



Contents lists available at ScienceDirect

LWT

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Fortification of durum wheat fresh pasta with red chicory by-product powder: Effects on technological, nutritional, and sensory properties

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ARTICLE INFO

Keywords:

Red chicory by-product
Fortified pasta
Technical properties
Biocompounds
Sensory analysis

ABSTRACT

The fortification of staple foods and the exploitation of agri-food by-products are the goals of modern food technologies. In this study, we prepared fortified pasta by replacing semolina flour with 5%, 10%, and 15% dried red chicory by-product powder (RCP), rich in fiber (27%) and healthy bioactive compounds (4.3%). The UHPLC-DAD-HRMS indicated hydroxycinnamic acids (HA), flavonoids (F), anthocyanins (A), and sesquiterpene lactones (SL) as the main bioactive compounds of RCP. The extraction recoveries of phenolic-fortified pasta (ranging from 0.2 to 13% for HAs to 5–28% for Fs and < 0.2% for As) suggest a high affinity with the semolina components, likely gluten or starch. The addition of RCP influenced ($p < 0.05$) the pasta's technological properties by inducing an increase in cooking loss, fully cooked time, and a decrease in the swelling index. The texture analysis showed that firmness and adhesiveness increased in the fortified sample. The sensory characterization showed a greater perception of vegetable and bitter flavor with increasing fortification levels. The RC15 sample had optimal technological, nutritional, and sensory characteristics. In conclusion, RCP could be an ingredient used to produce pasta with high fiber and bioactive compounds.

1. Introduction

Chicory (*Cichorium intybus* L.) is a perennial herbaceous plant of the Asteraceae family. The commercial interest of this vegetable is considerable, and its cultivation is widely distributed in Asia, North America, and Europe. In a north-eastern Italian region (Veneto) are present the most extensive red chicory (locally called “radicchio”) plantations; currently, the main varieties of *radicchio* cultivated in Veneto are “red chicory from Chioggia” (*Radichio di Chioggia*), “early red chicory” (*Radichio Rosso di Treviso*), “late red chicory” (*Radichio Rosso tardivo di Treviso*), “Marbled chicory” (*Variegato di Castelfranco*) and “Verona red chicory” (*Radichio Rosso di Verona*) (Barcaccia et al., 2003). In the Veneto region, in 2022, the cultivated surface in the open air was 4.602 ha, and the total crop production was 759.465 quintals (ISTAT, 2022).

Besides its economic and gastronomy features, red chicory is noteworthy for other reasons. Firstly, it belongs to traditional medicine since all parts of this plant contain potentially healthy bioactive compounds (Satsbekova et al., 2018). Then, red chicory represents an excellent example of functional food with health benefits. It has a prebiotic function in animal models (de Godoy et al., 2015) on bifidobacterial and lactobacilli due to its content in fermentable fiber such as inulin. Inulin is a plant-storage carbohydrate that ranges from 11 to 45 g/100 g in leaves and roots (Nwafor, Shale, & Achilonu, 2017).

From a human health point of view, dietary fiber plays an essential role in human health (Simonato, Trevisan, Tolve, Favati, & Pasini, 2019). Numerous studies have highlighted its role in promoting digestive functions, preventing gastrointestinal disorders, reducing the cholesterol levels in the blood, lowering the risk of type 2 diabetes, and

Abbreviations: RCP, red chicory by-product powder; CP, control pasta; RC5, 5 g/100 g red chicory powder fortification; RC10, 10 g/100 g red chicory powder fortification; RC15, 15 g/100 g red chicory powder fortification; DM, dry matter; a_w , water activity; FCT, fully cooked time; CL, cooking loss; SI, swelling index; FRAP, ferric reducing antioxidant power; ABTS, 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic); TE, Trolox equivalents; SDF, soluble dietary fiber; ISD, insoluble dietary fiber; TDF, totally dietary fiber; UHPLC-HRMS/MS, ultra-high performance liquid chromatography–high-resolution tandem mass spectrometry.

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<https://doi.org/10.1016/j.lwt.2024.116358>

Received 20 March 2024; Received in revised form 13 June 2024; Accepted 14 June 2024

Available online 26 June 2024

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preventing obesity (Anderson et al., 2009; Da Rocha et al., 2021; Simonato et al., 2019). The red chicory is also rich in antioxidant molecules (Innocenti et al., 2005), such as flavonoids, anthocyanins, and caffeic acid derivatives, with the content of total phenolic compounds in the range of 1.4–78 g/kg, according to the variety (Ferioli, Manco, & D'Antuono, 2015). These polyphenols are bioactive compounds that act against pathogens of bacterial and viral origins and, in addition, protect from degenerative and age-related pathologies (Mainente, Menin, Alberton, Zoccatelli, & Rizzi, 2019). Antioxidant, anti-inflammatory, hypolipidemic, gastro-protective, and anti-diabetic are some bioactivities related to this plant (Perović et al., 2021). In addition, sesquiterpene lactones, the compounds responsible for the bitter taste of *Cichorium* species, may play a highly significant role in human health, both as part of a balanced diet and as nutraceuticals, due to their potential for the treatment of cardiovascular disease and cancer (Chadwick, Trewin, Gawthrop, & Wagstaff, 2013). For all these reasons, the production and consumption of this plant should be encouraged, as proposed by the Chicory Innovation Consortium (CHIC PROJECT).

Chicory crops produce many by-products that might be valorized from the circular economy point of view. Indeed, residues (leaves, stems, and others) constitute 40–50% of the total harvested material (Lante, Nardi, Zocca, Giacomini, & Corich, 2011). Like the edible parts of the plant, these by-products are rich in healthy bioactive substances and represent a promising ingredient for functional food formulations.

Pasta is a staple wheat-based food that is pivotal in the Mediterranean diet. This food has a high energy value and is rich in complex carbohydrates but insufficient in some vitamins, minerals, and dietary fibers (Da Rocha et al., 2021; Tolve, Pasini, Vignale, Favati, & Simonato, 2020). For these reasons, several studies focused on fortifying pasta with vegetal by-products rich in fiber, such as olive, grape, vegetable, apple, and okara powder, which elevates pasta's total polyphenol content and antioxidant capacity but is accompanied by a modification in technological properties (Bianchi et al., 2021).

This study aimed to assess the effects of red chicory by-product powder (RCP) on fresh spaghetti at different levels of semolina replacement in the following ratio of RCP: Semolina; 0:100, 5:95, 10:90, and 15:85. The phenolic profile of RCP, pasta samples and the *in vitro* antioxidant capacity were investigated. The cooking properties, color, texture parameters, and sensory profile of pasta have been evaluated.

2. Materials and methods

2.1. Red chicory powder and experimental pasta preparation

Geofur (Legnago, Verona, Italy) kindly supplied the red chicory by-product. The leaves were dried at 50 °C for 16 h. Later, dried leaves were milled with Grindomix GM 200 (Retsch, Haan, Germany) and sieved through a sieve with meshes of 0.2 mm to achieve a fine powder with a particle size of less than 0.2 mm.

Molino Casillo (Corato, Bari, Italy) kindly provided durum wheat semolina with the following nutritional content: carbohydrates 70 g/100 g, protein 12 g/100 g, fat 2 g/100 g, fiber 2.8 g/100 g. We prepared pasta, replacing semolina with 0, 5, 10, and 15% of red chicory by-product powder (RCP). Initially, we mixed the dry ingredients and added 35% of water at 40 °C. The blending process was carried out for 10 min employing a professional pasta machine (Mod. Lillodue, Bottene, Marano Vicentino, Italy; mixed capacity: 1 kg of flour; pasta output: 3 kg/h; dimensions: 25 x 53 x 25 cm; motor: 0.37 kW). Subsequently, the dough was extruded through a 1.75 mm bronze spaghetti die, resulting in spaghetti strands cut at the standard industrial length of 25 cm.

2.2. Proximate composition of RCP and pasta samples

The proximate composition of RCP and the fortified sample was evaluated in triplicate following standard procedures. Moisture content was determined using method 44–15A (AACC, 2000). The protein

content using method 976.05, total dietary fiber (TDF) using method 985.29, total fat using method 948.22, ash using method 942.05 (AOAC, 2007), and free sugars using the Megazyme assay kit K-SUFRG 06/14 (Megazyme, Wicklow, Ireland). All the measured chemical components were expressed as g/100 g of dry matter (DM).

2.3. Pasta properties determination

The fresh pasta's fully cooked time (FCT), and cooking loss (CL) were determined following the AACC methods 66–51.01, and 66-50 (AACC, 2000).

The water activity (a_w) was measured using a Hygropalm HC2-AW-meter (Rotronic, Milano, Italia) at 23 °C.

The swelling index (SI) was determined following the procedure outlined by Cleary and Brennan (2006). In brief, cooked pasta samples were weighed and placed in a steel container. After drying at 105 °C for 16 h, samples were cooled and weighed again. The results were calculated as follows:

$$SI = \frac{\text{weight of cooked pasta (g)} - \text{weight after drying (g)}}{\text{weight after drying (g)}} \quad (1)$$

2.4. Texture analysis

A TX-700 Texture Analyser (Lamy Rheology, Champagne au Mont d'Or, France) with a 25 kg load cell was used to measure firmness and adhesiveness of cooked samples. The firmness test was performed as Simonato et al. (2020) described, with minor modifications. We arranged a sample of five spaghetti strings in parallel on a sample holder (130152 R) to secure the samples in place and compressed them using a cylindrical probe (diameter 2.5 cm) operating at a speed of 0.1 mm/s. We compressed the samples at 50% of their original height (i.e., where the trigger force of 0.1 N was achieved). Firmness is the maximum force (N) needed to compress the pasta samples, whereas adhesiveness (N) is the negative peak force necessary to separate the probe from the sample surface.

2.5. Color analysis

Color analysis was conducted on the surface of raw fortified and control pasta samples using a reflectance colorimeter (Minolta Chroma meter CR-300, Osaka, Japan), following the CIE - $L^* a^* b^*$ color system. Spaghetti strands were disposed in an adapted stand that allowed a flat surface to be formed as reported by Doxastakis et al. (2007). The analyses were carried out at five different points on each pasta sample, and Minolta Equations (2) and (3) were applied to calculate the total color difference (ΔE):

$$\Delta E = \sqrt{\Delta L^2 + \Delta a^2 + \Delta b^2} \quad (2)$$

$$\Delta L = (L - L_0); \Delta a = (a - a_0); \Delta b = (b - b_0) \quad (3)$$

where L, L_0 , a, a_0 , b, and b_0 are the measured values of experimental samples or the control.

2.6. Extraction of red chicory phenolic compounds

RCP and dried uncooked pasta samples (CP, RC5, RC10, RC15) were extracted according to Balli, Cecchi, Innocenti, Bellumori, and Mulinacci (2021). Briefly, 100 mg of dried samples were extracted twice with 2 mL of EtOH:H₂O 7:3 v/v (HCOOH, 1%) for 30 min at 30 °C in a thermostat-controlled ultrasound bath (Labsonic LBS2, Treviglio, Italy) and then stirred for 12 h. The supernatants were separated by centrifugation (5000 g, 5 min) and dried with SpeedVac concentrator. The residues were dissolved in appropriate volumes of MeOH:H₂O 1:9 v/v (HCOOH, 1%) to fall within the dynamic calibration ranges, and

centrifugated before the UHPLC analysis. A further set of extractions was performed on unprocessed blends of semolina with RCP (0%, 5%, 10%, and 15%).

2.7. UHPLC-DAD-HRMS/MS analysis

The analyses for the phenolic profile of RCP and fortified pasta samples were carried out using a Vanquish Flex UHPLC system interfaced to Diode Array Detector FG and Orbitap Exploris 120 mass spectrometer (ThermoFisher Scientific, Milano, Italy), equipped with a heated electrospray ionization source (HESI-II). Chromatographic separation was performed using a Kinetex C18 column (2.1 × 100 mm, 2.6 μm; Phenomenex, Bologna, Italy), protected by a C18 Guard Cartridge (2.1 mm I.D.) and thermostated at 30 °C, and a binary gradient (0–6 min, 5–12%B; 6–16 min, 12–20%B; 16–21 min, 20–98%B; 21–24min 98%B) of H₂O (A) and MeCN (B), both containing 1% of HCOOH, at a flow rate of 500 μL min⁻¹ (injection volume, 5 μL). MS detection was performed in positive and negative ionization modes and using a Full MS data-dependent MS/MS acquisition mode. The resolution of the Full MS scans (scan range 150–1500 m/z) and dd-MS2 scans was set at 30k (FWHM). A stepped collision energy HCD (20, 40, and 60) was applied. Instrument control and spectra acquisition were carried out using Xcalibur software (Version 4.4, ThermoFisher Scientific). UV spectra were acquired in the 200–600 nm range and three wavelengths.

2.8. Quantitative analysis of red chicory phenolic compounds

Phenolic compounds in RCP were quantified by UHPLC-UV analysis at 330 (hydroxycinnamic acid), 350 (flavonoids), and 510 (anthocyanins) nm, using chlorogenic acid, quercetin-3-*O*-glucoside, and cyanidin-3-*O*-glucoside, respectively, as reference compounds. Levels of main sesquiterpene lactones were estimated by UHPLC-HRMS (extracted ion chromatogram, mass tolerance ± 5 ppm) and expressed as lactucin equivalents.

The external standards method was employed to determine the levels of main RCP compounds. A mixture of reference standards (chlorogenic acid, quercetin-3-*O*-glucoside, cyanidin-3-*O*-glucoside, and lactucin) was prepared at concentration of 1 mg/mL, and the calibration levels (concentration range of 0.5–50 μg/mL) were prepared by appropriate serial dilution and analyzed in triplicate. The linearity of the calibration curves was tested with the analysis of variance (ANOVA), and the linear model was found appropriate over tested concentration range (R^2 values > 0.999). The levels of the compounds were expressed as mg per g of dry material (mg/g DM) ± standard deviation ($n = 3$ extraction replicates).

The levels of the main RCP compounds in pasta samples were determined by UHPLC-HRMS (extracted ion chromatogram, mass tolerance ± 5 ppm) and expressed as recoveries compared to the unprocessed blends semolina/RCP.

2.9. Determination of ABTS, and FRAP assay

The pasta sample was dried at 50 °C for 16 h, finely ground, and sieved (0.2 mm). Five hundred milligrams of powdered pasta sample and RCP were extracted for 16 h, at room temperature, with 7.5 mL of MeOH: HCl (97:3) under continuous stirring in the dark.

After centrifugation at 3500g for 10 min at 15 °C, the supernatant was utilized to assess the antioxidant potential (FRAP), and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) assay (ABTS) as reported by Tolve et al. (2020).

The results were expressed as μmol of Trolox equivalent (TE) per gram of DM using a Trolox calibration curve ($R^2 = 0.996$).

2.10. Sensory evaluation

A quantitative descriptive sensory analysis (QDA) was conducted in

the sensory test room of the University of Verona; the QDA was designed according to UNI ISO8589 standards.

The panelists were carefully selected based on their sensory sensitivity and capacity to describe and communicate sensory perceptions. The panelists, consisting of 12 people (8 females and 4 males) between the ages of 23 and 54, recruited from the staff and students of the Department of Biotechnology at the University of Verona, were trained for 12 sessions of 1 h each to evaluate the specific sensory attributes of pasta samples (Bianchi, Giuberti, Cervini, & Simonato, 2022; Simonato et al., 2020; Tolve et al., 2020). Before participating in the study, informed consent was obtained from each subject via the following statement: "I am aware that my responses are confidential, and I agree to participate in this survey". An affirmative reply was required to enter the survey. Panelists could withdraw from the survey at any time without giving a reason. The products tested were safe for consumption. The panelists generated 11 sensory terms and were qualified to identify them. The sensory attributes that encompassed aspects of appearance (color uniformity), taste (sweetness, bitterness, vegetable taste), aroma (pasta, vegetable odor), and texture (gumminess, adhesiveness, elasticity, roughness) were evaluated.

Each judge received 15 g of cooked pasta in a covered container. The sample presentation order was balanced and randomized across the judges. A 9-point scale was employed, ranging from 1 (the lowest intensity) to 9 (the highest intensity) for each attribute. The mean sensory scores for each attribute were subsequently calculated based on the evaluations provided by the panel members. Additionally, panelists were asked to comment on the pasta's overall acceptability. We set at 5 the threshold for good acceptability within a scale from 1 to 9 points, following Simonato et al. (2020) and Tolve et al. (2020).

2.11. Statistical analysis

The analyses were carried out in triplicate, and all data is reported as mean values ± standard deviation. The variables were tested for significance using one-way analysis of variance (ANOVA) with a post-hoc Tukey's test ($p < 0.05$) using the software XLstat Premium Version (2023.2.1413, Addinsoft SARL, France).

3. Results and discussion

3.1. Proximate composition of red chicory powder and pasta samples

The fat content shows minimal variation among the samples, suggesting that adding radicchio powder does not significantly affect the fat composition of the pasta (Table 1). The protein content is constant in the samples, while ash content decreased at higher RCP concentrations. Furthermore, sugar levels gradually increased with the increasing amount of RCP as the concentration of dietary fibers. Consequently, as reported in Regulation (EC) No 1924/2006 of the European Parliament and of the Council (European Parliament, 2006), the fortified pasta samples can be claimed as a "source of fiber" for the RC5 and RC10 fortified samples (TDF > 3 g per 100 g of product) and a "high fiber content" food product for the RC15 sample (TDF > 6 g per 100 g of product).

Dietary fiber plays an essential role in human health: promoting digestive functions, preventing gastrointestinal disorders, reducing the cholesterol levels in the blood, lowering the risk of type 2 diabetes, and preventing obesity (Anderson et al., 2009; Da Rocha et al., 2021; Simonato et al., 2019). The recommended fiber intake should be 25–30 g daily (Jane, McKay, & Pal, 2019). However, this value is often not reached due to modern eating habits (Beres et al., 2016). Considering the Italian suggested serving size for fresh pasta of 100 g (Dello Russo et al., 2021), one dish of our experimental pasta could cover about 16%–25% of RDA. Our results show that these experimental samples could represent an exciting opportunity to introduce fiber-enriched foods and thus increase daily dietary fiber intake.

Table 1
Chemical composition of red chicory by-product powder (RCP), control sample (CP), and fortified pasta samples (RC5, RC10, RC15).

Chemical composition (g/100g)	RCP	CP	RC5	RC10	RC15
Moisture	9.8 ± 0.05	33.08 ± 0.61 ^a	30.79 ± 0.83 ^b	30.40 ± 0.58 ^b	30.36 ± 0.35 ^b
Fat	3.3 ± 0.05	2.10 ± 0.06 ^a	2.11 ± 0.05 ^a	2.13 ± 0.01 ^a	2.21 ± 0.06 ^a
Protein	15.8 ± 0.04	12.09 ± 0.09 ^b	12.17 ± 0.05 ^b	12.18 ± 0.01 ^b	12.57 ± 0.02 ^a
Ash	10.2 ± 0.03	13.22 ± 0.05 ^a	13.24 ± 0.05 ^a	12.64 ± 0.04 ^b	12.71 ± 0.05 ^b
Starch	–	68.80 ± 0.07 ^a	66.71 ± 0.05 ^b	61.08 ± 0.07 ^c	59.06 ± 0.52 ^d
Free Sugar	23.5 ± 0.25	1.20 ± 0.03 ^a	1.71 ± 0.03 ^b	2.41 ± 0.03 ^c	3.55 ± 0.03 ^d
Total Dietary Fiber	27.4 ± 0.77	2.78 ± 0.06 ^a	4.12 ± 0.10 ^b	5.28 ± 0.03 ^c	6.52 ± 0.03 ^d

Data are expressed as mean ± standard deviation. Values with different superscripts within the same lines significantly differ for $p < 0.05$.

RCP: red chicory by-product powder.

CP: control pasta.

RC5: fortified pasta with 5% of RCP.

RC10: fortified pasta with 10% of RCP.

RC15: fortified pasta with 15% of RCP.

3.2. Pasta properties determination

The technological properties of pasta samples are reported in Table 2. The water activity ranged between 0.93 and 0.95 according to the limits allowed by the Italian law for fresh pasta (Simonato et al., 2020).

The addition of RCP caused a significant increase in fully cooking time (FCT) values that ranged from 3.5 min for the control sample to 4.5 min for the RC15 fortified sample. The rise in the FCT might be due to the competitive hydration tendency between fiber of RCP, starch, and gluten protein that decreased the starch swelling (Pongpichaiudom & Songsermpong, 2018).

Another essential parameter to evaluate pasta quality is cooking loss (CL). Low cooking loss is an index of the high quality of pasta. It depends on the capability of the gluten network to retain the starch granules, its structural integrity, and other compounds (i.e., phenols and protein) during cooking processes (Nilusha, Jayasinghe, Perera, & Perera, 2019).

Table 2
Technological properties of control pasta (CP) and pasta fortified with different percentages of red chicory by-product powder (RC5, RC10, RC15).

Pasta samples	a_w	FCT (min)	CL (%)	SI (g water/g dry pasta)
CP	0.93 ± 0.01 ^{ab}	3.5	3.23 ± 0.02 ^d	0.42 ± 0.01 ^a
RC5	0.95 ± 0.00 ^a	4.0	5.97 ± 0.57 ^c	0.40 ± 0.08 ^{ab}
RC10	0.94 ± 0.00 ^{ab}	4.0	8.14 ± 0.63 ^b	0.41 ± 0.00 ^a
RC15	0.93 ± 0.01 ^b	4.5	10.26 ± 1.00 ^a	0.39 ± 0.00 ^b

Data are expressed as mean ± standard deviation. Values with different superscripts within the same lines significantly differ for $p < 0.05$.

a_w : water activity.

FCT: fully cooked time (min).

CL: cooking loss (%).

SI: swelling index (g water/g dry pasta).

CP: control pasta.

RC5: fortified pasta with 5% of RCP.

RC10: fortified pasta with 10% of RCP.

RC15: fortified pasta with 15% of RCP.

In pasta samples, the CL increased from 3.23 for the control sample to 10.26 for the RC15 fortified samples. A 12% or less cooking loss was considered acceptable and indicative of good-quality pasta (Espinoza-Solis et al., 2019). Fortifying pasta resulted in a notable decrease in the SI value, and this trend was reported similarly in other studies (Marinelli, Padalino, Nardiello, Del Nobile, & Conte, 2015; Tolve et al., 2020). This decrease might relate to a weaker and less cohesive structure of the fortified sample, which could promote the leaching of soluble compounds (e.g., starch granules and RCP particles).

3.3. Texture analysis

Textural properties are reported in Table 3. The increase of RCP concentrations in the formulas enhanced the firmness of fortified pasta. Indeed, the lowest values were observed in the CP and significantly higher for the RC10 and RC15 samples. Our results agree with Rakhesh, Fellows, and Sissons (2015), who reported how the firmness of pasta enriched with barley, pollard, or retrograded starch increased compared to control pasta while obtaining opposite results for high-amylose starch, inulin, carboxymethyl cellulose, guar gum, bran, and β -glucan-enriched flour. Similarly, (Peressini, Cavarape, Brennan, Gao, & Brennan, 2020) reported that the firmness of pasta enriched with psyllium and beta-glucans increased significantly. The same authors (Peressini et al., 2020) argued that psyllium fiber could have entrapped the starch granules, generating a network that can reduce the water intake and swelling of starch, thus increasing firmness. Moreover, many authors reported higher firmness with pasta made with different by-products, namely grape pomace powder (Tolve et al., 2020), coconut residue (Sykut-Domańska et al., 2020), Okara powder (Kamble, Singh, Rani, & Pratap, 2019), potato juice (Kowalczewski et al., 2015), and tomato by-product (Padalino et al., 2017).

The adhesiveness increased with higher concentrations of RCP, with the highest value observed in the RC15 sample. Pasta adhesiveness is due to substances released from the gluten network during cooking (especially amylopectin) that adhere to the pasta surface (Aravind, Sissons, Egan, & Fellows, 2012) or water absorption by some fibers that form a viscous layer on the surface of the cooked pasta as reported by Rakhesh et al. (2015).

3.4. Color analysis

The pasta color analysis is reported in Table 4, while an image of the raw pasta samples is reported in Fig. 1. The results show a significant reduction in lightness (L^*) between the control sample and the fortified ones. In the samples, a significant increase in redness (a^*) was also evident for all fortified samples. Compared to the control sample, a notable reduction in yellowness (b^*) was observed with the incremental incorporation of RCP in fortified spaghetti. It must be considered that the inhomogeneity of the sample surfaces due to the irregular drying of pasta after extrusion that determine the high values of standard deviation.

Table 3
Texture analysis of control pasta (CP) and pasta fortified with different percentages of red chicory by-product powder (RC5, RC10, and RC15).

Pasta sample	Firmness (N)	Adhesiveness (N)
CP	12.16 ± 5.57 ^b	0.26 ± 0.11 ^{ab}
RC5	15.05 ± 5.24 ^b	0.26 ± 0.01 ^a
RC10	24.39 ± 1.48 ^a	0.29 ± 0.08 ^{ab}
RC15	25.92 ± 3.35 ^a	0.36 ± 0.08 ^b

Data are expressed as mean ± standard deviation. Values with different superscripts within the same lines significantly differ for $p < 0.05$.

CP: control pasta.

RC5: fortified pasta with 5% of RCP.

RC10: fortified pasta with 10% of RCP.

RC15: fortified pasta with 15% of RCP.

Table 4

Color analysis of control pasta (CP) and pasta fortified with different percentages of red chicory by-product powder (RC5, RC10, RC15).

Pasta sample	L^*	a^*	b^*	Sample confrontation	ΔE
CP	72.24 ± 1.84 ^a	0.71 ± 0.16 ^c	28.73 ± 2.02 ^a	CP-RC5	31.27
RC5	44.88 ± 1.72 ^b	7.37 ± 0.25 ^b	15.37 ± 0.63 ^b	CP-RC10	39.47
RC10	36.57 ± 1.62 ^c	7.91 ± 0.40 ^a	13.67 ± 0.65 ^c	CP-RC15	44.29
RC15	32.69 ± 1.77 ^d	7.33 ± 0.41 ^b	10.18 ± 0.56 ^d	RC5-RC10	8.50
				RC5-RC15	13.24
				RC10-RC15	5.25

Data are expressed as mean ± standard deviation. Values with different superscripts within the same lines significantly differ for $p < 0.05$.

L^* : lightness.

a^* : redness.

b^* : yellowness.

CP: control pasta.

RC5: fortified pasta with 5% of RCP.

RC10: fortified pasta with 10% of RCP.

RC15: fortified pasta with 15% of RCP.

3.5. Untargeted profiling of red chicory by-product powder (RCP)

UHPLC-DAD-HRMS performed a quali-quantitative profile of RCP to verify the presence and levels of phenolic compounds specific to red chicory in the by-product powder used for pasta fortification.

In Table 5 the detected phytochemicals and the RCP content of the significant compounds are reported.

Thirty-two secondary metabolites belonging to hydroxycinnamic acid (HA), flavonoid (F), anthocyanin (A), and sesquiterpene lactone (SL) classes, were detected in RCP and identified by UV and HRMS/MS data, along with MS data from literature and databases, chemotaxonomic data and the use of standard compounds, whenever available. According to previous studies on red chicory (Feroli et al., 2015), the HAs chlorogenic acid and chicoric acid, the Fs quercetin 3-*O*-malonylglucoside and luteolin *O*-glucuronide, and cyanidin 3-*O*-malonylglucoside resulted the main distinctive phytochemicals of RCP. Also, the major SLs of chicory leaves, lactucin, dihydrolactucin, 8-deoxylactucin, lactucopicrin, and their derivatives (Feroli et al., 2015; Giambanelli, D'Antuono, Feroli, Frenich, & Romero-González, 2018), were detected in RCP. Regarding the quantitative profile of RCP, chlorogenic acid (21% of total phenolic concentration), chicoric acid (21%), quercetin-3-*O*-malonylglucoside (8%), luteolin-*O*-glucuronide (12%) and cyanidin-*O*-malonylglucoside (28%) were the most abundant compounds. Their levels agreed with previous studies on red chicory varieties (Feroli et al., 2015). SL levels were in the 0.06–0.39 mg/g range, with a total SL content of 0.61 mg/g.

These data on RCP composition indicated that this by-product contains high levels of phenols compounds, and its use as a functional

ingredient provides added value to fortified pasta.

3.6. Phenolic profiling of fortified pasta and antioxidant activity

When the extraction procedure of red chicory phenolic compounds was applied to the fortified pasta samples, a substantial reduction in the content of extractable phenolic compounds than to the calculated levels was observed. The recovered amounts (data not shown) were widely variable, ranging from 0.2 to 13% for HAs to 5–28% for Fs, while anthocyanins were not recovered at all (<0.2%). Conversely, unprocessed blends of semolina with RCP (0%, 5%, 10%, and 15%) showed levels of compounds comparable to those calculated from RCP composition.

The poor extraction recoveries of phenolic compounds from fortified pasta are likely due to their interaction with the semolina components, namely gluten or starch, in the highly structured matrix formed during kneading and pasta processing. These non-covalent interactions (hydrogen bonds, hydrophobic interaction, and electrostatic and ionic interactions), as already reported, tightly complex the phenolic compounds in food systems, preventing their release during extraction or washing with alcoholic solutions (Nemli et al., 2024; Wang et al., 2023; Zhang et al., 2024). Thus, the apparent low levels of RCP bioactive extracted from pasta samples may be attributed to the difficulties of the extraction procedure in recovering these compounds from the matrix. However, literature data indicate that the complexation of phenolic compounds with starch and proteins affects starch digestion and phenolic compound bioavailability; therefore, these complexes in the food system can be used for both the modulation of glycemic response and the targeted delivery of phenolic compounds (Nemli et al., 2024; Wang et al., 2023; Zhang et al., 2024). So, further studies should focus on evaluating the bioaccessibility of phenolic compounds in RCP-fortified pasta through *in vitro* digestion.

Table 6 reports the FRAP and ABTS assay results of red chicory by-product powder and pasta samples. In the fortified spaghetti, the antioxidant activity assessed by FRAP and ABTS assays shows a notable increase with the addition of RCP. RCP15 antioxidant power resulted in 2-fold and 16-fold higher than the control sample for FRAP and ABTS assays, respectively. Bianchi et al. (2021), in pasta formulated by replacing wheat semolina with 15 g/100 g of maqui berries powder, reported higher antioxidant capacity detected through FRAP and ABTS assays of 57-fold and 50-fold compared to control pasta.

These differences could be attributed to the quantity and type of phenolic compounds in maqui berries, which better enhance antioxidant capacity. Moreover, as reported by Bianchi, Cervini, et al. (2022), the antioxidant capacity of muffins fortified with distilled grape pomace was positively correlated with certain polyphenols' classes: flavanols for FRAP, and anthocyanins, flavones, and flavonols for ABTS assay, respectively.

3.7. Sensory evaluation

A quantitative descriptive analysis (QDA) served as a conclusive



Fig. 1. Image of pasta samples. From left to right: control pasta without red chicory by-product powder (CP), pasta with 5% of RCP (RC5), pasta with 10% of RCP (RC10), and pasta with 15% of RCP (RC15). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

Table 5
UHPLC-DAD-HRMS data of compounds detected in red chicory by-product powder (RCP).

Metabolite class	N	Compound	RT (min)	Molecular Formula	Adduct	Precursor (m/z)	Error (ppm)	IL ^a	RCP Content (mg/g)
HA	1	Caffeoyl quinic acid	2.0	C ₁₆ H ₁₈ O ₉	[M-H] ⁻	353.0874	-0.7	2	-
HA	2	Caftaric acid	2.4	C ₁₃ H ₁₂ O ₉	[M-H] ⁻	311.0410	0.6	2	0.79 ± 0.01 ^b
	3	Esculin	2.4	C ₁₅ H ₁₆ O ₉	[M-H] ⁻	339.0721	-0.2	1	-
SL	4	Dihydrolactucin-sulfate	3.2	C ₁₅ H ₁₈ O ₈ S	[M+H] ⁺	359.0795	-0.3	2	-
HA	5	Chlorogenic acid	3.6	C ₁₆ H ₁₈ O ₉	[M-H] ⁻	353.0874	-0.7	1	9.03 ± 0.23 ^b
	6	Cichorioside	3.9	C ₁₅ H ₁₆ O ₉	[M + HCOO] ⁻	485.1664	-0.4	2	-
SL	7	Dihydrolactucin	4.7	C ₁₅ H ₁₈ O ₅	[M+H] ⁺	279.1227	-0.8	2	0.08 ± 0.01 ^c
SL	8	Oxalyl lactucin	5.2	C ₁₇ H ₁₆ O ₈	[M-H] ⁻	347.0772	-0.2	2	-
A	9	Cyanidin O-glucoside	5.3	C ₂₁ H ₂₀ O ₁₁	[M] ⁺	449.1077	-0.7	1	2.10 ± 0.18 ^d
F	10	Quercetin O-malonylhexoside-glucuronide	5.5	C ₃₀ H ₃₀ O ₂₁	[M+H] ⁺	727.1346	-0.9	2	-
SL	11	Lactucin	5.5	C ₁₅ H ₁₆ O ₅	[M+H] ⁺	277.1070	-0.8	1	0.06 ± 0.01 ^c
F	12	Taxifolin O-dimalonylhexoside	5.6	C ₃₃ H ₃₆ O ₂₃	[M+H] ⁺	801.1713	0.2	2	-
F	13	Taxifolin O-malonylhexoside	5.7	C ₂₄ H ₂₄ O ₁₅	[M+H] ⁺	553.1181	-0.4	2	-
HA	14	3-Feruloylquinic acid	7.1	C ₁₇ H ₂₀ O ₉	[M-H] ⁻	367.1033	-0.5	2	-
A	15	Cyanidin O-malonylhexoside	6.9	C ₂₄ H ₂₃ O ₁₄	[M] ⁺	535.1074	-0.6	2	-
A	16	Delphinidin O-malonylhexoside	7.3	C ₂₄ H ₂₂ O ₁₅	[M] ⁺	551.1026	-1.0	2	-
A	17	Cyanidin O-dimalonylhexoside	7.5	C ₃₃ H ₃₄ O ₂₂	[M] ⁺	783.1605	-1.2	2	-
F	18	Quercetin O-glucuronide	7.7	C ₂₁ H ₁₈ O ₁₃	[M+H] ⁺	479.0816	-0.7	2	-
A	19	Cyanidin O-malonylglucoside	8.3	C ₂₄ H ₂₃ O ₁₄	[M] ⁺	535.1074	-0.6	2	11.72 ± 1.2 ^d
SL	20	Dyhydrodeoxylactucin sulfate	8.6	C ₁₅ H ₁₈ O ₇ S	[M+H] ⁺	343.0845	-0.3	2	-
F	21	Rutin	9.7	C ₂₇ H ₃₀ O ₁₆	[M+H] ⁺	611.1602	-0.8	1	-
F	22	Quercetin O-glucuronide	9.9	C ₂₁ H ₁₈ O ₁₃	[M+H] ⁺	479.0816	-0.7	2	-
HA	23	Chioric acid	10.0	C ₂₂ H ₁₈ O ₁₂	[M-H] ⁻	473.0723	-0.9	2	9.07 ± 0.55 ^b
F	24	Luteolin O-glucuronide	10.2	C ₂₁ H ₁₈ O ₁₂	[M+H] ⁺	463.0868	-0.7	2	5.30 ± 0.14 ^c
SL	25	8-Deoxylactucin	11.0	C ₁₅ H ₁₆ O ₄	[M-H] ⁻	259.0976	0.1	2	0.39 ± 0.02 ^c
F	26	Quercetin O-malonylhexoside	11.4	C ₂₄ H ₂₄ O ₁₅	[M+H] ⁺	551.1028	-0.7	2	3.58 ± 0.16 ^c
HA	27	Dicafeoyl quinic acid	12.0	C ₂₅ H ₂₄ O ₁₂	[M-H] ⁻	515.1196	0.1	2	0.89 ± 0.17 ^b
F	28	Quercetin O-malonylhexoside	12.3	C ₂₄ H ₂₄ O ₁₅	[M+H] ⁺	551.1028	-0.7	2	-
HA	29	Caffeoyl-feruloyltartaric acid	13.9	C ₂₃ H ₂₀ O ₁₂	[M-H] ⁻	487.0882	0.1	2	-
F	30	Isorhamnetin O-malonylhexoside	15.0	C ₂₅ H ₂₄ O ₁₅	[M+H] ⁺	565.1183	-0.9	2	-
SL	31	Lactucopicrin 15-oxalate	17.6	C ₂₅ H ₂₂ O ₁₀	[M-H] ⁻	481.1138	-0.4	2	-
SL	32	Lactucopicrin	17.9	C ₂₃ H ₂₂ O ₇	[M-H] ⁻	409.1291	-0.3	2	0.07 ± 0.01 ^c

Data are expressed as mean ± standard deviation. Values with different superscripts within the same lines significantly differ for p < 0.05.

^a IL, Identification level were assigned according to Sumner et al., 2007.

^b chlorogenic acid equivalents.

^c lactucin equivalents.

^d cyanidin 3-O-glucoside equivalents.

^e quercetin 3-O-glucoside equivalents.

Table 6

Total phenolic component (TPC) and antioxidant activity (FRAP and ABTS) of red chicory powder (RCP), uncooked control pasta (PC), and uncooked fortified pasta with different percentages of red chicory powder (RC5, RC10, and RC15).

Samples	FRAP (μmol TE/g dm)	ABTS (μmol TE/g dm)
RCP	6.03 ± 0.42	77.87 ± 0.12
PC	2.46 ± 1.02 ^a	1.49 ± 0.10 ^a
RC5	2.78 ± 0.77 ^a	13.65 ± 0.95 ^b
RC10	3.57 ± 0.87 ^{ab}	18.46 ± 1.37 ^{bc}
RC15	4.67 ± 0.52 ^b	24.67 ± 1.65 ^c

Data are expressed as mean ± standard deviation. Values with different superscripts within the same lines significantly differ for p < 0.05.

μmol TE/g dm: μmol of Trolox equivalents per gram of dry matter.

FRAP: Ferric reducing antioxidant power.

ABTS: 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid).

RCP: red chicory by-product powder.

CP: control pasta.

RC5: fortified pasta with 5% of RCP.

RC10: fortified pasta with 10% of RCP.

RC15: fortified pasta with 15% of RCP.

examination of the pasta samples, aiming to detect changes in sensory attributes resulting from RCP enrichment (Fig. 2; Table S1- supplementary material). Regarding the taste attributes (sweetness, bitterness, and vegetable taste), CP was sweeter compared to the fortified samples, while RC15 was perceived as more bitter and vegetal taste compared to the other samples. Color uniformity ranged between 3.8 and 8.2, and the

CP sample showed higher color uniformity compared to other pasta samples. The uneven distribution and size of RCP particles in the dough could explain this difference. CP received relatively high ratings for gumminess, adhesiveness, and elasticity. The general flavor of the RC15 sample obtained a similar score compared to the control sample (Table S1 – Supplementary material). Furthermore, irrespective of the quantity of RCP added, the panelists rated the overall acceptability of the fortified pasta positively (the overall acceptability scores were higher than 5).

4. Conclusions

The results of the present study indicated that RCP is a plant by-product with high levels of fiber (27%) and healthy bioactive compounds (4.3%), which could improve the nutritional and nutraceutical value of fortified pasta. RCP phenolic compounds showed a high affinity with the semolina components, likely gluten or starch, as indicated by their low recoveries from fortified pasta under the adopted extraction conditions.

The amount of total dietary fiber in the RCP-fortified pasta samples allows RC5 and RC10 (4.12 and 5.28 g/100 g, respectively) to benefit from the claim "source of fiber" while RC15 could benefit from the claim "high fiber" containing 6.52 g/100 g. The fortification also influenced the technological properties of pasta. The cooking loss increased while the swelling index decreased. The texture analysis shows that firmness and adhesiveness increased proportionally with fortification. From a sensory viewpoint, vegetable taste, vegetable odor, roughness, and

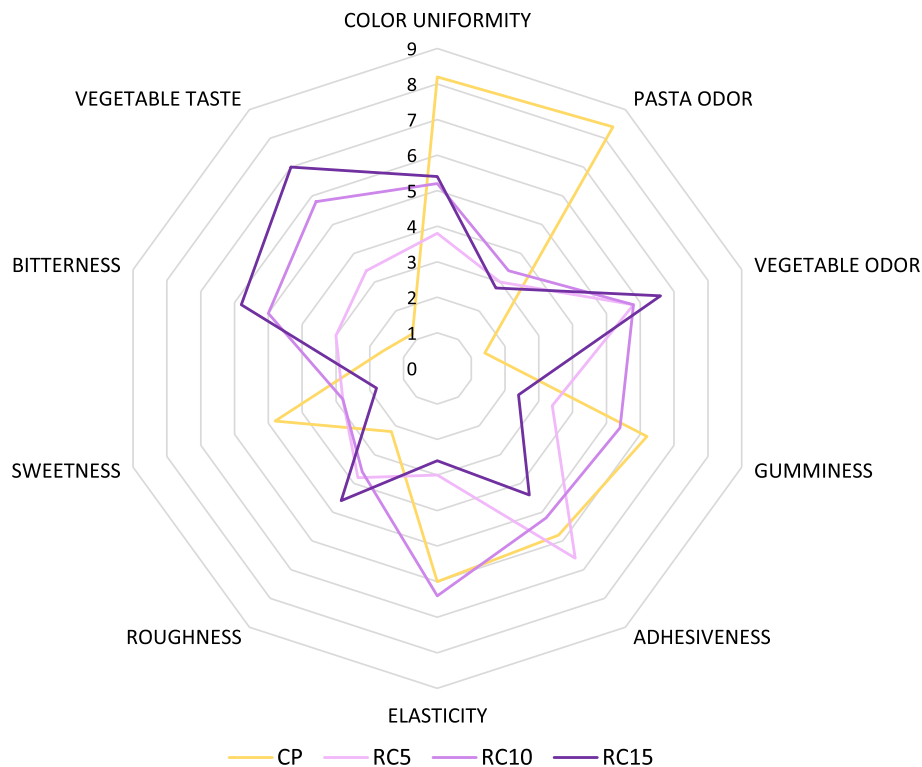


Fig. 2. Spider plot of the sensorial attributes and intensities of control pasta (CP) and pasta with 5% of RCP (RC5), pasta with 10% of RCP (RC10), and pasta with 15% of RCP (RC15).

bitterness increased with the fortification with RCP, while sweetness, adhesiveness, and gumminess decreased with the fortification. Moreover, panelists rated the overall acceptability of the 15% pasta and control by expressing equal ratings for both samples. In conclusion, RCP could constitute an intriguing ingredient for producing functional pasta fortified with antioxidant compounds and dietary fiber, potentially contributing to positive effects on human health and enhancing the economic value of the radicchio food chain by reusing this by-product.

Funding

This research received no external funding.

CRediT authorship contribution statement

Federico Bianchi: Writing – review & editing, Writing – original draft, Visualization, Supervision, Conceptualization. **Valentina Santoro:** Writing – original draft, Investigation, Formal analysis, Data curation. **Ilaria Pasqualoni:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis, Data curation. **Margherita Bruttomesso:** Writing – review & editing, Validation, Conceptualization. **Corrado Rizzi:** Writing – review & editing, Supervision, Data curation. **Anna Lisa Piccinelli:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Data curation. **Barbara Simonato:** Writing – review & editing, Supervision, Methodology.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The data that has been used is confidential.

Acknowledgments

The University of Verona is acknowledged for the support provided through the extraordinary university fund for Open Access publications. This study was carried out within the Interconnected Nord-Est Innovation Ecosystem (iNEST) and received funding from the European Union Next-Generation EU (PIANO NAZIONALE DI RIPRESA E RESILIENZA (PNRR) – MISSIONE 4 COMPONENTE 2, INVESTIMENTO 1.5 – D.D. 1058 23/06/2022, ECS00000043). This manuscript reflects only the authors' views and opinions, neither the European Union nor the European Commission can be considered responsible for them.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.lwt.2024.116358>.

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