
Methods: Essential oil and hydrosol extract were isolated respectively by hydrodistillation and liquid-liquid-extraction from the aerial parts of *R. pentaphylla*. The antioxidant activity was evaluated using 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical and ferric reducing antioxidant power (FRAP). Chemical compositions of various extracts were investigated using Gas Chromatography (GC) and GC-Mass Spectrometry (GC-MS)
Results: The results of GC and GC-MS analysis revealed 83 components in essential oil and 75 components in hydrosol extract representing respectively 96.9% and 92.5% of the total extract composition. The main constituents were hexadecanoic acid (31.5%) followed by spathulenol (14.9%) in the oil while the higher compound present in the hydrosol extract was spathulenol (14.2%). Otherwise, 13 and 18 fatty acids were identified in roots and fruits respectively. The highest levels of TPC and CTC were found in roots extracts while the highest quantity of TFC has been recorded in the leaves extract. Besides, hydrosol extract was able to scavenge DPPH and FRAP free radicals more efficiently than essential oil.
Conclusion: *R. pentaphylla*. contains bioactive substances that could be used as a new promising source of antioxidant compounds in different applications.
Keywords: essential oil, hydrosol, fatty acid, phenolic compounds, antioxidant activity, *Rhuspentaphylla.*

BMP30: Antioxidant and Antimicrobial Activities of Leaves Extracts of *Urtica dioica* L. from Algeria: Application in Apple Fruits

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Background and aim: Increasing interest in replacing synthetic food antioxidants by natural antioxidants has intensified the demand for plant materials for the identification of new antioxidants and also antimicrobials molecules. The aim of this work is to study the chemical composition of essential oil and determination of total phenolic content of different leaves extracts of *Urtica dioica* collected in the province of Tizi-ouzou, North of Algeria. Also, their antioxidant and antimicrobial activities.
Methods: The total content of phenolic compounds was determined by Folin-Ciocalteau reagent. The antioxidant activity was evaluated using free radical DPPH reduction method. DPPH. The essential oil was analyzed using GC-MS technique. The study of the antibacterial activity was carried out by the method of agar against bacterial and fungal strains.
Results: Major compounds found in essential oil of nettle were Carvacrol (27.42 %) and Naphtalene (6.86 %) as principal chemical components. Essential oil revealed an antibacterial activity with respect to the bacterial and fungal strains tested. The total content of phenolic compounds was 8.44, 1.41, and 1.02 mg GAE/g of dry weight of the aqueous extract, ethyl acetate and ethanol extracts respectively. All the studied extracts showed a good antioxidant activity than ascorbic acid, used as standard. The values of antioxidant activity determined by DPPH method increased in the following order: ethyl acetate, aqueous and ethanol extracts. It was revealed that *U. dioica* extracts, exhibited the antimicrobial activity at least one of the tested microbial strains. The essential oil was tested in apple fruits; experimentally inoculated with *Botrytis cinerea* at a concentration of 10^7 spores/ml. Results demonstrated that *U. dioica* essential oil exerted an antifungal effect against pathogen tested. This effect was evident from 24 hours of incubation; showing significant differences with untreated samples.
Conclusion: The data suggest a possibility that essential oils could be used as natural preservatives for improving life storage of apple fruits.
Keywords: Essential oil, Polyphenols, Antioxidant activity, Antimicrobial activity, Apple fruits.

BMP31: Optimization, purification and antioxidant activity of B-phycoerythrin from *Porphyridium marinum*

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Background and aim: Chemical molecules namely *tert*-butylhydroquinone (TBHQ), butylated hydroxyanisole (BHA), propyl gallate (PG) or butylated hydroxytoluene (BHT) are widely used in food and cosmetic industries. However, the use of synthetic antioxidant molecules is questioned because ofpotential toxicological risks and carcinogenesis. This why and in view of the desire of consumers to return to the use of natural products, the search for natural sources of antioxidants has attracted the interest of large specialized laboratories. In this context, the aim of the present study is to optimize the production of B-phycoerythrin (B- PE) from the red microalga *Porphyridium marinum* in order to purify it and study its antioxidant activities.
Methods: The optimization ofthe production of B-PE was carried out by adapted experience plans, Plackett-Burman and Box-Behnken design. The protein B-PE was then purified through an anion exchange column DEAE-Cellulose. Finally the antioxidant activities of purified B-PE was evaluated by various *in vitro* antioxidant assays such as DPPH scavenging activity, ferrous iron chelating capacity, reducing power and β-carotene bleaching test
Results: NaNO3 and K2HPO4 concentrations, luminosity, and metal solution were chosen, according to the Plackett-Burman design, as the most influent factors on the B-PE production by *P. marinum*. The optimization of these factors according to the Box-Behnken plan gave a maximum of B-PE production equal to 40 mg/g Dry Weight under the following conditions: NaNO3 = 3.4 g/L; K2HPO4 = 0 g/L; light intensity = 70 µmol photons/m^2/s and metal solution = 1.5 mL/L. The purified pigment showed a promising antioxidant capacity displaying a good DPPH radical scavenging activity (IC50 = 9.81 ± 0.64 µg/mL) and βcarotene bleaching inhibition (IC50 = 11.94 ± 0.13 µg/mL). It also showed an interesting capacity to chelate Fe^{3+} (IC50 = 13.91 ± 0.21 µg/mL) and a good reducing power (IC50 = 125 ± 0.53 µg/mL).
Conclusion: The present report, for the first time, describes antioxidant potential of optimized and purified B-PE from *P.marinum* that could be potentially used as a source of natural bioactive protein for cosmetic and pharmaceutical applications.
Keywords: B-phycoerythrin, *P. marinum*, Optimization, Antioxidant activities.

BMP32: Toxic effects of Nickel Chloride on Haematological Parameters and Enzymatic Activities in Male Albino Rats can be reversed with Fig (Ficus Carica.L) Extract Treatment

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Background and aim: Nickel is reported as a heavy metal that induces blood disorders and immunological effects because is a non-essential element for human body, it is one of the common environmental contaminant; Fig (Ficus Carica) is an important crop that can be consumed fresh or dried, the present study pertains to the protective and anti oxidative effect of Fig (FC) (Ficus carica.L) aqueous extract against NiCl2 induced toxicity in adult male albino rats
Methods: 18 male *albino* rats were intoxicated with 1% Nicl2/IP. After one month of the experiment, 6 rats among those given NiCl2 (**Gr_Ni1**) were sacrificed, Then, the rest of the Rats whose were given NiCl2 (12 rats) was divided into two groups: the first group, 6 rats received daily distilled water **(Gr_Ni2)**, the second group, 6 rats (**Ni+FC**) treated with 35% of FC aqueous extract by force-feeding. Four weeks later, the rats were killed, the Blood was collected, and plasma was prepared by low-speed centrifugation for the determination of the Haematological, Biochemical, and some oxidative stress Parameters.
Results: The intraperitoneal administration of 1% of NiCl2 during one month induced a significant increase *($P < 0.05$)* in the red blood cells count (RBC), hematocrit (Ht), and haemoglobin (Hb) levels, which are indication of anemia, and significant decrease in white blood cells count (WBC). In addition, an increase in plasma aspartate aminotransferase (AST) and alanine aminotransferase (ALT) levels was observed. The exposure to the NiCl2 also favored oxidative stress as shown in the significant increase *($P < 0.05$)* in lipid peroxidation (MDA-rate) and the decrease in the activity of the catalase (CAT) in the intoxicated groups **(Gr_Ni1).**
The orally administration of the Fig Fruits Aqueous extract enabled us to observe a significant decrease *($P < 0.05$)* in haematological parameters: decrease in plasma (AST, ALT), a Thiobarbituric acid reactive substances (TBARS) level (protect from lipid peroxidation) and markedly ameliorate a catalase activity in **(Ni+FC)** compared to **(Gr_Ni2).**
Conclusion: The present study was designed to ascertain the benefit effect of Ficus Carica Fruit aqueous Extract that prevented adverse effect of tested heavy metal (Nickel) on haematological and enzymatic activities.
Keywords: Ficus Caria, Nickel Chloride, oxidative stress, hematology, blood parameters.

BMP33: Aqueous extract of "*Rosmarinus officinalis*"and cerebral dysfonctions in rat after aluminium exposure

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Background and aim: *Rosmarinus officinalis* (RO) is a plant known for these Mediterranean culinary virtues and has been widely used in traditional medicines, mainly due to the presence of several phenolic compounds. The Aim of this study is designed to investigate the effect of the aqueous extract of Rosmary on deleterious effects caused by the toxicity of aluminum (AlCl3) in the rat brain**.**
Methods: Thirty-two wistar male rats weighing 60 ± 10 g were divided into four groups: Controls (receives the water); Intoxicated groupe (receives the (AlCl3) by an intraperitoneal injection of 60mg / kg / one times a week; Treated groupe (receives the aqueous extract of Rosmary orally by gavage of 1 50mg / kg / ml / Day, and the Intoxicated Groupe Treated (receive [60mg / kg / S (AlCl3) + 150mg / kg / ml / Day (RO)], during 90 days.
After 90days of experimentation, the rats are subjected to behavioral tests (Elvated plus maze and forced swimming test) for asses the ability of plant antioxidant molecular to restore a physiological homoeostasis. After that animals were sacrificed and the brains were immediately removed, stored at −80°C.
Later the brain were taken and minced into small pieces then homogenized with ten volumes of phosphate buffer (0.1 mol/L, pH 7.4), and the homogenates at 4 °C to yield the supernatant which was later used for the estimation of antioxidants parameters [superoxydedismutase (SOD), catalase (CAT), Thiobarbituricacidre active substances (TBARS) and acetylcholinesterase activity(AChE).
Results: The chronic exposure to (AlCl3) causes significantly modified stereotypic and cognitive behavior with marked depression and anxiety in rats; a significant inhibition of acetylcholinesterase activity in brain, accompanied by disruptions in the activity of antioxidant enzymes (SOD, CAT) and lipid peroxidation with an increase in the TBARS rate. However, the administration of the aqueous extract of Rosemary modifies the cognitive behavior (significant decrease in depression and anxiety) and enzymes activity, as well as a decrease in lipid peroxidation.
Conclusion: This study suggeste that '*Rosmarinus officinalis*' extract could possibly restore and protect the neurological function capacities and improvement of defenses against radical aggression caused by the Aluminum.
Keywords: *Rosmarinus officinalis*, Aluminium, brain.

BMP34: Analgesic potential of a sulphated polysaccharide isolated from a brown seaweed, *sargassum vulgare*

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Background and aim: Brown seaweeds are largely used in many domains such as pharmaceutical, cosmetic and food industries. In fact they are known to have many bioactive substances. Fucoïdan is a sulphated polysaccharide extracted from brown seaweed which exhibit many pharmacological activities. The aim of this study was to evaluate the analgesic activity of a fucoïdan isolated from a brown seaweed, *sargassum vulgare*, harvested from the coastal region of Bizerte.
Methods: Adult Swiss mice were used for this study. Acetic acid writhing test was used to test peripheral analgesic effect. Acetylsalicylate of lysine (ASL) was used as a reference drug . The number of abdominal constrictions episodes was recorded during 30 min. The antinociceptive activity (reduction in the number of writhes) was expressed as percentage of pain inhibition in treated mice. The central analgesic activity was evaluated using hot plate test. Tramadol was used as a reference drug. The latency time was barely recorded before challenge and at various time points after injection of saline or the tested compounds (30, 60, 90, and 120 min). The elongation in the response latencies is considered as an antinociception parameter.
Results: The pharmacological evaluation of the isolated fucoïdan showed a significant peripheral analgesic activity at the dose of 100 mg/kg with 63.6% of writhing inhibition. Moreover, at this same dose, fucoïdan showed an interesting central analgesic activity with a significant increase in the hot plate reaction time. The maximum of action was noted 60 minutes after injection.
Conclusion: The results of this study are promising. However complementary studies are required to understand the mechanisms of action and to screen others bioactivities.
Keywords: fucoïdan, sargassum vulgare, analgesic activity.

BMP35: Soil Bacteria Isolated from Tunisian Arid Areas Show Promising Antimicrobial Activities

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Background and aim: Antibiotic resistance is a major threat to public health, and the situation is aggravated by the shrinking of the antibiotic development pipeline. To fill the latter up again, novel natural products from microorganisms, the major source for novel chemical anti-infective scaffolds exhibiting new modes of action, have to be identified. The aim of this work was to identify soil bacteria isolated from unexplored arid sampling sites of Southern Tunisia, selected for their antibacterial activity, and to identify the bioactive compounds of one promising isolate.
Methods: The soil bacteria were isolated by standard serial dilution plate technique. Molecular identification of the isolates of interest was done by sequencing the 16S rRNA gene. MEGA 7.0 software was used to build the phylogenetic tree of these isolates. Antibacterial activity of identified isolates was tested against Gram positive and Gram negative bacteria. Two bioactive compounds were identified using LC-MS/MS and active NMR, respectively.
Results: Selected bioactive bacteria produce natural products effective against both Gram-positive and Gram-negative bacteria. The most of isolates belongs to the Firmicutes phylum (genus *Bacillus*); one isolate belongs to the Actinobacteria phylum (genus *Brevibacterium*) and one to the Proteobacteria phylum (genus *Pseudomonas*). The isolate *Bacillus* sp. M21, exhibiting high antibacterial activity, especially against clinical isolates of Gram-negative bacteria, was deeply investigated to identify the bioactive compound. Using LC-MS and NMR techniques the 1-acetyl-β-carboline was isolated. Furthermore, using LC-MS/MS and molecular networking analysis, isomers of bacillomycin, fengycin, and surfactin group were identified.
Conclusion: The strain collection of rhizospheric bacteria from Tunisian arid areas represents an important source to discover new antibiotically active compounds. The strain collection is dominated by the Gram-positive genus Bacillus, which seems to play an important role in shaping the microbiome by the production of bioactive compounds.
Keywords: Soil bacteria, Antibacterial activity, natural compounds, *Bacillus.*

BMP36: Valorization of hydromethanolic and methanolic extracts from leaves of *Ammi visnaga* and their antibacterial activity

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Background and aim: Interest to spontaneous plants is increasing, because it was recognised that these plants contain bioactive compounds and then, have medicinal virtues and a great importance to the health. The present work is aimed to study the antibacterial activity for the hydromethanolic (methanol-water 7/3) and the methanolic extrats from leaves of *Ammi visnaga*, a spontaneous plant growing in north of Algeria and very known with it's high antioxidant capacity [1].
Methods: The antibacterial capacity was evaluated using the disc diffusion method against 6 kinds of bacteria: *Escherichia coli* (ATCC 25922), *Klebsiella pneumoniae* (KPC^+), *Klebsiella pneumonia* (KPC^-), *Pseudomonas aeruginosa* (ATCC 27853), *Staphylococcus aureus* (ATCC 25923), and *Staphylococcus aureus* (MRSA) [2, 3].
Results: Among the extracts tested, the methanolic extract of the leaves show more antibacterial activity than the hydromethanolic extract, like a very good activity against KPC+ with 16 mm diameter of inhibition zone and a good activity against *P. aeruginosa* and *Staphylococcus aureus* with 14.8 mm and 14.1 mm. Also, the results indicate a medium activity against KPC- , *E. coli* and MRSA with 13.1 mm, 10.6 mm and 8.6 mm diameter of inhibition zone, respectively.
Conclusion: According to the results, we observe that the leaves of *Ammi visnaga* have a considerable antibacterial capacity due to their richness in bioactive compounds, which is confirmed by a previous study [1]; therefore these antibacterial proprieties might increase the therapeutic value of this spontaneous plant.
Keywords: *Ammi visnaga*, leaves, antibacterial activity.
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BMP37: Enhancement of insecticidal toxicity, structural study and molecular docking of *B. thuringiensis* Vip3Aa16-Cry1Ac chimera toxin

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Background and aim: *Bacillus thuringiensis* Vip3Aa16 protein is a promising insecticidal protein for crop protection and for delay resistance to existing insecticidal Cry toxins. Its sequence properties and its fusion with Cry1Ac activated protein were examined in order to enhance its insecticidal activity.
Methods: A fusion between *vip3Aa16* and the toxic core sequence of *cry1Ac* was constructed and then expressed by *B. thuringiensis*. The toxicity and the structure of chimera protein were investigated *in vivo* and *in silico*.
Results: Vip3Aa16-Cry1Ac chimera protein (150 kDa) was detected in the supernatant of *B. thuringiensis*. Bioassays tested against *Ephestia kuehniella* showed that the chimera toxin improved the toxicity to reach 90% ± 2 with an enhancement of 20% compared to the single Vip3Aa16 protein. So, the fusion protein could be a new approach to enhance Vip3Aa16 toxicity against lepidopteran species and could avoiding insect tolerance of *B. thuringiensis* delta-endotoxins. Additionally, we have predicted for the first time the whole 3D structure of a Vip3A toxin by computational study. Vip3Aa16 structure showed three domains like Cry toxins: an N-terminal domain containing hemolysin like fold as well as two others Carbohydrate Binding Module (CBM)-like domains. Molecular docking analysis of the chimera toxin and the single Vip3Aa16 protein against specific insect receptors revealed that residues of CBM like domains are clearly involved in the binding of the toxin to receptors.
Conclusion: The enhancement of insecticidal toxicity through application of chimera Vip3Aa16-Cry1Ac could be very useful in design of novel toxins to surmount the problem of *B. thuringiensis* tolerance or resistance to Cry toxins.
Keywords: *B. thuringiensis*, Vip3Aa16, Cry1Ac, Chimera toxin, Insecticide, Model prediction, Protein-protein docking.

BMP38: design of *Rosmarinus officinalis essential oil, Phoenix dactylifera phenolic extract* and acetic acid: Optimization of their antibacterial effect against Psychrophilic *Pseudomonas* strains

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Background and aim: Preservation at low temperatures is conscientiously applied. However, it promotes the development of psychrotrophic microorganisms, especially during the break of the cold chain. The purpose of this work is to accentuate the cold preservation with using natural agents.
Methods: Essential oil of *Rosmarinus officinalis L.* was obtained with hydrodistillation, while phenolic compounds from *Phoenix dactylifera L.* were extracted by Microwave assisted extraction. A simplex centroid mixture design was used for optimizing the antibacterial effect against psychrophilic *Pseudomonas* strains at mesophilic and low temperatures.
To maximize phenolic compounds recovery from stem of *Phoenix dactylifera*, Microwave assisted extraction was coupled with Response Surface Methodology. The optimum conditions extraction was 600W for power irradiation, time of 90s and pH=7.
This study showed that *Pseudomonas* sp. isolated from frozen fish and *Pseudomonas aeruginoasa* were resistant to phenolic extracts, while essential oil exhibited a considerable activity with a diameter of the zone inhibition of 23 ± 1.41mm and 26 ± 2.81mm against *Pseudomans* sp. and *Pseudomans aeruginosa*, respectively. Acetic acid gived a diameter of 30 ± 0 mm. The results show no significant difference between the effect of the agents at 37 °C and at 4°C. However, a synergistic effect has been recorded between the essential oil and acetic acid against *Pseudomonas aeruginosa* by using a simplex centroid mixture design.
This work thus highlighted the interest of combining essential oils and organic acids to preserve food. This will allow one side to accentuate their effect, and another reduces the undesirable effects of high concentration on the taste and texture of products.
Keywords: Psychrophilic strains, *Phoenix dactylifera L, Rosmarinus officinalis L,* acetic acid, synergy.

BMP39: Formulation and optimization by experimental design of dairy desert based on *Lupinus albus* and *Stevia rebaudiana* extracts

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Background and aim: Fresh milky desserts include a wide variety of products from milk based traditional and familial recipes. These preparations contain mainly milk, which is mixed to other ingredients such as sugar, coffee, chocolate, cream, flavors, and colors.
Lupin seeds contain large number of bioactive compounds that improve metabolism, when they are regularly consumed. Interestingly, ingestion of lupine isolates and/or lupine-containing foods has powerful benefits to prevent obesity, diabetes and cancer, hyperlipidemia and cardiovascular disease. In diabetic patients, lupine proved its hypoglycemic activity.
Lupinus has not received sufficient attention by the industrial sector. However, this situation may change; by extending studies about the creation of food mixture that contain mainly vegetable proteins. Thus, substitution or supplementation of cow's milk with lupine milk would improve the production of healthy cream dessert.
Methods: The aim of the present study is to evaluate physicochemical, microbiological and sensorial properties of cream dessert, made with mixtures of partly skimmed reconstituted milk and lupine milk at different concentrations. To reduce the amount of sugar, from 11% in the conventional protocol to 7.15%, 3.85% of *Stevia rebaudiana* extract was used.
Results: Compared to the reference product, eighteen elaborated trial protocols had high protein content ranging from 8.36 to 16.25 g / 100g. Furthermore, some mixtures had high amounts of fat, ash and calcium. Microbiological analysis revealed complete absence of pathogenic organism in the eighteen tests, which proved their safety for consumption. In this study, we developed a good mixture protocol to prepare cream dessert which is highly rich in protein (14,646 g / 100 g) and calcium (97,796 mg / 100 g), with an estimated acceptability of 3,424.
Conclusion: Modeling the responses according to different factors such as partly skimmed reconstituted milk, aqueous extract of *Lupinus albus*, and texture agents (starch and pectin), reviled the individual effect of each component on the overall properties of cream desert. The obtained formula gave products with good nutritional values, textural and taste properties.
Keywords: cream dessert, experimental design, *Lupinus albus*, pectin, *Stevia rebaudiana.*

BMP40: New compounds and antibacterial activity of *Haloxylon scoparium* Pomel from Algeria

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Background and aim: *Haloxylon scoparium* Pomel (Amaranthaceae) (Hs) is a spontaneous species, endemic to desert regions. It is traditionally used in Algeria in various diseases such as urogenital infections and in some cases of cancer. Some studies on the species, in Tunisia and Morocco, have revealed the presence of alkaloids and flavonoids. These studies remain insufficient for the Algerian context considering the important potential of the genus *Haloxylon*, and the phytochemical variability due to climatic and edaphic factors. The aim of the present study was to isolate new molecules from Algerian Hs and to evaluate the antibacterial activity of its enriched extracts.
Methods: Preparative chromatography technics as exclusion chromatography and medium pressure liquid chromatography (MPLC) on various supports were carried out on the ethanolic extract of the aerial parts to isolate pure products.
Spectroscopic methods: SM and 1D and 2D NMR were used for the structural determination. The microdilution technic was performed on enriched extracts to evaluate antibacterial activity on four reference strains and two pathological strains compared to gentamycin.
Results: Three new natural products were isolated, an alkaloid pyridinic derivative, a sequiterpenoid and an aliphatic acid derivative. Another lipid derivative witch was previously elucidated in the genus. The ethanolic extract and the alkaloid enriched fraction shown an antibacterial activity against all strains. The activity against *E.coli* and *E. enteritidis* were most important than gentamycin.
Conclusion: The phytochemical profile of Hs is complex with different classes of compounds, alkaloids, sesquiterpenoids and aliphatic derivatives, the chromatographic profile lead to research other new compounds. The antibacterial activity of the aerial parts attest the traditional use in urogenital affections.
Keywords: *Haloxylon scoparium* Pomel, Natural product, Phytochemistry, Antibacterial activity, Chromatography, Spectroscopy.

BMP41: Evaluation of the antioxidant and anti-inflammatory activity of the methanolic extract of *Origanum majorana L.*

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Background and aim: Marjoram (Origanum majorana L., Family: Lamiaceae) contains many bioactive compounds of interest including polyphenols. These organic molecules arouse the interest of scientists and industrialists because of their antioxidant properties. The aim of this work is to evaluate the antioxidant and anti-inflammatory activity of the methanolic extract of the leaves of marjoram.
Methods: The antioxidant activity of the methanolic extract of Origanum majorana L. was evaluated by the following tests: phosphomolybdate (CAT) test, iron reducing power, and DPPH antiradical activity. As well as the evaluation of the anti-inflammatory activity of this extract was carried out by the method of inhibition of the denaturation of bovine serum albumin.
Results: The antioxidant activity proved to be important in the methanolic extract (0.82 mg EAA / 100 g of the dry matter) for 2 ppm of the extract in comparison with that of the standard used (ascorbic acid). The leaf extract of Marjoram has a moderate antioxidant and antiradical potential compared to the standard antioxidant used (EC50 = 0.66 mg / ml, 0.269 mg / g DPPH the results of the tests of reducing power and DPPH respectively). The protein denaturation inhibition test of the leaf extract of Marjolaine and of sodium declofenac (anti-inflammatory reference), showed that the declofenac have the best thermal anti-denaturation efficacy of the BSA than the extract. But at a concentration of 0.125 mg / ml, the extract presents a strong activity than that of the déclofénac with respect to the thermal denaturation of the BSA.
Conclusion: The results of this work have allowed us to affirm that the antioxidant and antinflammatory activity of the raw extract of the marjoram leaves at different concentrations essentially returns to the phenolic compounds.
Keywords: *Origanum Majorana L.*, antioxidant activity, anti inflammatory, CAT, reducing power, DPPH, BSA.

BMP42: Identification of a novel heteromultimeric protease Prot CT2 for the hydrolysis of legume seed proteins

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Background and aim: Lately, microbial proteases represent the major group of industrial hydrolytic enzymes that find application in pharmaceutical industries and biotechnological research.The most important application of microbial proteases concerns their involvement in the synthesis of peptides, the production of food ingredient by the hydrolysis of food proteins or the improvement of the digestibility of proteins. Although legumes are rich in proteins, they have a low nutritional value due to reduced digestibility. Thus, the search for new proteases that can improve the digestibility of these proteins is quite interesting. The aim of this study is to identify a previously described high molecular weight alkaline protease (Prot CT2) produced by *Bacillus halotolerans* CT2strain and provides information on its potential use in legume proteins hydrolysis and digestibility.
Methods: We have firstly purified the Prot CT2 through ammonium sulfate precipitation, Sephacryl S-200 and SP-sepharose. Purified protease was analyzed by Tricine SDS-PAGE after denaturation; subunit bands were excised and subjected to in-gel trypsin digestion. The resulting peptides were analyzed by mass spectrometry performed with ESI-Q-TOF. Proteolytic activity was evaluated against modified and natural substrates. Hydrolysis of legume proteins by Prot CT2 was also performed and DH (%) was determined.
Results: Fractionation of concentrate prot CT2 extract by Sephacryl S-200 column yielded two peaks. The main proteolytic activity was eluted in peak 2 with 28.8 purification fold. Cation exchange chromatography step showed one major peak with 34.8% purification fold. Prot CT2 migrated as a single band at about 250 kDa. Tricine SDS-PAGE analysis under denaturating conditions detected two protein bands: S1 (30 kDa) and S2 (25 kDa).Analysis of the protein bands peptides by mass spectrometry performed with ESI-Q-TOF mentioned peptide sequences identified in subunit S1and S2 showing 100 % similarity with subtilisin AP01 and peptidase S8.Prot CT2 exhibited a potent catalytic activity towards natural and chemical protein aceous substrates and showed high hydrolytic activity against legume proteins.
Conclusion: ProtCT2 enzyme is heteromultimeric presenting high homology with the subtilisin-like serine proteases.It showed high catalytic efficiency and has a good ability to hydrolyze legume seed proteins to generate bioactive peptides.
Keywords: high molecular weight, heteromultimeric, legume proteins, hydrolysis.

BMP43: Chemical Composition and Antioxidant, Analgesic, and Anti-Inflammatory Effects of Methanolic Extract of *Euphorbia retusa* in Mice

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Background and aim: *Euphorbia retusa* of family Euphorbiaceae is an annual herb abundantly found throughout the Mediterranean area. It is used in traditional medicine as a cure for warts, trichiasis, and venomous bites. The aim of this study is to investigate the phytochemical constituents of the methanolic extract of *Euphorbia retusa* and to evaluate their antioxidant, anti inflammatory and analgesic activities.
Methods: Dry powder of *Euphorbia retusa* was extracted with methanol and analyzed by HPLC and by chemical assay reactions. The antioxidant activity in vitro was evaluated by the DPPH and the FRAP assays. The antinociceptive activity of methanolic extract of *Eupohrbia retusa* was assessed using acetic acid and hot-plate models of pain in mice whereas the antiinflammatory activity was evaluated by carrageenan-induced paw edema model.
Results: the phytochemical results have revealed the richness of the methanolic extract of *E. retusa* in active compounds, in particular polyphenols, flavonoids, and tannins. In vitro, the extract of *E. retusa* showed significant dose dependent antioxidant activity. In vivo, Oral pretreatment of mice with the methanolic extract of *E. retusa* (200 mg/kg) exhibited a significant inhibition of pain induced either by acetic acid or by the heating plate and in a manner comparable to the standard drug paracetamol. *E. retusa* significantly reduced paw edema starting from the 3rd hour after carrageenan administration by increasing the activity of antioxidant enzymes (SOD, CAT, and GPx) in liver and paw tissues and decreasing the levels of MDA.
Conclusions: Our results can confirm that this plant has an interesting therapeutic potential for oxidative stress, and disorders related to pain and inflammation.
Keywords: *Euphorbia retusa*; antioxidant; anti-inflammtory; analgesic.

BMP44: Optimization production of *ViciaFaba* protein hydrolysate using response surface methodology: Anti-biofilm activity evaluation

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Background and aim: *Candida albicans* is an opportunistic pathogenic commensal yeast for human microbiome and is the principal causative agents of candidiasis. This infection is frequently associated with biofilm formation. *C.albicans* biofilm represents one of the major virulence factors and contributes to high mortality rates even with treatment using available drugs which know several limits in terms of efficiency, toxicity, drug interaction and high cost. Consequently, the search of new antifungal agents from natural sources, especially peptides derived from the hydrolysis of dietary proteins, has become a public health priority. The aim of this work is to optimize the hydrolysis conditions of *Vicia Faba* by microbial protease, using response surface methodology (RSM) in order to obtain a high degree of hydrolysis and a good anti-biofilm activity.
Methods: The optimization of hydrolysis conditions of *Viciafaba* proteins was carried out by the response surface methodology using Box-Behnken design to evaluate the effect of three factors including hydrolysis time (h) (X_1), substrate concentration (w/v) (X_2) g/100 mL and substrate to enzyme ratio (w/w) (X_3) and their interactions on two responses variables (Y_{DH}: degree of hydrolysis and Y_{AA} : inhibition of *C. albicans* ATCC 10231biofilm formation). Experimental data were analyzed by STATISTICA (version 7.0) software.
Results: Based on RSM analysis, the optimum conditions for the independent variables were obtained as follows: substrate concentration of 0.5 % (g/100 mL), a ratio for enzyme to substrate of 2 % (g/100g) and hydrolysis time of 2 h. Under these optimum conditions, the obtained hydrolysate displayed a high degree of hydrolysis (93.74 %) with potent biofilm inhibition of *C. albicans* (84.38 %).
Conclusion: These promising results showed that the protein hydrolysate of *Viciafaba* could be an attractive agent to prevent pathologies related to the formation of *C. albicans* ATCC 10231 biofilm.
Keywords: protein hydrolysate, RSM, Box-Behenken, anti-biofilm activity.

BMP45: Optimization of total flavonoid hydro-methanolic extraction from Bee Pollen and their antioxidant activity

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Background and aim: The present study consists of establishing the optimal conditions to have the best yield of flavonoid pollen grains and their antioxidant power activity.
Methods: The material used is the pollen grains of region of Sidi bel abbes in west of Algeria harvested during the months April and May 2015.
The extraction parameters tested and their variations are: Type of organic solvent (methanol), concentration of the organic solvent (50%, 60%, 70%, 80% and 90%), temperature of extraction (30, 40, 50 and 60°C) and extraction time (2h, 4h, 6h and 8 hours). The flavonoid assay was done by the aluminium trichloride and sodium hydroxide method.
Flavonoids are determinate by Ultraviolet and Visible Molecular Absorption Spectrometry.
The antioxidant activity was tested by the method of free radical scavenging with DPPH.
Results: The results show a low concentration of flavonoids (1,539 mg EC/g) at (30°C, 2h, 50%). While a maximum of (5,939 mg EC/g) at (60°C, 8h, 90%).
The antioxidant activity test revealed that the flavonoid optimum has a high antiradical potency IC50 (4.830 mg/ml) compared with the standard extract of a maceration in methanol which gives an IC50 of (10.088 mg/ml).
Conclusion: The optimal extract of flavonoids revealed a very important antioxidant activity in the optimal conditions found according to our experimentation and it would be very interesting to valorize these extracts by tests of various biological activities and study the possibility of exploitation in industrial scale.
Keywords: bee pollen, hydro-methanolic extraction, flavonoids, antioxidant activity.

BMP46: Sublethal effects of *Bacillus thuringiensis* (Vectobac G), on the survival rate, longevity and fecundity of mosquito specie

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Background and aim : Mosquitoes are vectors of several diseases like malaria, filariasis, dengue fever, yellowfever, etc., causing serious health problems to human beings. These insects are generally controlled by conventional pesticides. Further, the indiscriminate use of neurotoxic insecticides caused various environmental aspects toxic problems to non target organisms and insecticide resistance. An alternative approach for mosquito control is the use of natural products such as plant and microorganisms. In the present study bioassay was conducted to test the larval toxicity and longevity of *Bacillus thuringiensis* vectobac G on *Culex pipiens* and *Culesita longiareolata*.
Methods: Toxin preparation used in this study was lyophilized powder of spore and crystal mixture of lysed cultures of *Bacillus thuringiensis* variety *israelensis*. Laboratory bioassays were conducted on the efficacy of a granule (G) formulation of *Bacillus thuringiensis* variety *israelensis* (Vectobac G; active ingredient: 200 *Bt i*international toxic units [ITU]/mg) at different concentrations, against newly ecdysed larvae for all different larval stages (L1 to L4) of *Culex pipiens* and *Culesita longiareolata*.
Results: Bioassays showed that *Bacillus thuringiensis* (Vectobac G) have a high toxicity against the two species, *Culex pipiens* and *Culiseta longiareolata*. The mortality varied with concentration for the different larval stages and the both mosquito species. However the adult longevity of both mosquito species was considerably reduced by the treatment of *Bt.* Vectobac G. The Effect of *Bt.* Vectobac G on reproduction was evaluated on different parameters, of the females emerged from the treated fourth instar larvae of *Cx. pipiens* and *Cs. longiareolata*. The fecundity for both species was also affected and a significant decrease was obtained for the two lethal concentrations.
Conclusion: From the present study we conclude that *Bt.* Vectobac G proved good larvicidal agent against *Cx pipiens and Cs. longiareolata* larvae in laboratory and also reduced the longevity of different developmental stages, egg productions and fecundity**.**
Keywords: Mosquito, *Culex pipiens*, *Culiseta longiareolata*, toxicity, *Bacillus thuringiensis*.

BMP47: Lactic Acid Bacteria Protects Synergistic effects of Aflatoxin B_1 and Fumonisin B_1in Mice

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Background and aim: Aflatoxins are important food-borne mycotoxins implicated in human health and have immunocytotoxic effects. The aims of this study were to evaluate a new aflatoxin B_1 (AFB_1) and fumonisin B_1 (FB_1)-binding/degrading microorganism for biological detoxification, examine its ability to degrade AFB_1 and FB_1 in liquid medium, and evaluate its potential *in vivo* protective role against any synergistic effect of AFB_1 and FB_1 on DNA and gene expression in splenocytes, caspase-3 activity and mRNA levels of inflammation-regulating cytokines.
Methods: Female Balb/c mice were divided into eight groups (10/group) and treated daily for 2 wk by oral gavage with AFB_1 (80 µg/kg BW) only, FB_1 (100 µg/kg) only, AFB_1 + FB_1, or a lactic acid bacteria (*Lactobacillus paracasei* BEJ01, 2 × 10^9 CFU/L, ~ 2 mg/kg) alone or in combination with the AFB_1 and/or FB_1. At the end of experiment, the spleen was collected for the determination of caspase-3 activity, lipid peroxidation (LP), and glutathione (GSH) content, as well as for any alterations in expression of anti-oxidation protective enzymes (GPx and SOD) and mRNA levels of inflammation-regulating cytokines (e.g., *IL-10*, *IL-4*, *IFNγ*, *TNFα*). The thymus of each mouse was also recovered for analysis of apoptosis among thymocytes.
Results: The results indicated that, in the spleen, exposure to the mycotoxins led to increased caspase-3 activity, LP, and *IL-10* and *IL-4* mRNA levels, but decreased GSH content and down-regulated expression of GPx and SOD and *IFNγ* and *TNFα* mRNA. Co-treatment with AFB_1 or FB_1 plus LAB suppressed the levels of DNA fragmentation, normalized splenic LP and increased GSH levels, up-regulated expression of GPx and SOD, and normalized mRNA levels of the analyzed cytokines.
Conclusion: It is concluded that AFB_1 and FB_1 might have combinational (synergistic moreso than additive) toxic effects *in situ*. Further, it can be seen that use of LAB induced protective effects against the oxidative stress and (immuno) toxicity of these agents in part through adhesion (and so likely diminished bio-availability).
Keywords: Aflatoxin, Fumonisin, Lactic acid bacteria, Immunotoxicity, antioxidant; prevention.

BMP48: Chemical characterization of bioactive components of Rosa canina extract and study of its protective effect against DSS-induced IBD model in mice

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Background and aim: Rosa canina is a well known medicinal plant that exhibits various beneficial health effects.
Methods: In the present work, the phenolic components of Rosa canina flowers methanolic extract (RCME) were identified and quantified and it protective effect on dextran sulfate sodium (DSS)-induced colitis in mice was then investigated. Particularly, chemical characterization of bioactive components of Rosa canina extract and study of its protective effect against DSS-induced IBD model in mice was also investigates.
Results: The chemical analysis of RCME was done by LC-HRESIMS and experimental animals received RCME (100, 200 and 400 mg kg-1, p.o. P.C.) one week before induction of colitis by oral DSS administration during seven days. 15 phenolic compounds have been identified by LC-HRESIMS whose phenolic acids and flavonoids were the major compounds. In addition, we showed that DSS intoxication altered colonic epithelium by inducing severe histological changes associated with an oxidative stress state as assessed by increased levels of lipoperoxidation and depletion of antioxidant enzyme activities and non-enzymatic antioxidants, as well as a colon overload of reactive oxygen species (ROS) such as OH• and H2O2. We also found that DSS administration provoked the disruption of some intracellular mediator's levels such as free iron and ionizable calcium. The overall results protective effect the RCME treatment effectively against all histological and biochemical alterations induced by DSS intoxication.
Conclusion: The RCME treatment effectively protected against all histological and biochemical alterations induced by DSS intoxication.
Keywords: Rosa canina, phenolic compounds, DSS intoxication, DSS-induced IBD model in mice.

PB Session
Plant Biotechnology

ORAL COMMUNICATIONS
(PBO)

TC1-PB/PBO1: How do the most prevailing abiotic stresses affect legume-rhizobia symbiosis and its contribution to agrosystem sustainability?

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Background and aim: Legume crops are able to establish a nitrogen fixing symbiosis with soil rhizobia that allows them, when it operates normally, to ensure their necessary nitrogen nutrition. Thanks to this unique biological process, these species improve the soil nutrient balance and are cropped in rotation or association with cereals to make these latters benefit from the advantages of the formers. Although, in the Mediterranean agrosystems this symbiosis is generally submitted to the most prevailing abiotic constraints that limit its performance. Thus, the selection of most performing legume-rhizobia symbiosis could improve the agroecological services of legume crops. The aim ofthis work is to select more performing legume-rhizobia symbiosis under abiotic stresses and to assess their benefit for the associated crops.
Methods: The responses of legume-rhizobia symbiosis to the most prevailing osmotic and phosphorus deficiency stresses, separately or combined were studied in faba bean, common bean and Alfalfa under greenhouse and field conditions. The effects of the soil nutrients balance and the promotion of associated cereal crops were analyzed.
Results: The results showed that P deficiency affected negatively the performances of legumerhizobia symbiosis with significant variation between the combinations. This constraint increased acid phosphatase (APase) and phytase activities under controlled and field conditions. The nodulated legume roots increased the available P content in the rhizospheric soil compared the bulk. Legume crop improved nutrition and growth of associated cereal crops in the field. Also, osmotic stress affected negatively growth and yield of stressed legume rhizobia symbiosis with variations between combinations. This constraint affected also nitrogen fixing performance and nodule membranes stability. Under field conditions, the legume yield was negatively affected under osmotic stress. When this osmotic stress was combined with P deficiency, the reduction of symbiosis performance was enhanced and the variations between the assessed symbiotic combinations were evident.
Conclusion: The variation between legume-rhizobia combinations for there responses to osmotic and P deficit stresses allowed identification of some performing symbiosis. The benefit of this symbiosis for associated crops involved improvement of plant nutrition.
Key words: Legume, phosphorus, osmotic stress, intercropping, Agrophysiology.

PBO2: PGPR effect of microbial phytases isolated from the bean rhizosphere

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Background and aim: Phosphorus (P) is often one of the most limiting elements for plant growth because it is not very mobile and the most inaccessible in soils. This P deficiency affects particularly leguminous plants because of the symbiotic fixation of nitrogen that consumes more energy than mineral assimilation. In this context, our objectives have been to isolate and characterize the functional bacterial community of the soil that allows the mineralization of phytate (organic phosphorus), and to study the ability of these bacteria to produce phytohormones in the rhizosphere ofthe bean.
Methods: Characterization of the bacterial strains was done on a generalist medium and on a specific phytate mineralization medium and different assays and tests allowed to test the PGPR functions.
Results: The evaluation of the density of bacterial community able to mineralizing phytate by the secretion of phytase in the rhizosphere of *Vicia faba L* has shown the high ability of certain isolated strains to solubilize P. The characterization of total bacterial and phytate mineralizing strains showed that most isolates are Gram-, catalase + and oxidase-, round in shape and in different colors. The study of the Plant Growth Promoting Rhizobacteria (PGPR) effect of rhizobacteria as phytostimulation and biofertilization agents showed that 100% of the assays tested are able of producing auxin at 40.62 µg / ml concentration (Strain 1P3b4), while 90% of isolates were able to grow on a medium containing only phytate as the sole source of phosphorus so they are able to degrade phytate.
Conclusion: The use of these bacteria as an inoculant appears to be a promising solution to improve bean production in low input agriculture.
Keywords: Rhizobacteria, bean, PGPR effect, phosphate nutrition.

PBO3: Nodulation and mycorrhization of red pea (*Lathyrus cicera*) in Tunisia

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Background and aim: Red pea *(Lathyrus cicera*) has been used essentially as green manure, animal feed and for its high level of biological active compounds. However, the study of the diversity of associated microorganisms has never been realized. Thus, the aim of this work is to study the diversity of rhizobia and mycorrhiza in association with *L. cicera* grown in 24 different localities in Tunisia.
Methods: The nodulation, the frequency and intensity of mycorrhization were examined. Isolated bacteria were identified using PCR-RFLP of the 16S rRNA, *nod*C and *nif*D-K IGS. The *nod*A, *rrs*, *rec*A, *thr*C and *dna*K genes were sequenced for six representative strains. The associated arbuscular mycorrhiza fungi (AMF) were determined morphologically.
Results: *L. cicera* was nodulated by three different species. The most abundant ribotype was assigned to *R. leguminosarum*. The two other species were assigned to *R. laguerreae* and *E. meliloti*. Symbiotic diversity was assessed by PCR-RFLP of *nod*C gene and *nif*D-K IGS. Five symbiotic genotypes were detected. The *nod*A phylogeny showed that strains belonging to *R. laguerreae* and *R. leguminosarum* harbored the symbiovar viciae. These strains induced effective nodules. However, *E. meliloti* strains were associated to the symbiovar meliloti and induced ineffective nodules. A wide fluctuation in the intensity and the frequency of mycorrhization was noted. According to the International Culture Collection of Arbuscular Mycorrhizal Fungi (INVAM), three different genera were identified: *Glomus*, *Gigaspora* and *Septoglomus*.
Conclusion: Red pea is nodulated by three different species of rhizobia with the dominance of *R. leguminosarum.* Except for *E. meliloti*, all isolates tested fixed nitrogen. A large fluctuation in frequency and intensity of AMF colonization was detected. A high diversity of the mycorrhiza spores morphology was noted and three genera were identified.
Keywords: Red pea, Rhizobia, Nodulation, Mycorrhization, Diversity.

PBO4: Biodiversity within *Medicago truncatula* genotypes toward response to iron deficiency: Investigation of main tolerance mechanisms and effect of efficient *Sinorhizobium* strains' inoculation

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Background and aim: The aim of this work was to investigate the biodiversity within *Medicago truncatula* to identify tolerant genotypes in order to assess the main tolerance mechanisms and the effect of the rhizobium inoculation into the response to this constraint.
Methods: A screening test was performed on 20 *Medicago truncatula* genotypes to identify contrasting genotypes. The second part of the work was conducted in hydroponic system and three treatments were used: control (C), direct deficiency (DD) and induced deficiency by bicarbonate (ID). To study the implication of rhizobia inoculation on *Medicago truncatula* tolerance to iron deficiency, plants were inoculated with two *Sinorhizobium* sepcies: (*S. meliloti*, TII7) and (*S. medicae*, SII4). Biometric, physiological and biochemical parameters were analyzed as well as the differential expression of some genes families potentially involved in the stress response.
Results: The screening results suggested an biodiversity within stress response and the presence of contrasting tolerant sensitive behaviors. Some contrasting genotypes were identified to investigate the main mechanisms of Fe deficiency tolerance. The better tolerance behavior was correlated with a best capacity of these genotypes to maintain higher Fe acquisition efficiency by roots via rhizosphere acidification, greater stimulation of Fe (III)- chelate-reductase activity, the stimulation of catalase (CAT) and peroxidase (POX) activities and related genes 'expression, as well as the accumulation of polyphenols. TN1.1 1 showed the highest lipid peroxidation and leakage of electrolyte under iron deficiency conditions, which suggest that TN1.1 1 was more affected than A17 and TN8.20 by Fe starvation. Notably, the inoculation of Fe-deficient plants with TII7 and SII4 mitigate the negative effect of Fe deficiency on plant growth and oxidative stress compared to nitrogen-fertilized plants.
Conclusion: The identified contrasting genotypes toward iron deficiency response represent an original material for more deep investigation of Fe tolerance mechanisms. Our study contributes that plant inoculation with bacterial strains manifesting high PGPR performances increase plant growth and vigor under Fe deficiency conditions.
Keywords: Antioxidant capacity, Biodiversity, iron deficiency, *Medicago truncatula*, rhizobia, Tolerance.

PBO5: Biological control of phytopathogenic *Fusarium* spp. fungi by *Bacillus halotolerans* bacteria

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Background and aim: Phytopathogenic fungi *Fusarium* spp. are known for their broad host range, and by their adaptability to infect several plant species sharing the same environment. That's why, the main objective of this work was to apply the *in vitro* and *in vivo* biological control method against 15 *Fusarium* spp. through 3 antagonistic *Bacillus halotolerans* bacteria.
Methods: *In vitro* (direct confrontation method) and *in vivo* (biological tests on vegetable crops) tests, microscopic observation.
Results: Firstly, a pathogencity test was carried out on olive branches (*Olea europaea* L. cv. 'Chemlali'), and on potato tubers (*Solanum tuberosum* L. cv. Spunta); this test led to conclude that the most aggressive olive fungi are *Fusarium solani* (FZ and FK2). *Fusarium solani* (FZ) was also the most pathogenic fungi infecting potato tubers.
Then, *in vitro* biological control tests were carried out by direct confrontation method using 3 strains of *Bacillus halotolerans* against the whole collection of *Fusarium* spp. Indeed, a significant antifungal activity resulting in an inhibition of mycelial growth of 52% compared to the control, has been demonstrated by the bacteria LMRE36.
Finally, preventive, curative and concomitant treatments were carried out *in vivo* on olive twigs, potato tubers and tomato fruits (*Solanum lycopersicum* L. cv. Rio grande), using the bacteria LMRE36. The results obtained prove the capacity of the rot severity reduction which reaches up to 75%.
Conclusion: Vegetable crops intercropped with olive trees must be responsable of dieback diseases on olive trees in open fields. The use of antagonistic *Bacillus halotolerans* bacteria may be effective against *Fusarium* spp. phytopathogenic fungi.
Keywords: Biological control, *in vitro*, *in vivo*, confrontation, phytopathogen, *Fusarium* spp., antagonist, *Bacillus halotolerans.*

PBO6: Effectiveness of biotized substrate based on date-palm wastes peat and native *arbuscular mycorrhiza*l fungi on lettuce transplants performance

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Background and aim: Quality of vegetable seedlings is affected by nursery management practices essentially growth media and fertilization. Sphagnum peat has been widely used as standard media for seedling production. However, there is an increasing concern about the environmental impact associated with its extraction. For sustainable greenhouse horticulture, it is important to characterize biologically-based approaches to improve transplants quality. The aim of this study is to investigate the possibility of developing a biotized growth substrate for transplant production, based on arbuscular mycorrhizal fungi (AMF) and datepalm wastes peat as alternative of commercial sphagnum peat.
Methods: A greenhouse experiment was established in order to assess the possibility of developing a biotized substrate for nursery production of lettuce (*Lactuca sativa* L.) seedlings. Different rate of date-palm wastes peat (0, 25%, 50%, 75%, 100%) were tested as total or partial substitute of Sphangium peat. Lettuce seedlings were inoculated with and without different consortia of native AMF isolated from plots with conventional or organic production system. Lettuce plantlets performances were evaluated in terms of growth and mycorrhizal parameters.
Results: When lettuce plantlets attained commercial size, a microscopic observation revealed that the establishment of mycorrhizal association was successful in all mixtures of growing media. A significant increase in growth parameters (germination, height shoot, collar diameter, biomass) were observed on lettuce transplants inoculated with indigenous AMF isolated from organic plot compared to those isolated from conventional plot and to control (without mycorrhizal inoculation). Commercial peat partial-substitution with date-palm wastes peat in proportion of 25 % to 50% improves better the growth of lettuce seedlings than the complete-peat substitution. Thus these mixtures seem to be an appropriate formulation for sustainable nursery production.
Conclusion: It can be concluded that date-palm wastes peat is a promising substitute substrate for commercial peat and inoculation with native AMF consortia enable stronger growth of seedlings in nursery stage.
Keywords: Biotization, Date-palm wastes peat, growing media, native arbuscular mycorrhiza, nursery production

PBO7: Induction of tomato plant defense against *Fusarium* wilt disease using *Trichoderma harzianum* and *Meyerozyma guiliermondii*

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Background and aim: The vascular wilt pathogen, *Fusarium oxysporum f. sp. radis lycopersici* (FORL) is one of the most devastating pathogens for tomato crop in Tunisia. Management of this disease through the application of benefic microorganisms may be a solution, by enhancing the plant defense response. In this study we evaluate the pathways of inducing the defense oftomato after inoculation by FORL.
Methods: Tomato was carried in hydroponic system with unlimited growth cultivation. Treated Plants with suspension solution of *Trichoderma harzianum* and *Meyerozyma guilliermondii*, were infected by the identified wilt pathogen FORL. Biochemical and molecular analyses performed to improve the induction of defense plants.
Results: The application of *T. harzianum* and *M. guilliermondii* directly in the hydroponic medium reduce the incidence of the *Fusarium* wilt of plants treated, besides, the combination of both species was more effective in controlling the disease compared to their application as alone. After inoculation by FORl, the biocontrol agents and their combination induce the accumulation of phenols and peroxyadase activity in leaves. The phenolic content was recorded to be higher in all the treatments. Moreover, the accumulation of phenolics reached maximum at the 6th day after inoculation. β- 1,3 glucanase and chitinase activity were induced to accumulate at higher levels at 6 days of challenge inoculation in treated plants comparing the control. In this study, molecular analyses appears that the expression of the gene PR, chitinase, glucanase and peroxydase were highly significant expressed in the leaves after the inoculation of plants.
Conclusion: These results suggest that the induction of defense enzymes, involved in the accumulation of peroxydase and phenolics compounds, might have contributed to restriction of invasion of FORL in tomato roots. The combined application of *Trichoderma* and *Meyerozyma* also enhanced the activities ofboth ofthe enzymes and gene defense.
Keywords: Tomato, FORL, enzyme activity, gene defense.

PBO8: Halophilic bacteria able to promote plant growth

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Background and aim: Extremophiles that live in extreme environments produce extremozymes and extremolytes that have the potential to be valuable resources for the development of a bio-based economy through their application to biotechnologies. Here, we provide potentialities behavior of halophilic bacteria in agriculture areas. Plant growthpromoting bacteria (PGPB) are those that can enhance plant growth and protect plants from disease and abiotic stresses through a wide variety of mechanisms. Commonly PGPB could be endophytes bacteria or isolated from the rhizosphere of the plant. In this work, we evaluate the ability of halophilic Bacteria isolated from Tunisian extreme saline soil to promote plant growth.
Methods: The application of bacterial suspension adjusted to 10^7CFU/ml, on tomato or cereals disinfected seeds, on MS media and pot. The determination of antibiotic or volatils compounds, dosage metabolites secondary, IAA, Siderophores, PR-proteins production.
Results: Halophilic bacteria can promote tomato growth compared to untreated one under in vitro conditions on MS media (improvement in weight, plant size and root length). The PGP efficiency of these bacterial species was confirmed by the inoculation of plant seeds on pot. Several plant growth promotion (PGP) traits were produced by our tested bacterial strains. First: various protection characteristics were demonstrated: the production of antibiotic or volatils antifungal compounds against all tested phytopathogenic fungi as well as a nutrient competition, antifungal enzymes, and morphological modifications. Secondly numerous plant promoting characteristics were observed; as siderophores and phytohormones productions. Furthermore an induction of systematic plant resistance and secondary metabolites production such as polyphenols, flavonoids and tannins.
Conclusion: In the light of these data, Halophilic bacteria species success in improving plant growth, but several processes involved can influence the efficiency oftheir soil inoculation, as for example the exudation by plant roots, the bacterial colonization in the roots, and soil health can contribute to increase agronomic efficiency by reducing production costs and environmental pollution.
Keywords: Halophilic bacteria, plant growth promoting,

TC2-PB/PBO9: Root proteome response of Tunisian barley landraces to salinity stress at the seedling stage

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Background and aim: Salinity is a severe abiotic stress factor which limits plant growth and productivity of Tunisian barley landraces. Breeding for higher yield from saline soils needs the understanding of related mechanisms and identification of corresponding traits. Here, genetically characterized accessions previously identified as contrasted pairs under salt stress conditions, were used to analyze the root proteome response at the seedling stage in order to explore biochemical pathways and identify candidate proteins associated with salinity tolerance.
Methods: The Tunisian barley Boulifa (salt-tolerant) and Testour (salt-sensitive) were subjected to a hydroponic salt stress assay (200 mM NaCl) during four days. The total root proteins were extracted and separated by bi-dimensional gel electrophoresis (17 cm IPG strips in the range pH 4-7). Image analyses were performed by PDQuest software and the Student test at 99%. The differentially expressed spots were identified following Mass spectrometry by MALDI-TOF-TOF and searches in Triticeae-NCBI 20170816, Hordeum-JGI 20170816 and NCBI prot 20170125 databases.
Results: Comparative proteomic analyses of control and salt-treated plants showed differences between root proteome responses of both genotypes. Under salt stress, a higher number of differentially expressed proteins was revealed in Testour compared to Boulifa. A combined study ofprotein variation allowed selecting 58 differentially expressed spots based on up-regulation or down-regulation of the protein expression. Analyses grouped these spots into six groups. Among these spots, 49 proteins were successfully identified and were implicated in different pathways. The results showed that tolerance mechanisms in Boulifa genotype would be associated with the up-regulation of proteins involved in selective degradation of proteins, glycolysis, purine metabolism, amino acid metabolism. In addition, other proteins which expression level was maintained in Boulifa and down-regulated in Testour would be also implicated.
Conclusion: The root proteome response analyses showed notable differences between the selected barley landraces at the seedling stage. The identification of proteins allowed the annotation of some metabolic pathways which would be associated with salt tolerance mechanisms of Tunisian barley. The validation of the defined proteins will be investigated in order to select key traits.
Keywords: *Hordeum vulgare*, proteomics, root, salt tolerance.

PBO10: Grain proteome analysis of the Tunisian barley landraces with contrasting salinity tolerance

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Background and aim: Barley is considered to be one of the most cereal crops ranking second in terms of production in Tunisia. However, in arid and semi-arid regions, salinization has severely affected its growth and yield. Breeding strategies are therefore needed for an efficient selection and improvement of genotypes in order to directly exploit saline soils. Here a comparative proteomic study of the albumin/globulin grain fraction of Tunisian barley landraces with contrasting salinity tolerance was performed in order to identify biochemical traits.
Methods: Mature grains of selected Tunisian barley Boulifa (tolerant) and Testour (sensitive) were used for albumins and globulins fraction extraction. Proteomic analyses were based on bidimensional electrophoresis with 17 cm IPG strips in the range pH 4-7. To assess protein abundance, proteome profiles were compared using PDQuest softw are based on Student test ($p<0.05$). Spots with an accumulation enhanced by 1 .5-fold in grain were subjected to MALDI TOF/TOF MS and Barlex database search for identification.
Results: Soluble protein extraction and 2-DE were successfully applied. Image analysis by PDQuest software of all gels led to the detection of sixty-four spots with significant differential accumulation. Notably, in Boulifa, 40 spots were more abundant compared to Testour. Additionally, four genotype specific spots were detected in the tolerant genotype. Mass spectrometry spot identification allowed determining three groups (i. abundant proteins in Boulifa; ii. accumulated proteins in both genotypes; iii. proteins with higher expression in Testour). Considering the protein annotations, acquisition of salinity tolerance in Boulifa would be associated to the abundance of proteins involved in protein folding, processing, defence, desiccation tolerance and in osmotic homeostasis as heat shock and late embryogenesis abundant proteins. Moreover, additional pathways such as pentose phosphate, cell wall modulation and growth, starch metabolism, tricarboxylic acid cycle, lipid signalling and organelle transport were also defined.
Conclusion: Proteomic contrast between selected genotypes was clearly distinguished. Specific heat shock and other defined proteins were identified and validation will be investigated. Key candidates would provide useful biochemical traits for barley breeding programs to face salinity threat.
Keywords: Barley, grain, proteome, albumins and globulins, salinity tolerance.

PBO11: Effect of Salt on spike development and final yield in Tunisian Barley

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Background and aim: The big challenges which we face in the twenty-first century are producing sufficient food to meet demands of the increasing global population. Abiotic stresses caused by climate change, such as salinity, drought, low or high temperatures, flooding and frost are predicted to become more severe and widespread. The losses of crop productions due to salinity and drought stresses have increased progressively over the last decades. Common cereal members such as wheat (*Triticum spp.*), maize (*Zea mays* L.), rice (*Oryza sativa* L.) and barley (*Hordeum vulgare* L.) are the main important members of most worldwide nations in food and feed system. Barley is one of the oldest cereal crops known to be cultivated since about 10,000 years. It has a natural tolerance to salinity, drought, and fungal diseases, thus making it a model plant in stress biology research. We aim in this study to understand the effect of salinity stress on the spike developmental stages and the final grain yield of five Tunisian barley cultivars.
Methods: Experiments were performed at IPK-Gatersleben, Germany. Seeds were sown under controlled condition in green house (LD condition, 16/8 h day/night and ⁓ 20/ ⁓ 16°C day/night). A saline treatment with 10gNaCl/l was imposed starting 7 days after sowing and was compared to a nonsaline control treatment.
Results: Results obtained show that, in addition to its impact on plant growth, salt has a severe and marked effect on spike development and final yield. It caused delaying in inflorescence developmental stages and decreasing in floral units numbers. Anthesis and flowering also occurred later in treated plants compared with control plants. The measurement of ten main-spike components indicated that salt negatively affected number of spikelets primordia/spike, number of grains/spike, spike length and thousand grain weight, as well as number of tellers, number of leaves and plant height. Cultivars Ardhaoui and Rihane showed to have faster spike developmental stages and more yield than the other three under salt stress conditions.
Conclusion: Delaying in the spike development and reduction in spikelet primordia numbers decrease the yield in salt-stressed barley are the main results.
Keywords: Tunisian barley, salinity, spike development, yield, growth traits.

PBO12: Genetic variation of ionic accumulation in roots and leaves levels under salinity stress in the Tunisian populations of *Hordeum marinum ssp. marinum*

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Background and aim: Genetic improvement of salt tolerance is of high importance due to the extent and the constant increase in salt affected areas. The aim of this work was to evaluate the genetic variation of salinity tolerance of *Hordeum marinum*i n Tunisia by using physiological indicators.
Methods: In the present study we examined growth and ionic contents in leaves and roots of 150 lines of *Hordeum marinum ssp. marinum* in response to salt stress. Plants were grown in pots at 0 and 200 mM NaCl in a randomized block design with three replications and were harvested at seedling and at maturity stages.
Results: The results showed that studied populations differed dramatically in their responses to salt in terms of plant biomass and ionic accumulation in roots and leaves. The Na^+ content in both leaves and roots increased with increases of salinity. Increased treatment levels of NaCl induced decreases in K^+ and to a lesser degree in Ca^{2+} and Mg^{2+}in all the plants. Regardless the presence or absence of the salt in the medium culture, the amount of Na^+ at the leaves exceeds that of the roots. Even though, the reduction of potassium is much more pronounced in leaves than roots in all the studied populations, K^+ contents are approximately two to three folds higher in leaves than those in the roots. Consistent with these findings, the ratio K^+/ Na^+ was lower in roots than in leaves, indicating discrimination in Na^+ and K^+ transport. The relation of biomass with respect to the ratio of selectivity showed that the tolerance to salinity is related to the maintenance of the ratio K^+/ Na^+ comparable to the control medium in leaves and roots. The populations Mouthul and Bkalta showed high salinity tolerance, maintaining significantly higher ratios K^+/ Na^+ and higher dry weight, whereas Lessouda was the most affected.
Conclusion: Overall, the reported results may provide novel insight into the potential responses associated with salt stress in Tunisian populations of sea barley.
Keywords: *Hordeum marinum ssp. marinum*, genetic diversity, salt stress, cation accumulation, growth, cation/ Na^+selectivity.

PBO13: Identification of the NaCl-responsive metabolites in Citrus roots: A lipidomic and volatomic signature

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Background and aim: The main objective here was to exploit a targeted metabolomic method utilizing gas chromatography mass spectrometry (GC-MS) in order to identify metabolic signatures in response to salt-stress in *C. aurantium* roots. Specifically, this study examined the influence of salt treatment on fatty acid composition and chemical composition of the essential oil of *C. aurantium* roots.
Methods: Here, targeted metabolomics utilizing gas chromatography mass spectrometry (GC-MS) was employed for the first time to identify metabolic signatures in response to salt-stress in *C. aurantium* roots. Specifically, this study examined the influence of salt treatment on fatty acid composition and chemical composition of the essential oil of C. *aurantium* roots. A lipidomic and volatomic profiling proved an altered emission/accumulation of chemical compounds affected by salt stress.
Results: Under salt treatment a redirection of the lipidic metabolism towards synthesis of unsaturated fatty acids as revealed by the increase of double bond index and linoleic desaturation ratio was pointed out. On the other hand, reduction of saturated fatt y acids was spotted. Such treatment also induced marked quantitative changes in the chemical composition of the essential oils from C. aurantium roots and increased markedly the rates of monoterpenes, while the sesquiterpenes decreased significantly. On the basis of the parameter VIP> 1, both primary and secondary metabolites were found to be significantly salt responsive, including 1 fatty acid (palmitoleic acid) and 6 volatiles (E-2-dodecenal, tetradecanal, γ-Elemene, trans-caryophyllene, α-Terpinene and germacrene D). This metabolomic analysis resulted in an identification of a signature containing metabolites potentially implicated in response to salt stress.
Conclusion: Plasticity at the metabolic level may allow Citrus plants to acclimatize their metabolic ranges in response to changing environmental conditions. Information derived from metabolite datasets can provide a blueprint framework for better understanding the mechanisms that govern Citrus plants response to salt stress, with insights into molecules that can be used for crop improvement.
Keywords*: C. aurantium*; roots; lipidomics, volatomics; salinity; responsive metabolites.

PBO14: Delta-7-stigmastenol: quantification and isomeric formation during chemical refining of olive pomace oil and optimization of the neutralization step

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Background and aim: The aim of this study is to evaluate the formation of the increase of Δ-7-stigmastenol during the chemical refining of pomace olive oil (POO) and the optimal neutralization by NaOH concentration of 20 °Be at a temperature of 70 °C. A comparison has been made between virgin olive oil (VOO) and consecutive steps of refining process in the amounts of Δ-7-stigmastenol of the POO oil samples. Among the oils, refined olive oil particularly the neutralized olive oil (NOO) by soda (NaOH) contained a high-level of Δ-7- stigmastenol.
Methods: Determinations of free fatty acids (*FFA*) were made following ISO procedures (*ISO 660, 2009*). The analyses of ultraviolet (*UV*) evaluation (*K270*), trans-fatty acids (*TFAs*), waxes esters, sterols, and triterpenic dialcohols were performed according to the methods of the International Olive Council.
Results: A mean result found in NOO by different concentration of NaOH from 15 to 25 °Be showed increased values significantly ($p < 0.05$) from 0.70 ± 0.01% to 0.78 ± 0.01% of Δ-7- stigmastenol and increased significantly ($p < 0.05$) the levels of erythrodiol and uvaol from 26.34 ± 0.39% to 28.11 ± 0.42%. Then, the concentration of the Δ-7-stigmastenol was evaluated using a GC–MS instrument. Besides, further analyses were performed to ensure the uniqueness of the peak of Δ-7-stigmastenol and absence of any overlap. In all cases, the level of Δ-7-stigmastenol was higher than the limit set by the International Olive Council.
Conclusion: The most important step can cause a change in the sterol composition which is the neutralized step, this variation can affect the final product. Our findings, the alkaline neutralization of POO by NaOH also transforms a part of the *β*-Sitosterol into Δ-7- stigmastenol by delocalization of the double bond. The content of Δ-7-stigmastenol by NaOH in POO was significantly higher than in oils neutralized by Ca(OH)2. Behind this analysis, the purpose of this study is to focus only on parameters that have significant influence on Δ-7- stigmastenol rather than focusing on several insignificant parameters. To provide oils that meet the expectations of consumers and industrial user (Δ-7-stigmastenol < 0.5%, IOC).
Keywords: Δ-7-Stigmastenol, Chemical refining processes, Neutralization, Optimization, GC/MS

PBO15: The resistance of the olive tree varieties to the leaf-spot disease: The study of the phenolic metabolism

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Background and aim: The leaf-spot disease caused by *Spilocaea oleagina* represents the most widespread fungal disease of olive-tree (*Olea europea)* in the world. The aim of this work is to explore the role ofphenolic compounds metabolism in the defense of the olive-tree against this fungus.
Methods: The defense components of two varieties of olive-tree: a resistant (Picholine Languedoc) and a susceptible (Moroccan Picholine) were studied. Parietal phenols are extracted from the residue of cell walls after depletion of soluble compounds. They are characterized by various chromatographic techniques and identified by their retention time. The phenolic compounds and the lignin contents are determined by reading the optical density respectively at 760nm and 280nm against a control. While, the phenylalanine ammonia-lyase activity was determined after the extraction ofthe trans-cinnamic acid.
Results: The inoculation of olive-tree leaves by *Spilocaea oleagina* induces a foliar necrosis whose speed of onset and expansion distinguishes clearly the two studied varieties according to their behavior to the leaf-spot disease. For the resistant variety, these symptoms are composed of small necrotic lesions, whereas for the susceptible variety, they appear as extended necrotic spots. These symptoms are accompanied by an increase in the accumulation of the contents of soluble and parietal phenols, the intensification of the lignification and the induction of the phenylalanine ammonia-lyase activity of which the speed and intensity plainly distinguish both varieties under study.
Conclusion: The response of phenolic metabolism to the resistance of the olive tree to the leaf-spot disease appears to occur in the early stages of infection leading to an increase in the biosynthesis of the contents of three defense components (soluble phenols, parietal phenols and lignin).
Keywords: Lignin, Olive, Phenols, Phenylalanine ammonia-lyase, *Spilocaea oleagina.*

POSTER PRESENTATIONS
(PBP)

PBP1: Comparative study of exogenous proline effects on *Triticum aestivum* l. and *lens culinaris* medik. tolerance to drought

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Background and aim: Accumulation of compatible solutes such as proline has been often regarded as a basic strategy for tolerance of plants under water stress. Thus, exogenous application of proline is a possible technique to avoid the deleterious effects of the drought on plant growth. The objectives of this study are to investigate the impact exogenous proline on the physiological behavior of two plant species, bread wheat (*Triticum aestivum* L.), a monocot, and lentil (*Lens culinaris* Medik.), a dicot, under water stress conditions.
Methods: After several preliminary tests, optimal concentrations of exogenous proline were determined (6 Mm for bread wheat and 2 mM for lentil) and both species were treated in normal and drought conditions. Biometric, physiological and biochemical parameters were measured and compared to control.
Results: The results showed that water deficit affected both species leading to a reduction in growth and chlorophyll content. Likewise, the polyethelene glycol (15 % PEG-6000) caused a strong production of Malondialdehyde (MDA). We also noted a remarkable decrease in Catalase (Cat) and Ascorbate Peroxidase (APX) activities which was probably due to the oxidative stress caused by water stress. Exogenous proline counteracted the effects of PEG6000 on physiological and biochemical parameters. The application of proline in stressful conditions reduced the deleterious effects caused by the stress on both species, due, particularly, to increase of chlorophyll content and Cat and APX activities. This mitigation effect of exogenous proline was especially noticeable for bread wheat.
Conclusion: Application of exogenous proline in stressful conditions could increase the tolerance ofbread wheat and lentil plants.
Keywords: Drought, PEG-6000, Wheat, Lentil, Exogenous proline, Tolerance.

PBP2: Optimization of ultrasound extraction of *Phoenix dactylifera L.* polyphenols by response surface methodology and their antioxidant activities

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Background and aim: Optimum conditions for extracting phenolic compounds and antioxidant activity by ultrasound extraction from date fruit *(Phoenix Dactylifera L.)* have been investigated using RSM.
Methods: The Box–Behnken design was used to investigate the effects of three independent variables, time, temperature and solvent-to-solid ratio, on the response.
Results: Regression analysis showed that most of the variation was explained by the models. P-value for the lack of fit was insignificant which confirmed the validity of models. Response surface analysis showed that the optimal extraction parameters that maximized antioxidants extraction were 30 min, 25 C° and 74.4mL/g. Under optimum conditions the experimental values for TPC, TFC, DPPH and CUPRAC were 1 .634mg GAE/g DW, 0.3095mgQE/g DW, 60.683 1 mg EAA/100g DW and 293.3422 mg EAA/100g DW respectively.
Conclusion: The ultrasound extraction process was successfully applied to optimise the conditions for the extraction of antioxidant compounds from date fruits. The experimental values are in accordance with those predicted, indicating the suitability of the model and the success of RSM in optimizing the extraction conditions. This study can be useful to promote industrial extraction of date antioxidants.
Keywords: *Phoenix dactylifera* L.; Polyphenols; Ultrasound extraction; Response surface methodology; Antioxidant activity.

PBP3: The physiological behaviour of two varieties of durum wheat (*Triticum durum Des.f*) under drought conditions

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Background and aim: Global warming is expected to worsen drought in the Mediterranean areas and this may limit the plant growth of cereals in the semi-arid environment of Tunisia. The aim of this investigation was to evaluate the effect of water stress on the physiological behaviour of two durum wheat varieties: Karim and Razzak. For this reason, the dynamic of soil moisture and its relations with the leaf water potential, photosynthesis, transpiration and stomata conductance were followed.
Methods: A randomised block design with four replicates and two varieties was adopted for the pot experiment. The experiment was carried out at the experimental station of the National Institute of Agronomic Research of Tunisia (INRAT) at Ariana (Tunis), in a typical semi-arid climate. Water stress was applied at different development stages: irrigation was stopped when the stage of development is initiated whereas the control pots were irrigated to 70% of reserve in useful water.
Results: It was showed that water deficit resulted in stomata closure which was followed by reduced plant photosynthesis and transpiration. Moreover, the shortage of water leads in variation physiological activity with the stage of development.
It was revealed also that Karim variety may support drought stress better than Razzak variety due to its developed and performed root system which allows it to extract water at very low leaf water potential even over permanent wilting point.
Conclusion: It was demonstrated the great susceptibility of the tillering stage compared to others. It was seemed that the young plants tend to avoid the water stress better than the old ones which respond to the deficit in water by a form of tolerance.
Keywords: durum wheat, water stress, physiological parameters, moisture, stage of development.

PBP4: *Vigna unguiculata* seed priming is related to redox status of gemmula, radicle and cotyledons

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Background and aim: Pre-germination treatments represent the physiological methods that improve plant production by modulating the metabolic activities of germination before the emergence of the radicle. This treatment consists of an osmotic (osmopriming) or a hormonal (hormopriming) and/or redehydration (hydropriming) treatments. The approach allows the elimination of dormancy, homogenization (synchronization) of germination, better growth, earlier flowering and a tolerance to abiotic stresses such as drought and salinity. It was suggested that ROS play a crucial role in signaling seed germination. Our work consisted in studying changes in the redox status in the embryonic axis (radicle and gemmula) and in cotyledons without and with priming, by estimating antioxidant activities and production of reactive oxygen species (ROS).
Methods: In the cotyledons, radicle and gemmula taken from control *Vigna unguiculata* seeds and having undergone osmopriming, simple hydropriming and double hydropriming **s**ome antioxidant enzymatic activities (SOD, Cat, APX, GPOX) as well as total nonenzymatic antioxidant capacity (TAC) were measured. The production of hydrogen peroxide (H2O2) by DAB and superoxide anion (O2 -) by NBT was also assessed.
Results: The results obtained showed that priming allows activation of antioxidant enzymes, especially in the gemmula. On the other hand, our results showed that priming caused an accumulation of ROS in embryonic tissues. This may also explains the improvement of seed germination performance according to the oxidative window model.
Conclusion: Priming induced changes in the redox environment at the seed level. These changes were closely related to the type of seed initiation. Indeed, a double cycle of hydration-rehydration induced the broadest spectrum of modifications of the redox status by inducing better germination and better growth under favorable or unfavorable conditions.
Key words: germination; priming; antioxidants; redox; ROS; pre-germination treatments; *Vigna unguiculata*.

PBP5: Effect of water Deficit during the Flowering Period of Grass Pea (*Lathyrussativus* L.)

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Background and aim: Grass pea (*Lathyrussativus* L.) is a multipurpose grain legume with an indeterminate growth habit. It offers an attractive choice for sustainable food production, it is a legume crop in arid and semi-arid areas, is widely acknowledged as highly drought tolerant and have great agronomic potential as grain and forage legumes. Grass pea is a promising crop for adaptation under climate change because of its tolerance to drought and salinity, and being almost free from diseases.
Methods: The experiment design was split- plot with six replications. When plants reached flowering, drought was imposed to stress plots by withholding water, while non-stressed plots continued receiving irrigation.
Results: The objective of this study was to identify adaptation mechanisms of grass pea to water deficits. This was done by imposing water deficit during the reproductive period on plants of *L. sativus* grown in pots in a glasshouse. In this experiment, water deficit was imposed on plants of grass pea by withholding water from first flowering. At maturity, dry matter, seed yield and number of pod decreased significantly compared with the control. This study showed that under water deficit, grass pea avoids dehydration through a reduction in leaf area and fresh biomass.
Conclusion: Grass pea offers an attractive choice for sustainable food production, it is a legume crop in arid and semi-arid areas, is widely acknowledged as highly drought tolerant and have great agronomic potential as grain and forage legumes. Therefore it seems to be a suitable candidate for breeding programs. The forage pea has been shown to exhibit abiotic constraints and their association with grasses may be beneficial in forage crops.
Keywords: *Lathyrussativus L*; Water deficit; Flowering period; Dry matter, Grain and pod number.

PBP6: *Cytisus villosus* benefits on growth and cork quality

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Background and aim: Cork oak (*Quercus suber L.*) is a species of great ecological, economic and social value with a natural distribution limited to the western Mediterranean basin. The cork oak is the native hardwood forest species the most represented on the Tunisian territory Although still productive, from 80,000 to 100,000 quintals of cork per year, this forest remains constantly threatened (DGF, 2010). Actually, it was in a severe degradation, accentuated during these last decades by various factors: aging of stands, stands in southern exposure, fires, insects and fungi attacks, human overexploitation, and recently by the climatic changes characterized these last years by marked summer drought. The association of cork oak with autochthonous plant species is an original research action envisaged by INRGREF institute to resolve this problem. As part of this reflection, the association of the cork oak with leguminous species such as *Cytisus villosus* could positively influence its growth by providing the nutrients necessary for its survival in the face of climatic constraints.
Methods: A morphological study of cork oak trees and their cork quality (porosity, humidity, durability....) was carried out in four cork oak populations in Feija and Ain Drahem areas in Jendouba gouvernorate (AD1 with Cytisus; AD2 without Cytisus ; F1 Cork oak with Cytisus and F2 without Cytisus).
Results: Morphological characterization (height, circumference) showed significant differences especially in AD1 population with highest diameters. Statistical analysis has shown that AD2 oaks have poor corks quality. The same, cork oak trees in F1 population have a higher diameters, higher height and better cork quality than in F2 population. Between areas, the cork oaks with *Cytisus villosus* at Feija have the caps of good qualities.
Conclusion: We can conclude that the cork oak behaves better and have higher growth and better cork quality in the presence of legumimous species like Cytisus villosus.
Keywords: Cork oak, Cytisus villosus, cork quality, association.

PBP7: Ethnobotanical study of medicinal and aromatic plants used in the city of Ain Temouchent in Algeria

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Background and aim: Medicinal plants constitute a precious heritage for the humanity, they are natural chemical factories, producing biochemical active substances. In Algeria, we resorted for a long time to the traditional medicine thanks to the wealth and the diversity of its flora. Ethnobotanical study was made in order to gather general information about the therapeutic uses practiced in the region of Ain Temouchent.
Methods: The ethnobotanical investigation was realized with 500 informants at random of different ages, gender and intellectual levels. Who informed us about the therapeutic and traditional local applications of the population of Ain Temouchent with the help of questionnaires beforehand established containing information concerning the informant profiles, information on the nature and pharmaceutical techniques of the medicinal plant used, information on their use in the treatment of different kind of diseases.
Results: The surveys accomplished allowed us to identify 77 species belonging to 44 different plant families. The family of Lamiaceae is the most dominant. The analysis of the results, concerning the relations existing between the medicinal species and types of diseases treated has shown that most of these species are used primarily in the care of the digestive and respiratory system. These devices are handled mainly by the foliage which is the plant organ most used and the infusion which is the method of preparation the most dominant.
Conclusion: This work is a source of information that can serve as a basis for pharmacological studies to evaluate the therapeutic efficacy and safety of medicinal plants.
Keywords: Medicinal plants, Ain Temouchent, Ethnobotanical study, Traditional medicine, Survey.

PBP8: Biodiversity of Faba bean (*Vicia faba* Minor) genotypes within response to combined salt-flooding stresses: Modulation of growth capacities and metabolic adaptations

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Background and aim: Tunisian agricultural lands are widely belonging to the semi-arid bioclimatic stage characterized by the abundance of drought stress. Paradoxally, these regions are submitted to seasonal flooding periods that affect huge surface of cropping systems. Aiming at investigating biotechnological solutions to these situations, the present study focuses on the selection of salt and flooding tolerant faba bean genotypes and the isolation of efficient associated rhizobial strains to enhance plant productivity and soil quality in marginal soils.
Methods: Six varieties of faba bean (*Vicia faba Minor*) were grown on either salinized or non-salinized soils, and subsequently submitted to flooding (resulting in root hypoxia). Leaves and roots were analyzed at biometric, physiological and biochemical levels to explore plant biodiversity toward stress response.
Results: This study showed that the six analyzed varieties had different behaviors depending on the soil on which they are grown. Overall one variety (Najeh) showed the best adaptation to stressed conditions. Based on the production of biomass and the ability to form nitrogen fixing nodules, the Najeh variety is the most productive and shows the best tolerance when grown in saline and flooded soils. The analysis of salinity response markers (proline) and hypoxia (alanine) does not differentiate the different varieties. On the other hand, levels of accumulation of nitrogen compounds (nitrate, nitrite ammonia) and of enzymatic activities nitrate reductase and nitrite reductases, suggest that the nitrate reduction pathway in response to hypoxia is more developed in the Najeh variety.
Conclusion: The applied aim of our study is the selection of faba bean varieties that can perform relatively high growth rate, high biomass production, good nodulation and achieve their life cycle in salinized soil under flooding stress. Our results suggest the possibility to exploiting this kind of immerged soils by using flooding-salt adapted cropping systems.
Keywords: Biodiversity, combined salt-flooding stresses, Faba bean, growth capacities, metabolic adaptations.

PBP9: Insecticidal effects of *Ziziphus jujuba* Mill. and *Ziziphus lotus* L. roots and barks on *Tribolium castaneum* development

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Background and aim: *Ziziphus* species possess multiple medicinal properties such as antifertility, analgesic, and antidiabetes. The aim of this work is to estimate the insecticidal properties of roots and barks extracts (aqueous and ethanolic) of four provenances of *Ziziphus jujuba* and *Ziziphus lotus*.
Method: Braks and roots of *Zizphus* species were collected from five localities: Mahres, Mahdia, Sfax (*Ziziphusjujuba*) and Souasi, Kasserine (*Ziziphus lotus*). Collected organs, airdried and grinded, were macerated each with 1000 ml of distilled water for 24 h. The extracts were filtered through Whatman N°1 filter paper, pooled and concentrated under vacuum. The topic application, used to investigate the insecticidal activities, consisted to insert 10 insects of *Tribolium castaneum* aged from 7 to 10 days in Petri dishes. 2 mg of crud extract/ml of solvent were than deposed on each insect using a micropipette. Essays were kept at temperature of 30°C and Petri dishes were sealed with plastic wrap to prevent the loss of moisture and avoid contamination. The control groups were prepared in the same conditions. The lethal concentrations $_{LC50}$ ^{and} $_{LC90}$ of each extract against the *Tribolium castaneum* larves were calculated. Insects were considered dead if it did not move when observed out of the container.
Results: Results showed that lethal concentrations varied among the provenance, organs and species. For roots, *Ziziphus lotus* of kasserine provenance were the most toxic (LC50= 2.07). Mahres provenance posses the less level of lethal concentrations (LC50=5.29µl/l air). This effect became more important for braks and lethal concentrations levels ranged between 1.09 and 3.37 29µl/l air for Mahdia and Sfax provenance, respectively.
Conclusion: *Ziziphus lotus* barks could be included as an insecticide agent originated from a natural resource.
Keywords: Lethal concentrations, provenance, *Tribolium castaneum*, *Ziziphus* organs.

PBP10: Effect of Pectine Lyases (Pel) and Phenyl Alanine ammonia-Lyase enzyme (PAL) on disease severity in potato and carrots infected by plant pathogen bacteria (*Dickeya* and *Pectobacterium* sp)

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Background and aim: Potato diseases, due to *Pectobacterium* and *Dickeya* bacteria, constitutes a serious handicap for Algerian agri food economy by inducing important losses in culture or/and storage conditions. Given the ineffectiveness of conventional control methods, it becomes imperative to develop alternative technics based on the detection and quantification of metabolites released during infection process. Our study's aim is highlighting the implication of pectinases and PAL enzymes in the induction of systemic resistance.
Methods: 02 potato cultivars (Désirée; Spunta) and 1 carrot variety are used. Samples were infected by inoculating a bacterial solution from 3 strains "*Pectobacterium carotovorum* (8822), *P. atrosepticum* (5384) and *Dickeya solani* (99)". 100 µl are taken from infected tissues and centrifuged twice in order to quantify Pectate lyases enzymes by spectrophotometer (230 nm). PAL activity was detected by using Borate Buffer, and with Phenylalanine as precursor.
Results: Following data collected by calculating infection parameters (weight, length, water volume and depth of macerated tissues), we can assume that whites kinned cultivar Spunta show less susceptibility to pathogens then Désirée (Red Skin) regarding the values. Moreover, atrosepticum seems to get more virulence comparing to *Carotovorum* and *Dickeya* strains. In addition, *Pa* had highest rates of pectinase activity especially on carrots (> 12 IU), while cultivar Spunta present lower enzymes production then Désirée.PAL enzymes are also better induced by susceptible cultivar Désirée comparing to Spunta. Pels contents are more available in intermediary tissues then rotten ones. At the opposite, PAL are most presents in rotten area comparing to intermediary tissues.
Conclusion: PAL accumulation are related to vegetal reaction towards pathogens, wich show its role in inner resistance. So, we constate that highestyields of PAL were found from susceptible potatoes as a direct response to pathogen infection.
Keywords: Enzyme activity, PAL, Pectobacterium, soft rot, potato.

PBP11: Fatty acid of the lipid from the leaves of *Centaurea sp*(Asteraceae) from Algeria

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Background and aim: Plants are a precious source of interesting biologically-active compounds. Biological and chemical screenings are complementing approaches for the rapid detection of plant constituents.Additionally,in order to replace the synthetic products, the quest for effective natural compounds with antioxidant activity has been intensified in recent years.
Methods: The various parts of the plant Centaurea-Sp were collected monthly from (January until July), at the middle of each month in 2017 from Dellys (located in the central east of Algeria, limited 36°54'48"nord, longitude 3°45'51"est altitude 200m). The fatty acids FA composition of the lipid was determined after having converted the fatty acids FA into FA methyl esters (FAMEs) then analyzed by gas chromatography (GC).
Results: For the overall samples were collected monthly from (January until July), the main saturated fatty acids components were lauric acid (C 12:0 = 3.53 -16.64%), myristic acid (C14:0 = 0.09 –1.67%) and palmitic acid (C16:0 = 11.73 - 25.37%). Alternatively, the main Monounsaturated fatty acids were palmitoleic acid (C16:1W7 = 0.76 -10.31 %) and Oleic acid (C18:1W9= 3.51–17.94%) and most importantly linolenic acid (C18:3ω3 =0.7–26.98 %), which is known for its multiple dietary, pharmaceutical and clinical benefits.
Conclusion: The results of fatty acids methyl esters identification using (GC)showed a clear variability in the composition of fatty acids. The main result of this study illustrates the nutritional potential (richness in Saturated fatty acid such as C 12:0), presence of essential fatty acids such as (C18:2W6 = 21.84 % in March)at all the stages of maturation, richness in omega(W-3) fatty acids such as (C1 8:3W3 = 26.98 %in April)of the lipid of ***Centaurea sp*** leaves, which can provide opportunities for rational exploitation for medicinal products.
Keywords: Extraction, Soxhlet, Centaurea Sp, Lipid, Fatty acid, gas chromatography.

PBP12: Beneficial effects of silicon in sea barley (*Hordeum marinum* Huds.) under cadmium stress

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Background and aim: Silicon (Si)represents the second most abundant elements, after oxygen, in soil. Although silicon is not listed among the essential elements for higher plants, it has been recently documented to play an important role in providing beneficial effects on growth and yield, especially in plants under stressful environments, such as cadmium (Cd) stress. Cd is a metal pollutant that accumulates in soils and plants and has detrimental consequences in terms of food safety. The aim of this study is to investigate the role of silicon (synthesized from Tunisian silica sand) in the alleviation of the negative effects of Cd in sea barley (*Hordeum marinum* Huds.)and in improving the mechanisms of tolerance. This species, with forage potentialities, presents a very interesting capacity for rehabilitation of non - productive marginal areas.
Methods: In this study, sea barley plants were grown for 17 days on media supplemented with silicon (0, 1 ,or 2 mM Si)under cadmium (0 or 25 µM CdCl2).
Results: Our results showed that Cd stress altered plant growth, water status, chlorophyll content, photosynthetic gas exchanges, chlorophyll fluorescence as well as membrane integrity.
However, these effects were significantly mitigated when we added Si. Indeed, Si restored morphological aspect of plants, improved negative effect on growth parameters, and established water status, and photosynthetic activity. Interestingly, silicon had a high potential to contribute to toleration of cadmium toxicity and mitigate its adverse effects, likely through alleviate Cd uptake.
Conclusion: Such behaviour showed the benefits of silicon applications on sea barley plants grown under Cd stress.
Keywords: Silicon, Cadmium, *Hordeum marinum*, growth, water status, photosynthetic activity.

PBP13: Effects of drought stress on some physiological parameters and stressrelated gene expression in two genotypes of *Vicia faba* L. Minor

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Background and aim: Currently, there is a great interest in improving *Faba bean* tolerance to abiotic constraints especially water deficit. For this purpose, physiological and molecular studies are needed. Here, we assessed the potential implication of two *WRKY* transcription factors as well as other important physiological parameters in drought stress response in two faba bean genotypes.
Methods: Hara and Giza 3 Faba bean cultivars were cultivated under optimal, moderate and limiting water supply (90, 60 and 30% of field capacity (FC), respectively) as well as under PEG (20%) treatment. Thereafter, these plants were used for *VfWRKY1* and *VfWRKY2* kinetic expression analysis by q-RT-PCR analysis besides of evaluating some physiological traits.
Results: Under water stress conditions, Giza 3 genotype showed the most pronounced decrease in the total chlorophyll content mainly under 30% of FC compared to the control (90% of FC). Also, Giza 3 exhibited the highest level of lipid peroxidation (5 5%) compared to that of Hara genotype under drought stress. Hara genotype showed an enhanced accumulation of osmolytes such as soluble sugar (58 1%), and proline (128%) compared to those of Giza. Moreover, Hara genotype showed an enhanced accumulation of antioxidant enzymes compared to those of Gisa 3 including superoxide dismutase (37%), catalase (547.11%) and ascorbate peroxidase (75.6%). On the other hand, significant differences were observed between the two genotypes (Hara and Giza 3) regarding the expression of *VfWRKY1* and *VfWRKY2* genes under osmotic stress. In fact, *VfWRKY1* and *VfWRKY2* were more rapidly expressed and highly induced in Hara genotype compared to those in Gisa 3.
Conclusion: Our study demonstrated contrasting physiological responses to water and / or osmotic stress among *Vicia faba* genotypes (Hara and Giza 3). The fact that *WRKY*regulators were differentially expressed in the contrasting genotypes; this suggest their involvement in mitigating drought stress in *Vicia faba* L.var.minor.
Key words: *Antioxidant enzymes, RT-PCR, water stress, VfWRKY, Vicia faba.*

PBP14: Diversity, geographic distribution and biocontrol of fungal strains infecting field-grown common bean (*Phaseolus vulgaris* L.) in Tunisia

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Background and aim: Common bean (*Phaseolusvulgaris* L.) crops are facing serious surfaces and production declines in Tunisia; resulting in high price for consumers. One major limiting factor is attributed to fungal diseases. We propose in this study (i) to characterize the fungal species causing the threats, and (ii) to assess the biocontrol potential of common be an associated bacteria against the pathogens.
Methods: A collection of 103 fungal strains was established from infected field-grown common bean in three geographic regions Boucharray, Chatt-Mariem, and Metline. Fungal isolates were identified using ITS region. Endophytic and rhizospheric bacteria were characterized, tested for antibiosis and biocontrol properties against the fungi, and the antifungal activities were analyzed at molecular and biochemical levels.
Results: The pathogenicity test carried out on Common bean cv. Coco blanc showed that among the fungal collection, 41% of fungal strains were assigned to be highly pathogens. Serious cases of seedling damping-off, and significant reduction in root and shoot biomass were noticed (up to 90% biomass reduction). A wide generic and specific diversity of the pathogenic fungi was noticed. *Fusarium* spp. strains were dominant and represented 67%,fllowed by *Alternaria* (22%), *Rhizoctonia* (4%), *Ascomycota* (4%), *Macrophomina* (1 0%) and *Phoma* (4%). The highest richness levels were found in Chatt-Mariem and Boucharray regions, showing the highest generic and inter specific diversity. A variability of the abundance and geographic distribution of fungal species was noticed. Fungal strains recovered from Metline were represented exclusively by *Fusariumoxysporum*. However, the genus *Fusarium* represented about 66% of fungal strains recovered from Boucharray, and only 20% from Chatt-Mariem. The genus *Alternaria* represented 11% and 40% of total fungal isolates in Boucharray and Chatt-Mariem, respectively. The biocontrol assays showed the effectiveness of *Bacillus* spp. strains to reduce the fungal disease incidence on common bean and showed others PGPR activities. Genes mediating antifungal activities in selected bacteria were amplified and underlined the multifunctional options for their uses.
Conclusion: The present work provides a database that should be considered for surveying common bean fungal-diseases, and proposes a set of promising biocontrol agents for formulation and application programs.
Keywords: Common bean, Fungi, Pathogenicity, Diversity, Biocontrol.

PBP15: Interaction between UV-A radiation and water stress: effects on the biochemical parameters of *Triticum aestivum* wheat plants

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Background and aim: Ultraviolet radiation causes changes in the physiological processes of plants. This sensitivity to UVs can be influenced by other environmental factors, such as drought. The aim of this work is to see, through physiological parameters, how a plant reacts under the effect oftwo combined environmental constraints, water stress and UV-A.
Methods: Two main groups of *Triticum aestivum* wheat plantlets are grown, one of which is exposed to UV-A radiation during its growth while the other group will not be irradiated. Each group is further subdivided into two subgroups, depending on whether water stress is applied. Thus, we count four distinct lots: one having been exposed to UV and water stress (WS + UV), one having undergone only UV (UV) radiation, one having only undergone the water stress (WS), and a last one being the control which has not undergone any of the two treatments (C).
Results: Different parameters were measured for all lots, those parameters are: catalase activity, proline, soluble sugars and photosynthetic pigments contents. Results of the analysis show the same pattern; where there is a small variation between the (WS) and the (UV) lots, and compared to the control lot (C). On the other hand, the parameters values (CAT, Proline, soluble sugars and pigments) in the lot that was subjected to the two environmental factors (WS + UV), are much higher than those found in the other three lots. This is explained by the fact that water stress combined with UV-A rays, exert a very strong pressure on the plant.
But instead of weakening, the plant can show, on the contrary, a very strong resistance to these constraints, as has already been reported in other work. The plant's defense capabilities are magnified by an additive effect.
Conclusion: All the measured parameters clearly demonstrate a synergistic action of the two combined treatments. Hence it could be deduced that the plant is stimulated in its defense mechanisms, in particular the antioxidant systems, by the cumulative effect of the two factors, that are the radiation exposure UV-A and water stress.
Keywords: UV-A, Stress, Drought, Wheat, Antioxidant.

PBP16: Hormo (GA3) priming improves germinative performances and seedlings growth under favourable and stressful conditions on wild and cultivated barley

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Background and aim: Various techniques have been developed to improve the quality of seeds and their resistance to environmental constraints. The most commonly method is known as "seed pretreatment or priming". Several agents are used in this process including hormopriming. The aimed of this study was to investigate the effect of soaking seeds with GA3on the germinative parameters in cultivated and wild barley and to evaluate their growth in a wide range of salinity.
Methods: Seeds have been divided into two lots. The first one was un-primed and the second one was moistened with GA3.Seeds priming was carried out in the dark for 12 hours. After priming, seeds were dried at room temperature for 24 h until they reach their initial moisture. The un-primed and hormo-primed seeds were then sown in covered Petri dishes on two filter paper layers imbibed with NaCl solutions (0, 50, 100, 150 and 200 mM NaCl).Measurements were based on germination parameters, growth parameters, α-amylase activity and osmolyte accumulation.
Results: Soaking seeds in GA3accelerated the germination compared to un-primed seeds by reducing the latency phase. Moreover, it improved the germination parameters (T50%, GI) as well as the final germination rate for*H. vulgare* and*H. Maritimum* Soliman ecotype under favourable and stressful conditions. The measurement of the α-amylase activity showed that the priming of the seeds with GA3 attenuated the effect of salt on this activity. On the other hand, the stimulating effects of hormopriming were well marked on seedlings vigor. The primed seedlings have a better ability to lower their root osmotic potentials depending on the salt medium and to accumulate more compatible compounds for better hydration. Additionally, our results suggested that these compounds (proline and carbohydrates) can protect plants against the destabilization of membranes by the reactive oxygen species produced under salt stress.
Conclusion: Hormo-priming improves germination performance, growth and development of plants under favorable and stressful conditions. GA3 is involved in the metabolic processes mainly under the high doses of salt (150 and 200 mM NaCl), starting with the reserve degradation in seeds to the carbon skeleton and biomass production.
Keywords: *Hordeum maritimum*, *Hordeum vulgare*,hormo-priming, germination, α-amylase.

PBP17: How Vapor pressure deficit affect fruit quality and calcium content?

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Background and aim: Blossom End Rot (BER) is a physiological disorder of peppers caused by a calcium deficiency. The most common cause of blossom-end rot is fluctuating soil moisture, vapor pressure deficit and salinity. VPD plays an important role in calcium uptake in the plant.
In fact, when VPD decreases, leaf transpiration is reduced impacting xylemic Ca2 + transfer into the leaves, and increases its movement into the fruit. When a plant does not get the amount of calcium needed, malformed fruit and mushy lesions are produced.
The objectives of this study were to determine the effect of VPD in Blossom End Rot incidence and Ca^{2+} partitioning in pepper leaves and fruit.
Method: Sweet pepper seedlings (*Capsicum annuum* L. cv. Marconi) from a commercial nursery were transplanted to a glass house with two vapor pressure deficit: Control VPD (1.14 KPa) and high VPD (2.3 8 KPa).VPD was calculated based on the hourly measured relative humidity and air temperature.
Result: High VPD (2.38 KPa) increased the percentage of fruit with BER (40%) and reduced the level of calcium content in the basal and apical parts of fruit respectively for 35% and 27%.
There were no significant differences in the level of calcium in leaves and peduncle fruit Blossom End Rot is induced on fruit when Calcium content in the apical part was less than 0.03%.
Conclusion: High VPD increases the BER disease apparition and reduce calcium content essentially in apical part. This result is due to the reduction of xylemic bundles number that ensure alone the transport of calcium to the different parts of the plant.
Keywords: *Capsicum annuum L.*, Vapor Pressure Deficit, Blossom End Rot, calcium, fruit.

PBP18: Salinity and potassium deficiency induce differences in polyphenol accumulation and their antioxidant activity in *Sulla carnosa*

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Background and aim: Salinity and K^+ deficiency represent two environmental constraints that generally occur simultaneously in the field and seriously limit plant growth and productivity. Hence, the present study aimed to investigate the effects of salinity alone or in combination with K^+ deficiency on growth, secondary metabolites content, and related antioxidant capacity in *Sulla carnosa* stems and roots.
Methods: Seedlings were grown hydroponically under sufficient (6 mM) or low (60 µM) K+ supply without or with 100 mM NaCl (C: complete nutrient solution (6 mM K^+ and 0 mM NaCl), KD: K^+-deficient nutrient solution (60 µM K^+ and 0 mM NaCl), C+S: salt treatment (control nutrient solution containing 6 mM K^+ and 100 mM NaCl), and KD+S (combined treatment between salt and K^+ deficiency containing 100 mM NaCl and 60 µM K^+) to determine their effects on growth, phenolic compound contents, and their related antioxidant capacity (total antioxidant capacity, $DPPH^{\cdot}$radical scavenging ability, iron reducing power, and β-carotene bleaching test).
Results: Salinity, either alone or in combination with K^+ deficiency significantly restricted plant growth. The depressive effects are more pronounced when the two stresses are applied simultaneously. Contents of polyphenol, their composition, and their antioxidant capacity vary considerably on applied treatment with a more significant dependence on K^+ deficiency.
Conclusion: As a whole, these data suggest that K^+ deficiency aggravates the deleterious effects of salt stress. In addition, salinity, K^+ deficiency either applied alone or in combination modulates quantitative and qualitative of phenolic compounds accumulation and their anti-oxidative capacity with K^+ nutrition may be considered a suitable manipulation for the production of antioxidants compared to the other treatments.
Keywords: Antioxidants; K^+ deficiency; Polyphenols; Salinity; *Sulla carnosa*.

PBP19: The physiological and biochemical basis of zinc toxicity in safflower (*Carthamus tinctorius* L.)

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Background and aim: The toxicity of heavy metals is one of the environmental factors affecting plant growth and development. Their toxicity varies with plant species, particular metal, metal concentration and the chemical form. The aim of this work is to evaluate the physiological and biochemical responses of safflower plants in the presence of 15 and 75 jtM zinc during the flowering stage.
Methods: The effect of zinc on safflower plants was studied through the evaluation of growth parameters, colorimetric assay of phenolic compounds, protein content and antioxidant activity.
Results: The analysis of the results shows that at the flowering stage, zinc affects plant growth parameters in the presence of 15 and 75 jtM (dry weight, stem length and leaf number). While the water content shows a 2 fold increase at the leaf and root in the presence of 75 jtM of zinc.Zinc toxicity also causes chlorosis in in the presence of 75 jtM.These changes are accompanied by changes in chlorophyll pigment content, the levels of chl a, chl b and carotenoid decrease as a function of the increase in zinc concentration. Zinc also affects the levels of phenolic compounds (polyphenols and flavonoids) in the leaves, roots and florets, especially in the presence of 75 jtM. Moreover, the content of total soluble sugars is stimulated in the presence of 15 jtM, then it decreases in the presence of 75 jtM in the florets, this part also shows a progressive stimulation in the starch content in the presence of both concentration in zinc. In addition, this work shows that under the effect of zinc, the protein content and the antioxidant activity are stimulated at the leaf but they show a decrease in the florets in parallel with the increase of the zinc concentration.
Conclusion: In the present study, our results show a particular vulnerability of safflower in the presence of zinc at the flowering stage.
Keywords: antioxidant activities, chlorophylls, flavonoids, polyphenols, proteins, safflower, starch content, total soluble sugars, zinc.

PBP20: New *Pseudomonas* spp isolated from citrus orchards

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Background and aim: The main objective of this study is to characterize and clarify the taxonomy of these putative new *Pseudomonas* species
Methods: Strains were deeply analyzed morphologically; genotypically by multilocus sequence analyses of the rpoD, gyrB and 1 6S rRNA gene sequences and phenotypic characteristics were determined by API20NE and Biolog GENIII.
In a *Pseudomonas* study realized in 2017, 12 strains were isolated from different regions: 7 from Kairouan; 3 from Nabeul; and one from Ben Arous. All were identified as fluorescent Gram-negative bacteria. Phenotypic studies (API 20NE, Biolog GEN III, MALDI-TOFF) together with genotypic analyses of the housekeeping genes *rpoD*, *gyrB* and 1 6S *rRNA* confirmed their identification as *Pseudomonas* spp. The genotypic tests showed that the percentages of similatity were below the stablished threshold to assign a strain to a species of the genus *Pseudomonas*: 95% for the rpoD gene and 97% for the concatenated genes. KC strains share with *P. grimontii*a *rpoD* simmilarity of 94.2% and 95.3% with the concatenated gene analysis. Others have percentage values of 93, 8 % and 95, 9 % with rpoD and the concatenated genes respectively with *P.canadensis.* These strains are able to cause citrus black pit following their injection on lemon fruits, but they gave negative results in the hypersensibility tests on tobacco leaves, aggressiveness on melon and also in the production of syringomicin. The genomes of both representatives were sequenced and the results obtained support the conclusion that the strains represent two novel species.
Conclusion: Considering the phylogenetic, chemotaxonomic, genotypic and phenotypic characteristics presented, we propose two new species, *Pseudomonas* sp. nov.
Keywords: Cirus black pit, MLST, *Pseudomonasfluorescens* sub group, *Pseudomonas fluorescens* group, *New Pseudomonas* spp.

PBP21: Effect of soil moisture and temperature on carbon mineralization

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Background and aim: The mineralization is considered as the assimilation of the organic matter (OM) by soil's organisms in which OM is transformed into mineral compounds. Mineralization of soil organic matter is an important source of nutrients for plants. The rate of mineralization is strongly dependent on the nature of the OM, the environmental factors and the biological and physicochemical properties of the soil.
Methods: In this study, an experimental design was carried out in the laboratory under controlled conditions to give an idea of the way in which the carbon mineralization takes place and the effect of the addition of litter, the variation of humidity and the temperature on this phenomenon. In this regard, two soils have been tested. These soils come from two different sites of a pure vegetation of cork oak, one in Ain zena and the other in Khroufa. The mineralization of carbon was determined by the microbial respiration method.
Results: The results obtained suggest that adding litter of different plant species to both soil types, Khroufa and Ain Zena, clearly increases microbial activity.
Furthermore, we have found that a fairly high humidity (80% of the field capacity) favors the microbial activity of the soil and there is a significant evolution of the carbon mineralization under different types of soils. This activity decreases as the soil dries up (40% of the capacity in the field).
Similarly, a strong response of carbon mineralization rates to temperature was observed. Indeed, the quantity of mineralized carbon is greater at the temperature of 35 ° C than at the temperature of 5 ° C.
Conclusion: The results found from the experimental design shows that the addition of litter of different species have a stimulated effect on Carbon mineralization for different sites. Similarly, the moisture and the temperature have an effect on the carbon mineralization which is more pronounced respectively at the moisture content of 80% of field capacity and at 35°C.
Keywords: Mineralization, Organic matter, Carbon, Moisture, Temperature.

PBP22: Effects of the major environmental constraints on the germination of autochthonous *Vacheiia* seeds of Algeria

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Background and aim: The autochthonous *Vachellia* trees are frequent, widespread and emblematic species that grow in Algerian desert areas. *V. tortilis* (Forssk) Hayne subsp. *raddiana* (Savi) Brenan and *V. nilotica* (L.) Willd Del. subsp. *adstringens* (Schum & Thonn.) Roberty are two species that present high potentials for desert areas rehabilitation. Our aim is the study of the effects of most frequent environmental stresses encountered in these areas such as salt and water stresses on germination stage are of outmost importance for regeneration and consequently successful establishment of plantations.
Methods: For successful seed germination and establishment three pre-germination treatments (manual scarification, boiling water and concentrated sulphuric acid) were used to overcome seeds dormancy. The impacts of two abiotic stresses (salinity and water deficiency) were examined on their seeds germination using eight levels of salinity (0, 50,100, 150, 200, 250, 300 and 350mM of NaCl) and six concentrations of $_{PEG6000}$ (0, -2, -4, -6, -8 and -10 bars). Germination rate and mean germination time were calculated and data were subjected to analysis of variance (ANOVA) followed by Newman-Keuls test.
Results: The application of pre-treatments indicated that manual scarification improved the germination of *V. tortilis* (Forssk) Hayne subsp. *raddiana* (Savi) Brenan and *V. nilotica* (L.) Willd Del. subsp. *adstringens* (Schum & Thonn.) Roberty.
V. tortilis (Forssk) Hayne subsp. *raddiana* (Savi) Brenan and *V. nilotica* (L.) Willd Del. subsp. *adstringens* (Schum & Thonn.) Roberty are revealed to be tolerant to salt and water stresses and they can germinate even at high concentrations ofNaCl (350 mM) and $_{PEG6000}$ (- 10 bars).
Conclusion: The work show that *V. tortilis* (Forssk) Hayne subsp. *raddiana* (Savi) Brenan and *V. nilotica* (L.) Willd Del. subsp. *adstringens* (Schum & Thonn.) Roberty two native species present a good choice and could be suitable for land rehabilitation, restoration and afforestation programs in these areas.
Keywords: Autochtonous *Vachellia*, Algeria, seeds, germination, salt stress, water stress.

PBP23: Diversity, Antagonistic activities and PGPR traits ofActinobacteria isolated from *Artemisia herba-alba*Asso Rhizosphere

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Background and aims: To our knowledge, the study of the diversity of actinobacteria isolated from the rhizosphere of the medicinal plant *Artemisia herba-alba* Asso from arid regions of Algeria have never been reported previously. This study aims to reveal the actinobacterial diversity harboured in the rhizosphere of this plant and their morphological characterization. In addition, the screening for their*in vitro* potential as biocontrol agents against selected human and plant pathogens (bacteria and fungi) and some PGPR traits were evaluated.
Methods: The rhizospheric soil of 18 healthy plants was collected from six regions in the districts of Batna and Biskra (Algeria). Eighty purified cultures of actinobacteria were isolated from these samples using the chitin-vitamins agar medium. All the isolates were morphologically identified on ISP2 and ISP4 media. The disc diffusion method was used to evaluate their antimicrobial activity against five bacteria (a sensitive *Staphylococcus aureus*, a methicillin resistant *Staphylococcus aureus* 639C, *Bacillus subtilis* ATCC 30300, *Escherichia coli*E.52 *and Pseudomonas aeruginosa ATCC 27856*), four fungi(*Fusarium culmorum, Aspergillus ochraceus*ATCC 3174, *Aspergillus carbonarius* M333 *and Umbelopsisramanniana* NRRL 1829) and the yeast *Candida albicans M3*.The qualitative screening for various plant growth promoting traits (phosphate solubilization, siderophore, ammonia and hydrogen cyanide productions and enzymatic activities like cellulose, amylase and protease were evaluated.
Results: On the basis of the morphological identificationon ISP2 and ISP4, the isolated actinobacteria were assigned to seven genera: *Streptomyces*, *Saccharothrix*, *Pseudonocardia*, *Amycolatopsis*, *Nocardioides*, *Saccharopolyspora* and *Planomonospora* with a dominance of *Streptomyces* (84%).The results of the antimicrobial screening revealed an important antifungal activity of two *Saccharothrix* and four *Streptomyces* particularly against *Fusarium culmorum* which suggest their possible use as biocontrol agents. In addition, several other strains showed a medium activity especially against Gram – bacteria. In another hand, some of the tested isolates possessed *in vitro* Plant Growth Promoting (PGP) Traits like the strain BKS15 and BKS 39 as phosphate solubilizers and BTS3 and BKS30 as siderophore producers.
Conclusion: The result indicates that isolates of actinobacteria from *Artemisia herba-alba* rhizosphere could be interesting sources of antifungal bioactive substances as well as growth promoting substances.
Keywords: Actinobacteria, *Artemisia herba-alba*, PGPR.

PBP24: Exploring the diversity of endophytic bacteria inhabiting the roots of *Zygophyllum album*, a desertic halotolerant plant from El-Golea, Algeria

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Background and aim: In the Algerian desert (Sahara), populations of the pioneer plant *Zygophyllum album* are growing in many locations on high and low salt level soils. We hypothesize that the microbiome hosted by this plant species particularly in the endophytic compartment, play a major role in facing harsh environmental conditions. We aim to answer the following questions:

- What is the composition of the microbiome of the endophytic compartment of *Zygophyllum album*?
- Does the salt level effect the composition of the microbiome?
- Is the composition of the microbiome species dependent?

Z. album plants were collected in July 2016 from two distinct localities, one containing a high salt level and a low salt level from El-Golea. To determine whether the *Z. album* genome is a major player in shaping the microbiome we also collected *Imperata cylindrica* a pioneer *Poaceae* plant that grows at the same locations.
Methods: Total genomic DNA was extracted from roots of all samples and sequenced. Sequences with at least 97% identity were clustered and the resulting OTUs were classified using the RDP classifier and the GreenGenes database.
Results: The analysis of the microbial endophytic communities reveals the presence of 23 phyla, 220 families and 364 genera. Comparison of endophytic communities compositions at phylum level reveals that 70 % of the phyla are common to all the samples where *Proteobacteria* and *Actinobacteria* are the most abundant phyla. Comparison of endophytic communities composition at family level shows that all samples share 50% of families.
Conclusion: According to our results, both salt level and plant species are involved in determining the composition of the microbiome of the studied plants. These differences are visible in the most abundant OTUs.
Keywords: Microbiome, Pioneer plants, Salinity, Metagenomics.

PBP25: Modification of sugar homeostasis and of the anatomy of the vascular bundles in the floral stem of *Arabidopsis thaliana* under salt stress

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Background and aim: Sugar homeostasis is important for plant growth and abiotic stress responses and depends on adjusted carbon allocation between organs. The aim of this word is the study of the plant responses to salt stress with emphasis on sugar transport and homeostasis in adults plants.
Methods: The development of the main floral stem in *Arabidopsis thaliana L.* plants of the Columbia-0 *(Col-0)*, under salt stress (150 mM NaCl), with or without acclimation) was studied. Molecular, metabolic and anatomical approaches was investigated.
Results: Thirteen days after the beginning of salt stress, the content in sucrose and fructose was measured. We observed a higher sucrose and fructose content while starch content was lower in the floral stem in plants submitted to 150 mM of NaCl for 13 days. Expression analyses indicated that salt stress modulates the transcript levels of several target genes, including many genes involved in sugar homeostasis and sugar transport. Some of the responses differ significantly from those previously described in the rosette leaves at vegetative stage. We also observed that floral stem cross sections of plants subjected to salt stress exhibited lower xylem area than control plants, in addition to a significant number of collapsed xylem vessels and a modification of the composition of the secondary cell wall in xylem poles. In addition, we observed that an acclimation period of 4 days, with lower salt concentrations (50 mM and 75 mM) prior to beginning of salt stress alleviated the effect of stress on the anatomy of the vascular bundles in the main floral stem and on the composition of the secondary cell wall of xylem cells
Conclusion: These results suggest that the plant response to a salt treatment on starch and sugar homeostasis is developmentally and spatially regulated, most probably through the regulation of sugar transporters and effects on carbon allocation in competing sink organs.
Keywords: Salt stress, floral stem, sugar homeostasis, vascular bundle.

PBP26: The valorization of oak-cork acorns in the north west of Tunisia

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Background and aim: The Cork-oak forest consist an afforested areas of a great ecological, economical, and socio-economical interest. It offers a diversify production such as: Cork, timber, fodder, and acorns. In fact it presents an ecosystem over exploitative by the local population, that's way natural regenaration of this forest is very difficult.
Methods: In this study, we determined the physicochemical properties of acorns after and without conservation. Forage value was highlighted as well as its health status.
Results: In this way, an economic and social study was also carried in these sites; it showed a high activity of seed harvest and use by the forest local population, especially for animal food. This activity may partially explain the lack of natural regeneration of Cork-oak.
The study showed that acorns have an interesting chemical composition; it is rich in nutrients and energetic elements.
We noticed that after conservation, the fruits keep their excellent nutritional and germinative values; but the long conservation (+ 2 years) influences negatively the quality of the acorns. We have identified the oak balanin *Curculio elephas* (Coleoptera, Curculionidae) Synonym: *Balaninus elephas*, an insect that damages this genetic potential and negatively influences its germination and nutritional quality.
Conclusion: The production of cork oak acorns is a very important food resource for ruminants, especially during the scarcity period (autumn and winter). This study has shown that this potential has enormous properties and requires recovery for humans and their animals.
Keywords: *Quercus suber* L., acorns, physicochemical properties.

PBP27: Plant growth promoting and antagonistic activity of *Bacillus mojavensis* I4

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Background and aim: Plant growth-promoting rhizobacteria (PGPR) are advantageous bacteria that inhabit around plant roots and enhance plant growth by a wide variety of direct and indirect mechanisms. Utilization of PGPR is now gradually increasing in agriculture and offers an attractive way to replace chemical fertilizers, pesticides, and supplements. The aim of this work is to characterize *Bacillus mojavensis* I4 strain and antagonistic traits so that they can be exploited as a potential bioinoculant for plant.
Methods: The characterization of *Bacillus mojavensis* I4 strain was investigated through some enzyme activities and antagonistic activities.
Results:
Bacillus mojavensis I4 strain was able to produce the screened enzymes like amylase, cellulase, pectinase, protease, laccase and lipase. A preliminary screening of the strain using disk diffusion method for antifungal activities against six phytopathogenic fungi indicated that *Bacillus mojavensis* I4 strain displayed broad spectrum bioactivity against fungal pathogens like as *Fusarium solani*, *Fusarium oxysporium*, *Botrytis cinerea*, *Rhizoctonia solani*, *Fusarium verticillium* and *Fusarium culmorum*. *Bacillus mojavensis* I4 was also characterized for their potential of plant growth promotion according to indoleacetic acid (IAA) production, nitrogen fixation, solubilization of phosphate and production of siderophore. *Bacillus mojavensis* I4 have shown positive results for IAA, nitrogen fixation, phosphate solubilization and siderophore production.
Conclusion: From the above study it can be concluded that the strain *Bacillus mojavensis* I4 used in present study showed PGPR activities at various degree. This strain showed possibility to be exploited as biocontrol agent and may be used as bioinoculant to induce systemic resistance in plant against phytopathogens, thus proving an alternative to the hazardous chemical fertilizers.
Keywords: Endophyte, *Bacillus mojavensis* I4, phosphate solubilization, antagonistic activity.

PBP28: Genetic variability based on spike morphological characterization of Tunisian durum wheat landraces

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Background and aim: Tunisia is considered the second center of durum wheat origin. This work aims to characterize six Tunisian durum wheat populations collected from the center and the south by the National Gene bank of Tunisia (NGBT), based on ten spike morphological traits: spike shape (SS); spike color (SC); spike density (SD); awn color (AC); awn distribution (AD); glume shape (GS); glume shoulder shape (TF), glume shoulder width (LT); bec length (BL) and bec shape (BF).
Methods: 198 accessions of durum wheat landraces were used to evaluate spike morphological diversity based on ten parameters using UPOV descriptor (UPOV 2012)
Results: Analysis of the variance showed significant differences among populations based on spike shape, spike color, spike density, glume shoulder shape, bec length and its shape, a low difference with awn color, glume shape and glume shoulder width traits and no differences with the remaining traits. ACP and cluster analysis assigned the populations to 3 groups: The first group composed of two populations is characterized by spikes strongly colored, parallel sided and with an intermediate density; the second group constituted by three populations showing spikes slightly colored, pyramidal and compact and the last distant group with one population characterized by slightly colored, fusiform and lax spikes.
Conclusion: Our results highlight the useful spike traits to be considered for characterizing and evaluating the genetic diversity of the durum wheat landraces and also show the variability between the populations that could be exploited in the national and international breeding programs.
Keywords: UPOV descriptor, Diversity, Tunisia, Durum wheat, landraces

BP29: The *in vitro* antioxidant activity of *Reichardiapicroides* extracts

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Background and aim: *Reichardia picroides* (*R. picroides*) is a species mainly used for alimentary purposes; it is used in the traditional medicine as hypoglycemiant, diuretic, depurative, galactagogue and tonic agent. To our knowledge, there are no studies on the antioxidant effect of this plant. The objective of the present study was, therefore, to evaluate the *in vitro* antioxidant activity of crude extract (CrE) and its fractions.
Methods: Polyphenols extraction and fractionation from plant material was performed using solvent of different polarity giving the following phases: CrE, chloroform extract (ChE), ethyl acetate extract (EAE) and aqueous extract (AqE). The estimation of polyphenols and flavonoids was carried out using Folin-Ciocalteau method and aluminum trichloride method, respectively. The study of antioxidant activity included the DPPH scavenging and β-carotene bleaching assays.
Results: Results of polyphenols estimation showed that the highest amount of both total phenols and flavonoids was found in EAE (33 1.64 ± 3.11 µg gallic acid equivalent (GAE)/ mg of extract and 48.14 ± 0.70 µg quercetin equivalent (QE) / mg of extract, respectively).
The EAE and ChE were found to exhibit significantly ($p < 0.0001$) a strong scavenger activity (EC50 = 6.41 ± 0.51 and 28.94 ± 0.16 µg/ml, respectively) which was surprisingly similar to that of quercetin (EC50 = 2.57 ± 0.00 µg/ml). Results showed that the ChE (AA % = 80.78 ± 4.57 %) and EAE (AA % = 80.11 ± 4.24 %) were more powerful in inhibiting the percentage of β-carotene bleaching than CrE (64.30 ±13.81 %) and AqE (AA % = 54.76 ± 7.52%).
Conclusion: It can be suggested that *R. picroides* had potent antioxidant activities. Therefore, we can consider it as a natural source of antioxidant agent.
Keywords: *Reichardia picroides*, polyphenols, oxidative stress, anti-hemolytic activity, chelating activity.

PBP30: Genetic diversity of Tunisian perennial ryegrass using the chloroplast *trnH-psbA* intergenic spacer region

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Background and aim In Northern Tunisia, perennial ryegrass (*Lolium perenne* L.) constitutes an important cool-season bunch grass, widely used in pasture, to prevent soil erosion, safe guarding, lawns and hay. In order to enhance forage production and improve agronomic performance of this local germplasm, a molecular analysis was undertaken.
Methods: The *trnH–psbA* intergenic spacer region has been used to evaluate genetic diversity and relationships among 12 Tunisian accessions of *Lolium perenne* L.
Results: The chloroplast *trnH-psbA* spacers are used for study the genetic diversity and establish phylogenetic relationships. This region reveals a low level of polymorphism in the local species, *Lolium perenne L*. The low values of genetic distances argue in favor of their great genetic similarity, which is expected DNA cytoplasmic is much more conserved than genomic nuclear DNA. The hierarchical classification of the accessions studied highlights the degree of similarity between the accessions studied. However, their grouping is independent of their geographical origins. The similarities thus revealed make it possible to highlight the idea of a program to improve Tunisian ryegrass considered as a local plant genetic resource adapted to the valorization ofthe fodder courses.
Conclusion: This is the first detailed report of using the chloroplast *trnH-psbA* spacer for genetic diversity analysis of Tunisian *Lolium perenne* L.Overall, our results are able to provide useful information for the efficient utilization of *Lolium perenne* L. genome diversity and germplasm conservation.
Keywords: *trnH–psbA*, intergenic spacer region, Genetic diversity, *Lolium perenne*, DNA barcoding.

PBP31: Phenotypic and Molecular Characterization of *Verticillium dahliae,* the Causal Agent of Verticillium Wilt of Olive in Tunisia

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Background and aim: During the last two decades, verticillium wilt of olive has spread to young olive orchards where it causes severe yield losses and death of olive trees in southern and central regions of Tunisia. Therefore, identification of the causal agent as well as the study of its pathogenicity will be useful for design the appropriate management program.
Methods: This study was conducted using phenotypic and molecular methods to identify the causal agent ofthe decline and death of olive trees. The pathogen was recovered from infested tissues using a potato dextrose agar medium. Identity of the isolates was confirmed by ITSRFLP and sequencing. Pathogenicity of the isolates was evaluated by infection bioassay on young olive plants.
Results: All the fungal isolates were hyaline, flocculose and produced microsclerotia after 15 days of incubation at 25°C, which is in agreement with the identification key of *Verticillium* species. All the isolates were characterized by ITS-RFLP and sequencing of the internal transcribed spacer. All the isolates produced 470 bp using primers ITS1/ITS4. Digestion of the ITS product using *EcoRI* and *HaeIII* produced two fragments of 250 and 220 bp and three fragments of 300, 150, and 20 bp respectively. Pathogenicity was evaluated on two-year old olive plants using an artificial infection bioassay. After 15 days of inoculation, similar symptoms were produced as natural infection. Symptoms of wilt developed rapidly and caused the death of more than 50% ofthe inoculated plants.
Conclusion: *V. dahliae* is present in almost all growing olive regions of Tunisia. However, local *V. dahliae* populations in Tunisia is predominated by a highly pathogenic clone which is able to overcome the resistance of the main olive cultivar.
Keywords: Molecular identification; infection bioassay; ITS-RFLP; sequencing.

PBP32: Molecular identification and genetic variability among Tunisian strains of *Erwinia amylovora*

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Background and aim: *Erwinia amylovora* (Burill) Winslow *et al.* is the causal agent of the destructive fire blight disease that affects apple, pear and other plants in the Rosaceae family. In Tunisia, this disease first observed in 2011 has spread throughout all cultivation regions of pome and pear fruitspecies.The present study focused on the molecular characterization of a collection of *Erwinia amylovora* isolates recovered from different outbreaks in Tunisia between 2015 and 2018.
Methods: Analysis of 39 isolates, including the reference type strain CBS, revealed that all Tunisian isolates produced the expected amplicons with two primer pairs specifically used for molecular diagnostics of *E. amylovora.* We also evaluated the genetic diversity of these isolates by PCR fingerprinting, using specific primers for Internal Short Sequence Repeat.
Results: Overall, results obtained by this method, revealed that only7 primers out of 14 used, which produced an identical amplicon for all isolates. The remaining primers showed moderate variability among Tunisian isolates. Phylogenetic analysis following the UPGMA method revealed that all strains were clustered regardless of their geographical origin and their pathogenicity features.
Conclusion: These results suggest that local strains evolved from common ancestor, which could be introduced in Tunisia by infected plant material. Additionally, these results indicate that PCR fingerprinting using ISSR markers is a goodand reliable tool for discrimination of *E. amylovora* strains and for their identification in epidemiological studies.
Keywords: *Erwinia amylovora*, feu bactérien, PCR fingerprintings.

PBP33: Comparative flower biometry and correlation analysis with pollen viability in *Vicia narbonensis* L. accessions

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Background and aim: Since in annual legumes, autogamy predominated over allogamy and the mode of reproduction has a great influence on the genetic diversity of the species and their distribution, this work was carried out within the framework of the program for the conservation and valorization of genetic resources of legumes forage and pastoral local and introduced from INRAT. It aims to study the floral biology of five accessions of *Vicia narbonensis* L. of which four are introduced (VN1, VN7, VN10 VN14) and one is local (545) used as a check.
Methods: In order to characterize the accessions for the morphology of their flower, measurement of the floral pieces were carried out on five flowers by accession and plant and concerned the following parameters: length and width of the standard, length of the hull and wing, length of the ovary and width of the calyx. The pollen viability was also analyzed for the 5 accessions using the Alexander solution (1969).
Results: The one way analysis of the variance applied on the measured parameters showed that only the width of the standard differs significantly between accessions (Pr<0.05): it varies from 0.4 to 1.4 cm for respectively VN545 (from Tunisia) and VN10 (from Turkey). Analysis of variance showed that the viable pollen is lower for VN545 (34.4%) and higher for VN14 (40.4%). A strong and positive correlation was found between pollen viability and the length of the standard showing that more the standard of the flower is long more the pollen is viable (r= 0.9; n=5; P<0.04) and between the length of the hull and the length of the wing (0.96, n=5; P<0.01).
Conclusion: the data obtained on floral biometry of Narbon vetch accessions may have impact on the breeding system of this species and the viability of the pollen. These results may contribute to the conservation and future genetic improvement of Narbon vetch.
Keywords: *Vicia narbonensis* L., floral biometry, pollen viability, local population, accessions.

PBP34: Dark Septate Endophyte(DSE) more efficient than Vesicular Arbuscular Mycorrhiza (VAM) in facing drought? Case of *Pistacia atlantica* in arid land (Algeria)

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Background and aim: The Atlas Pistachio is among the few spontaneous tree species in the arid environments of North Africa. The flexibility of its root system and the efficiency of its strategy of occupying these indigent environments allow it great ecological amplitude. However, facilitating relationships with other organisms could also explain its adaptive performance. Thus, we have made an inventory of the main potential root fungal symbionts (VAM and DSE) in four Atlas pistachio populations located along a gradient of increasing climatic and edaphic aridity according to a north-south transect in Algeria.
Methods: We sampled the roots of four spontaneous populations of Atlas Pistachio, located on a gradient of increasing climatic and edaphic aridity (north-south transect). The fine roots are bleached, stained with trypan blue and observed with optical microscopy. Mycorrhizal fungi are identified according to the protocol of Brundrett et *al*., (1996) and Blaszkowski (2012). DSE identified by morphological characteristics (mainly: septate hyphae and microsclerotia).
Results: It appeared that the aridity increasing, the generic diversity of the VAM decreases, while the roots show more DSE.
Conclusion: As described by the authors, DSE could protect the roots against pathogens and other potential herbivores, improve their thermo-resistance more than VAM do. DSE can improve host plants efficiency in the acquisition of nutritive resources, especially of organic origin.
Keywords: VAM, DSE, drought adaptation, *Pistacia atlantica*.

PBP35: Metabolic mapping of *Agave americana* leave juice, Biochemical characterization of an α-amylase activity (AmyAa) and their prospective use in pulp industry

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Background and aim: The phytochemical and enzymological characterization of *Agava Americana* leaves juice have a great interest in various medicinal, food and cosmetic applications.
About biochemical characterization of the α-amylase activity, it is realized mainly to improve the quality of some national agro food industries.
Methods: The exploitation of *A. americana* has been studied through phytochemical, physiological and enzymatic characterization.
Results: We have exploited the chemical, enzymatic and antioxidant abilities of*Agave americana*. Indeed, we monitored the presence of various metabolites. The amount of proteins was estimated to 8.13 mg/g of dried leaves. In addition, *A. americana* leave juice contains lipase activity (280 $U.ml^{-1}$), tyrosinase one (27 $U.mL^{-1}$), peroxidase activity (20 $U.mL^{-1}$), protease activity (356 $U.mL^{-1}$), and also α-amylase activity (264 $U.ml^{-1}$). Given that amylases are important industrial enzymes that have been applied especially in pulp industries, the *A. americana* one was purified after two purification steps. Their optimal activity is showed at 70 °C and pH 9. Also, other physico-chemical properties are monitored. In another hand, the *A. Americana* scavenging system proves their antioxidative abilities. Eventually, those findings enhance their potential use in pulp industry in which α-amylase upgrades some parameter's like elasticity index, baking strength, humidity and fall time.
Conclusion: Eventually, phytochemical, enzymological, physiological and antioxidative characterization of *A. americana* leave juice is of great interest. Those findings seem constitute metabolic mapping of our biological matrix and enhance their plausible use in various medicinal, para-pharmaceutical, food and cosmetic applications. For example, many compounds are now under serious investigations related to one of the repeated diseases (Sahnoun et al. 2017; Sahnoun et al. 2018). Antioxidative potentialities are monitored to seek for their plausible use as antioxidant effectors. In another hand, purification and biochemical characterization of the AmyAa is realized in order to look for the improvement of quality of some of national agro food industries. The plausibility and encouraging use of the *A. americana* juice in pulp industry give birth for mort investigation and very efficient exploration of this biological matrix for biotechnological purposes. Also, their richness on vitamin C could justify their use in this field.
Keywords: *Agave americana*, phytochemical mapping, enzymological capacities, biotechnological tools, α-amylase activity, pulp industry.

PBP36: Phytochemical study and evaluation of the anti-haemolytic activity of leaf extracts of *Ruta graveolens* L. (Fidjel) from Tlemcen region (Algeria)

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Background and aim: *Ruta graveolens* or commonly called Fidjel, is a plant belonging to the family of Rutaceae, growing spontaneously, widespread in North Africa, particularly in Algeria, it is used in traditional medicine as anti-inflammatory, antioxidants and for the treatment ofcoetaneous pathologies.
Materials/Methods: The aim of this work is to carry out a phytochemical study based on qualitative and quantitative assays of phenolic compounds of different extracts prepared by maceration: hydromethanolic crude extract, ethyl acetate extract and n-butanol extract of *Ruta graveolens* leaves harvested in the region of Tlemcen (Algeria). A biological study based by evaluation of the anti-haemolytic activity, carried out by contacting the human red blood cells with extracts from the leaves of Ruta graveolens at different concentrations tested in the presence of AAPH (2,2'- azobis (2-aminodipropane, dihydrochloride).
Results: Phytochemical analysis showed the presence of alkaloids, flavonoids, tannins, free quinones, terpenoids and saponins.
Dosage of total polyphenols and flavonoids carried out according to the method of Folin Ciocalteu and aluminum trichloride, respectively, revealed that the ethyl acetate fraction is the richest in these phenolic compounds with total polyphenol contents order of 997.23 µg Eq AG/ mg ES and flavonoids of order 221.80 µg Eq C / mg ES.
The assay results of in vitro anti-haemolytic tests indicated that extracts of Ruta graveolens leaves have a protective effect on the erythrocyte membrane against the oxidative stress induced by AAPH, in a dose-dependent manner. The ethyl acetate extract represents the protective power the more potent with HT50 of order of 184.3, 202.1, 210.98 and 197.85 min for concentrations 10, 20, 40 and 80 µg / ml, respectively.
Conclusion: The ethyl acetate extract of Ruta graveolens leaves showed the highest levels of phenolic compounds and showed a very potent protective capacity against hemolysis of erythrocytes.
Keywords: *Ruta graveolens L.*, phytochemical analysis, polyphenols, anti-haemolytic activity, AAPH

PBP37: An efficient diazotrophic Halotolerant PGPB strain with plant growth stimulating activity: *Bacillus* sp.BCLRB2

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Background and aim: Olive tree and gramineaous plants were a two unique species that occurs naturally in Tunisia, has a high socioeconomic and nutritional value. Many farmers who cultivated wheat between olive tree rows observed an increased effect of promoting wheat growth and decreased incidence ofphytopathogenic infections.
The salinity is the major factor limiting productivity and vulnerability to several microbial plant attacks. Novel agricultural technologies are required to improve food production in saline and dry soils. Despite their different ecological niches, diazotrophic free-living rhizobacteria and diazotrophic endophytic bacteria use some of the same mechanisms to promote plant growth and control phytopathogens.
Methods: This work is designed to screen diazotrophic endophytic PGPB associated with olive tree for improving capability stress tolerance. For this, 4 strains were selected following a biochemical characterization of plant growth promotion activities such as ability of antimicrobial production, azote fixation, ACC deaminase production, growth hormone production, P solubilization... Furthermore, the efficient BCLRB2 strain was explored for in vivo pot plant growth and Biocontrol activity against wheat fungal wilt experiment, and finally the antimicrobial activity was partially biochemical characterized.
Results: Inoculated plants with bacterial strain, namely *Bacillus sp. BCLRB2*, showed the best growth of durum wheat seedlings compared to a control under salt stress and natural conditions. Total length, fresh weight, and total dry weight were significantly higher in inoculated plants compared to uninoculated plants. The BCLRB2 significantly suppressed wheat fungal wilt by 5 6.5% in the greenhouse experiment, whereas the uninoculated plant leaves were desiccated and slackened and the disease incidence was 89.9%. With TLC analysis, three bioactive spots were observed (Rf values of 0.10, 0.45 and 0.77) and one spot inactive with Rf 0.3 7. After checking out his powerful antagonistic effect against Fusarium sp., the analysis of the different fractions of the crude bacterial extract by HPLC reveals that this extraction by fractionation can give us a clear idea about the nature of the antifungal active substance.
Conclusion: It could be predicted that Bacillus sp.BCLRB2 improve plant growth under saline conditions and suppress fungal disease wilt ofwheat.
Keywords: PGPB, Salt stress, durum wheat.

PBP38: Bio-guided fractionation and biological properties of bioactive compounds of *Retama raetam* methanolic extract

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Background: *Retama raetam* is an aromatic and medicinal halophyte known for its hypoglycaemic activity and its role as an antidote against snake bites.
Materials/Methods: Fractionation by the open chromatography on silica gel of the methanolic extract of Retama raetam yielded 25 fractions which were analyzed by thin layer chromatography and grouped into four sub-fractions. These sub-fractions, in addition to the crude extract, were evaluated for their content on phenolic compounds (total polyphenols, flavonoids and condensed tannins) and for their antioxidant, antibacterial, antidiabetic and anti-cancer properties.
Results: The main results have discriminated fraction 3 as being the most active since it has the highest levels of phenolic compounds. Moreover, this fraction was characterized by an antioxidant activity as important in vitro as ex vivo, surpassing even that of the crude methanolic extract. Indeed, it exhibited an ORAC value equal to 8.1 µmol Trolox.mg^{-1} and anti-radical and ex-vivo antioxidant activities equal to 22 and 4.5 µg.ml^{-1}, respectively. However, the fractionation of the crude methanolic extract has considerably reduced the anticancer potential. In fact, all sub-fractions showed a low anti-cancer capacity, except from fraction 3 which exhibited a strong activity against A-549 cells with an $_{IC50}$ equal to 34 µg.ml^{-1}. Concerning the antidiabetic and antimicrobial activities, fractions 1 and 4 were the most active, respectively, and were even better than the crude methanolic extract.
Conclusion: Finally, these results emphasize the importance ofbioguided fractionation in the improvement of the biological activities of plant extracts as they highlight the richness of this species in active biomolecules, exploitable in pharmaceutics or food industries.
Keywords: *Retama raetam*; Open column chromatography; phenolic compounds; biological activities.

PBP39: Natural variation of drought response in *Brachypodium hybridum*

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Background and aim: *Brachypodium hybridum* belongs to the *Poaceae* grass subfamily. It has a close genetic relationship with temperate cereal crops, which means that it can be used as a model for temperate cereal and grass crops. The aim of this work is to evaluate the morphological,physiological and biochemical responses to water deficit in natural genotypes of *Brachypodium hybridium*
Methods: 145 lines of *B. hybridum* representatives of nine populations and all the eco-regions of Tunisia were used to evaluate the drought tolerance. Drought stress was imposed by withholding water for eleven days and the control plants were received regular irrigation during the treatment. The drought response index (DRI) of nineteen agronomical and physiological traits were used to evaluate the drought response of the 145 genotypes. Then, the tolerant and susceptible genotypes were evaluated by the malondialdehyde (MDA) content and the level of hydrogen peroxide H2O2.
Results: Results show a considerable variation between genotypes. Discriminant analysis revealed that four of the nineteen traits explained most of the phenotypic variation, , which was used to classify the tolerant and susceptible accessions. Four groups of accessions differing in drought tolerance were identified. A large variability is recorded in the content of malondialdehyde MDA and the level of hydrogen peroxide H2O2, the tolerant group had little leaf wilting and fewer increase in MDA and H2O2 in comparison to susceptible one.
Conclusion: The method used in this study allows the detection of material that has the ability to grow at drought conditions, which prevent the growth of other material. Thus, the large variation of *Brachypodium* in response to drought stress can be used to identify genes and alleles important for the complex trait of drought tolerance
Keywords: *Brachypodium hybridium*, drought stress, DRI.

PBP40: Diversity of rhizobacteria associated with the rhizosphere of ficus carica

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Background and aim: Nowadays, the excessive use of agrochemical products have many negative impact on environment Plant Growth Promoting Rhizobacteria (PGPR) can be used as bioferetilizers for a natural solution . The aim of this work is to identify bacterial communities associated with *ficus carica* rhizosphere soil and to study also their action mechanisms for further studies.
Methods: The diversity of these rhizobacteria was inspected through PCR amplification of 16S and Identification of isolates and PCR-DGGE analysis of bacterial communities also Characterization of PGP traits and abiotic stress resistance of these isolates.
Results: the PCR amplification of 16S and the identification of Isolates revealed a wide diversity into these bacterial communities associated with *ficus carica* rhizosphere we found several bacterial genera such *as Lysobacter , Bacillus , Arthrobacter , Pseudoxanthomonas. Pseudomonas* was the genre most frequently observed in the collection these cultivable bacteria showed a predominance of gram-negative bacteria favored than gram positive bacteria. These rhizobacteria were characterized for a series of PGP traits *in vitro*. The majority of isolates showed multiple PGP activities most common PGP trait was siderophore production followed by cellulase followed by protease activity and then lipases activity in the last position the solubilization of phosphates. We performed also further analyses to evaluate the adaptability of isolates to abiotic stresses (different high temperature , different range of PH and salinity also drought resistance) then we selected 29 strains that showed most of PGP traits tested we found in this group many genus such as *Pseudomonas , Bacillus , Arthorobacter, Flavobacterium.*
Conclusion: the bacterial communities associated with the rhizosphere of *ficus carica* showed a high diversity with a several genera of bacteria, many isolates showed many PGP activities can be selected for further studies .
Keywords: Rhizosphere; Bacterial communities, Diversity, PGPR activities, Abiotic stress.

PBP41: Specific micobial effects on C: N: P stoichiometry of four *Sullacarnosa* ecotypes under saline and non-saline conditions

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Background and aim: Sulla(*Sulla carnosa*) is a salt-tolerant forage legume that could be utilized for bioremediation of salt-affected soils.The main goal of this study was to assess the influence of plant-growth-promoting bacteria (*Bacillus* sp.) or/with the arbuscular mycorrhizal fungus *Rhizophagusintraradices* on plant growth and C: N: P stoichiometry and the interaction between them of four *Sulla carnosa*ecotypes (*SidiKhlif, Thelja, Kalbia* and *Kerker*) under saline and non-saline conditions.
Methods: Pot experiments were conducted in a greenhouse and *S. carnosa*seedlings were stressed with NaCl (200 mM) for 4 weeks. Plant biomass and mineral nutrition were assessed.
Results: Results showed that microbial effectiveness on plant growth ranged from insignificant in *Kerker* to highly positive in the ecotypes *SidiKhlif* and *Kalbia*. The growth of sulla plants did significantly decline under application of salt stress compared to the fresh water-irrigated controls, while the decline was much more pronounced in *Sidikhlif* ecotype.
Particularly in *Kalbia*, the shoot N and P concentrations were enhanced in inoculated plants with *R. intraradices*under stressed conditions compared to non-inoculated ones. Thus suggests that inoculation with AMF during nursery establishment contributes to alleviatesalt stress by maintaining a favorable nutrient profile in *Kalbia*ecotype. We found that shoot of plants inoculated with *Bacillus sp.*was demonstrated distinct C: N: P stoichiometry as compared to non-inoculated plants in *Sidikhlif.* There by indicating that *Bacillus* sp. could make the plants more tolerant to salt stress through nutrient allocation.
Conclusion: Present data suggesting that microbial inoculationchanged C: N: P stoichiometry and the change in C:N: P stoichiometry was associated with plant growth and salt tolerance.
Keywords: *Bacillus* sp.**,** *Rhizophagus intraradices***,** *Sullacarnosa* ecotypes**,** Nutrition C: N: P stoichiometry**,** Salinity.

PBP42: Biochemical and physicochemical analysis of a traditional olive oil and determination of its effects on lipid parameters

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Background and aim: In Algeria the extraction of olive oils, from the Oleaster or cultivated olive is done in a traditional way without considering the specific characteristics of each oil. The Oleaster gives particular oil used in pharmacology. This oil is very rare in Algeria despite the wealth of forest resources in Oleaster. In order to value the products of our area, the objective of this study is to evaluate: The physicochemical characteristics and fatty acid composition of the Oleaster oil extracted by traditional manual method, used in study area; the effects on the blood lipid profile of *Wistar* rats turned hyperlipemiants by gavage with vegetable and animal fats.
Methods: The oil was extracted by the traditional method after this extraction we tried to determinate Fatty Acids by GC**,** Physicochemical indexes were determined according to AFNOR and European standards finally sixty four female rats were used from Nutritional and Pharmacological Study of Oil.
Results: The physicochemical characteristics: acidity (0, 73%), peroxide value (14, 16 meqO2/kg oil) and iodine value (74, 08 g iodine / 100 g of oil) are consistent with international standards. The dosage of FA revealed a wealth of oil with UFA (76, 7%), mainly composed of 65.43% of MUFA whose major fatty acid is oleic acid (63, 57%). The experiment on rats receiving a diet rich in saturated fats and hydrogenated oils revealed that the consumption of Oleaster oil at the dose of 10g and 20g for 15 and 30 days improves plasma lipid profile by decreasing the rates of TC, TG, TL and LDL-C with an increase in the rate of HDL-C serum.
Conclusion: The physicochemical analysis of the oil shows that the acidity and peroxide values are in the standards (IOC, 2011, EEC, 2005), the fatty acid composition by GC revealed that the Oleaster oil is oleic type, The importance of treatment effects of oleaster oil on *Wistar* rats depends on the dose and period of treatment.
Keywords: Oleaster oil, fatty acid, *olea europea*, oleic acid, lipid profile.

PBP43: Morphological characteristics assessment and yield components in some durum wheat varieties (*Triticum durum* Desf.) under drought stress

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Background and aim: The present study was carried out at the agricultural experimental station of the field crop technical institute (ITGC) of Sétif during the 2016/2017 agricultural campaign.
Methods: The tests were carried out according to a device in randomized blocks with 4 repetitions, the elementary parcel is 6 rows x 5m long x 0, 20m inter-row spacing. Our objective was to study the morphological characters of tolerance to drought stress and the component evaluation of the yield of some varieties of durum wheat (*Triticum durum* Desf.).
Results: We have estimated the following parameters: Above-ground biomass (BIO, g/m²), seed yield (RDT, g/m²), height (Cm), weight of one thousand grains (WTG g), the number of grains/m², The number of ears/m², weight of ears/m², the weight of an ear (g) and the number of grains/1 epi. The results show that the (V1) variety has the best average values for above-ground biomass (BIO, g/m²), yield (RDT, g/m²), and height (Cm) compared to the (V2) variety and the control too. Variety (V1) and the control are distinguished by high average values of the number of ears/m² and weight of ears/m² compared to the (V2) variety. While the results of Weight of 1000 grains the highest is noted in the (V2) variety with slight deference between (V1) variety and the control. However, the weight of an ear is almost the same in all varieties. The variety (V2) has the lowest seed/ear count and the number of seeds/m².
Conclusion: The study carried out showed a very pronounced unequal effect of drought stress on the morphological behavior of different varieties.
Keywords: Durum wheat, variety, drought stress, tolerance, components yield.

PBP44: Study of the hydrogen potential correction effect of water on the behavior of the bean *Phaseolusvulgaris L.*

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Background and aim: The Correction of the hydrogen potential natural saline water improves considerably the use of water resources in arid areas. Indeed, four saline treatments were applied which is a water natural GASSI TUOIL saline and three fixed saline treatments by adding acids and micronutrients that make them balanced salt what facilitates their absorption by the plants. Indeed, natural saline water correction greatly improves the use of water resources in arid areas, thus the pH correction resulted in a significant increase on the studied eco-physiological parameters.
Methods: The experimental device is factorial two-factor total randomization. The first factor represents the four saline treatments, and the 2nd factor is represented by the studied variety. The repeat count is five pots by treatment.
Results: Enough salt concentrations prevent plants to absorb water and nutrients and therefore slow plant growth. It also notes that saline stress led to water stress. This lack of availability of trained water an ionic imbalance, disturbances in ion homeostasis and causes cell toxicity. Since the salt stress involves both ionic and osmotic stress, stopping the growth is directly related to the total concentration of soluble salts from the ground of the similar observations. Also, this slowdown in growth the vacuole allows a massive storage of Na^+, without damage to the functioning of the rest of the cell. When the ability of accumulation of the vacuole is full, the ions of Na^+ who continue to achieve aerial parts accumulate either in the cytoplasm, the cell walls. The saturation of the walls by an excess of ions causes a fluid imbalance resulting in a brutal water loss of cells that will dehydrate and die.
Conclusion: The study of the various parameters measured in the final stage of culture, allowed us that natural saline water use greatly limited growth and development. This is due mainly to high salinity and Ionic disorder in natural environments, as well as the lack of the useful nutrients for their growth including nitrogen, phosphorus and potassium, which has led to abortion rates up to 100%.
Keywords: Saltwater, potential hydrogen, Bean, Above ground.

PBP45: Effect of variations in nitrogen and potassium requirements on tomato (solanum lycopersicum) cultivated above ground

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Background and aim: This work aims to study the effects of variations in nitrogen and potassium requirements of a basic nutrient solution on the growth, production and quality of fruit from a variety of tomatoes (Saint-pierre) cultivated above ground.
Methods: Four (10%, 20%, 30%, and 40%) and three (10%, 20%, and 30%) decreases were made in the amount of nitrogen and potassium in the initial nutrient solution.
Results: The main results obtained show that the increasing nitrogen and potassium doses positively improve the studied parameters such as the number of leaves, fresh and dry weight of the aerial part of the plants. The best averages were obtained with T4 treatment. With respect to production parameters, it was found that increases in potassium and nitrogen in nutrient solutions significantly and positively influenced the total weight of fruit per plant. On the other hand, the treatments which were the subject of potassium and nitrogen restrictions, ie T5, T6 and T7, adversely affected the aforementioned parameter. Also it has been shown that the total sugar level as well as the vitamin C content were improved regardless of the increase achieved and that the titratable acidity of the fruit juice was decreased with the same treatments. In general, the nitrogen and potassium reductions achieved have an adverse effect on all the parameters measured.
Conclusion: the results obtained in this work show that the potassium and nitrogen increases achieved in nutrient solutions significantly and significantly influenced the parameters studied, this is related to the fundamental role of these two elements in physiology.
Keywords: Tomato - above ground crop - nutrient solution - nitrogen - potassium - mineral nutrition.

PBP46: Restoration of durum wheat growth under salt stress by Inoculation with the rhizobacteria *Pseudomonas fluorescens* RB 13

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Background and aim: Soil salinity is one of the major factors affecting agriculture. It causes a reduction in growth, yield and even death of plants. To grow in saline environments, bacteria and plants have to adjust their turgor pressure by accumulating compatible solutes as glycine betaine and proline. Inoculation of plants of economic interest, mainly wheat, by Plant Growth Promoting Rhizobacteria (PGPR) such as Pseudomonas species is an effective biological approach for the recovery of soils affected by salt. In addition, these bacterial species may be a good alternative to pesticides and fertilizers. They produce bioactive molecules stimulating plant growth [indole acetic acid (IAA), sidérophoeres, ammonia] and they solubilize phosphates. They protect plant by suppression of deleterious organisms through the HCN production or antibiotics synthesis. In order to restore plant growth on surfaces affected by salinity, the strain *Pseudomonas fluorescens* RB13 was applied as inoculant of durum wheat under saline stress conditions
Methods: *Pseudomonas fluorescens* RB13 isolated from the wheat rhizosphere was characterized by its ability to promote plant growth. The Wheat seeds were inoculated with the strain under different levels of salt. Germination rate and plant biomass were calculated.
Results: *P. fluorescens* RB13 solubilized 187.9 µg/ml of phosphate and produced 50,95 µg/ml of IAA. The seed inoculation of durum wheat with *P. fluorescens* RB13 increased the cumulative rate of germination and plant biomass. *In vitro*, the morphological and biochemical characteristics were significantly improved with the inoculated wheat under an osmotic stress of 200 mM/ NaCl.
Conclusion: These results are in favour of the inoculation of wheat seeds by active and halotolerant strains of *P. fluorescens*. This would be very beneficial for sustainable agriculture in the soils affected by salinity.
Keywords: Inoculation, *P. fluorescens*, PGPR, durum wheat, Salinity.

PBP47: Phylogeny and genetic diversity of *Erwinia amylovora* strains in Tunisia based on multilocus sequences analysis (MLSA)

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Background and aim: Fire blight disease caused by bacterium *Erwinia amylovora*is among the most serious threat to rosaceous, was introduced for the first time in Tunisia in 2012. Since, the disease propagated then in wide causing immense damages. Phylogeny and assessment of genetic diversity of strains is important for epidemiological studies of *E. amylovora* and to trying to improve our understanding of the mechanisms of its long-distance spread. In this context, the aim of this work is to study phylogeny and genetic diversity of Tunisian strains using multilocus sequences analysis (MLSA).
Methods: An MLSA scheme comprising the three house-keeping genes atpD, infB, and rpoB encoding, respectively, ATP synthase subunit B, translation initiation factor IF-2, and RNA polymerase beta subunit were used in this study.
Results: PCR amplification and DNA sequencing on both strands led to the generation of confirmed consensus sequences comprising 525bp, 726bp, and 489bp in length for the atpD, infB, and rpoB marker, respectively. For these markers, orthologous Erwinia sequences identified in the GenBank database ranged in similarity from 99% to 86% (atpD), from 100% to 87% (infB), and from 100% to 89% (rpoB). Similarities among amplified consensus sequences from Tunisian isolates as calculated in a p-distance matrix ranged from 100% to 99.6% for both the atpD and rpoB marker, whereas amplified infB gene sequences from all isolates were found identical.
Phylogenetic reconstruction based on the three markers independently located all Tunisian isolates in one clade together with the available reference sequences representing the species *E. amylovora* more exactly.
Conclusion: The results obtained demonstrates that all bacterial isolates investigated belong to the taxonomic species *Erwinia amylovora*. The low degree of variation of marker genes across the Tunisian isolates is for each single marker consistent with the average level of sequence dissimilarity across the *E. amylovora* reference sequences stemming from diverse geographic origins. Therefore, as judged from MLSA data, *E. amylovora* present in Tunisia do not form a particularly homogeneous population as might be expected for a recently emerged invasive organism.
Keywords: Fire blight; genetic diversity, MLSA.

PBP48: The major role of the double association Mycorrhizae-Rhizobium in adaptation to climate change requires the development of leguminous crops

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Background and aim: Soils and legumes provide a unique symbiosis that protects the environment, improves productivity, supports efforts to adapt to climate change, and provides essential nutrients to soils and other crops. In this context, it is urgent to develop and encourage the nitrogen fixing crop. The objective of this work is on the one hand the study of the germination, the growth and the cycle of development of Medicago sativa, and on the other hand; to compare the morphological and physiological mechanisms for two years with a different water supply and illustrates the role of mycorrhizal and rizhobium symbiosis in adaptation.
Methods: Our experiment concerns the cultivation of Medicago sativa in the field, soil analysis, germination and root coloring using the Phillips and Hayman method.
Results: The results we recorded showed that Medicago sativa develops a great resistance to water reduction (2nd year).
The soil has a sandy loam texture, a pH of 8.4, low organic matter. Identification of the type of mycorrhization (AM). During the water stress we noted that the slight decrease in the dry weight of the aerial part is due to a decrease in the number of leaves and branches of the stems. Moreover, the decrease in the dry weight of the roots is attributed to a depletion of the root reserves.
We noticed that the weekly growth in stem length in this year (124.75 cm) is significantly greater than that of the first year (65.7 cm).The proportion of leaves is greater in the 2nd year compared to the 1st year (407 leaves for the 2nd year, 192 leaves the 1st year). The leaf area is about 10.7 times that of the first year. It increases when the plant ages.
Conclusion: Through our study, we wanted to demonstrate that the development and intensification of Medicago sativa cultivation is an ecologically and economically interesting alternative to water stress. The results we recorded showed that Medicago sativa develops a great resistance to water reduction (2nd year) underlining the role of double mycorrhiza - rhizobium association.
Keywords: Medicago sativa, Mycorrhiza-Rhizobium, Physiology, Growth, Water deficit.

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