UNIVERSITA' DEGLI STUDI DI VERONA

DEPARTMENT OF

Surgery, Dentistry, Paediatrics and Gynaecology

GRADUATE SCHOOL OF

Medicine and Surgery

DOCTORAL PROGRAM IN

Cardiovascular Science

WITH THE FINANCIAL CONTRIBUTION OF

University of Verona

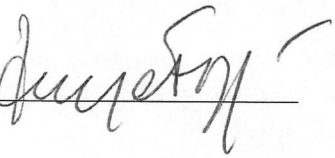
Cycle / year (1° year of attendance) 34° /2018

TITLE OF THE DOCTORAL THESIS

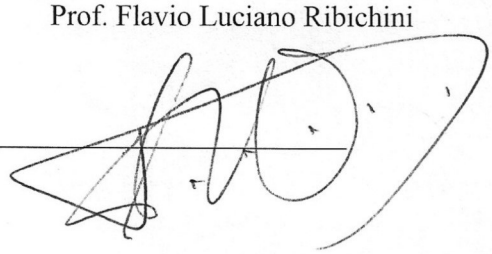
STRAIN AND CONVENTIONAL ECHOCARDIOGRAPHIC PARAMETERS AS PREDICTORS OF SUCCESSFUL VA ECMO WEANING

S.S.D. MED/11

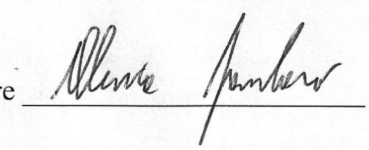
Coordinator: Prof. Giuseppe Faggian

Signature 

Tutor: Prof. Flavio Luciano Ribichini

Signature 

Doctoral Student: Dr. Alessia Gambaro

Signature 

ABSTRACT

Introduction: Venoarterial extracorporeal membrane oxygenation (VA ECMO) is a type of temporary mechanical circulatory support and extracorporeal gas exchange device for acute cardiovascular and respiratory failure. The population of VA ECMO patients is characterised by high mortality rates. The causes of death are several: haemorrhages, septic complications, haemorrhagic or ischaemic stroke, new episodes of cardiovascular failure after VA ECMO removal. The protocol for weaning from VA ECMO often changes from hospital to hospital and it has not been validated worldwide. The decision to wean a patient from VA ECMO is particularly challenging and crucial because it is necessary to understand whether the patient can survive without VA ECMO support, but it is also important to avoid any delays to reduce the risk of complications.

Objectives: the aim of this pilot study was to assess whether strain obtained through speckle tracking could give additional information to hemodynamic, and conventional echocardiographic parameters in identifying patients who will develop adverse outcomes within 3 (± 1) months from VA ECMO removal.

Methods: observational prospective pilot study delivered over 3 years at Royal Brompton and Harefield NHS Foundation Trust and Azienda Ospedaliera Universitaria Integrata Verona. VA ECMO patients have been screened, and the VA ECMO patients who underwent VA ECMO removal (not for palliation) after a VA ECMO weaning trial have been recruited and followed up. Conventional echocardiographic, haemodynamic and speckle tracking (strain) parameters of patients developing a composite clinical outcome within 3 (± 1) months post VA ECMO removal (death for any reason, new necessity of high dose of ino-vasopressors or mechanical circulatory support device after VA ECMO removal, new hospitalisation for heart failure/cardiovascular shock) have been compared with those of patients free from clinical outcomes. Furthermore, a sub-analysis on composite cardiac outcome development and exploratory ROC analysis have been performed.

Results: Over 3 years of recruitment, 92 VA ECMO patients have been screened. 21 patients met the eligibility criteria for the study. Of these, 19

patients could be analysed. 5 patients experienced the composite clinical outcome of interest (3 had cardiac outcomes, 2 were complicated by septic shock). At the lowest flow of VA ECMO support, the median ejection fraction (EF) of the clinical outcome + patients was 23.9% (IQR 15.4), conversely it was 45.8% (IQR 18.7) in clinical outcome – patients ($p = 0.06$). Considering right ventricular (RV) function, strain and haemodynamic parameters, no significant differences were found between clinical outcome + and clinical outcome – patients. The indexed end diastolic volume (iEDV) was significantly lower for patient free from clinical outcomes compared to the others (respectively 44.7 ml m^{-2} IQR 17.9; 70.4 ml m^{-2} IQR 43.0, $p < 0.01$). Analysing the recruited patients on the basis of cardiac outcome development, the median iEDV remained significantly higher in cardiac outcome + (112.5 ml $min^{-1}m^{-2}$ IQR 47.1; $p = 0.01$). Furthermore, the median circumferential strain was -5.6% (IQR 1.0) and the EF 23.0% (IQR 4.2) in cardiac outcome + patients. Conversely, circumferential strain was -15.5% (IQR 6.5) and EF was 45.8% (IQR 15.6) in patients free from cardiac outcomes. Graphically organising the echocardiographic findings, it was possible to observe that compared to outcome free patients, the absolute values of EF and circumferential strain were lower in cardiac outcome + , while RV free wall longitudinal strain was lower in clinical outcome + patients. At the ROC analysis, the best cut point to discriminate patients developing clinical outcomes and patients free from outcomes at the lowest flow of VA ECMO support was for EF 26.32% (AUC 0.79), for left ventricle outflow track velocity time integral 14.36cm, (AUC 0.78), for cardiac index 2.5ml min^{-1} m^{-2} (AUC 0.71) and for RV free wall longitudinal strain -12.0%. (AUC 0.75).

Conclusions: the decision to wean a patient from VA ECMO is complex and require the assessment of multiple variables (echocardiographic, haemodynamic and respiratory). According to our analysis, large iEDV and low EF predispose to the development of clinical outcomes. Furthermore, low values of circumferential strain and RV free wall longitudinal strain may be indicative of development of cardiac and clinical outcomes respectively. In our view, it is particular important to discriminate the reasons for VA ECMO implantation and

the causes of VA ECMO weaning failure to properly identify, in a future study, the predictors of VA ECMO weaning success.

SOMMARIO

Introduzione: Venoarterial extracorporeal membrane oxygenation (VA ECMO) è un tipo di supporto meccanico di circolo e respiratorio temporaneo usato in caso di gravi insufficienze cardiovascolari e respiratorie. La popolazione di pazienti supportati da VA ECMO è caratterizzata da un alto tasso di mortalità. Le cause di morte in corso di VA ECMO sono molteplici: shock emorragico, complicanze settiche, emorragie cerebrali o ictus ischemici, nuovi episodi di shock cardiogeno post rimozione di VA ECMO, ecc. I protocolli di svezzamento da VA ECMO differiscono da ospedale a ospedale, e non esiste un protocollo di svezzamento validato internazionalmente. La decisione di svezzare un paziente da VA ECMO è particolarmente difficile perché non solo è necessario capire se il paziente è in grado di sopravvivere senza tale supporto, ma è anche importante evitare qualsiasi ritardo nello svezzamento per ridurre il rischio di complicanze.
Obiettivi: lo scopo di questo studio pilota era quello di valutare se lo strain ottenuto tramite metodica speckle tracking fosse in grado di fornire delle informazioni aggiuntive ai parametri ecocardiografici convenzionali ed emodinamici nell'identificazione di pazienti che svilupperanno degli eventi avversi entro 3 (± 1) mesi dalla rimozione del VA ECMO.
Metodi: studio pilota osservazionale prospettico condotto presso gli ospedali Royal Brompton and Harefield NHS Foundation Trust e l'Azienda Ospedaliera Universitaria Integrata Verona nell'arco di 3 anni. I pazienti supportati da VA ECMO sono stati screenati e quelli che sono stati sottoposti a rimozione di VA ECMO (non a scopo di palliazione) dopo una prova di svezzamento sono stati reclutati nello studio e seguiti nel tempo. I valori dei parametri ecocardiografici convenzionali, emodinamici e speckle tracking (strain) dei pazienti che hanno sviluppato un outcome clinico composito entro 3 (± 1) mesi dalla rimozione di VA ECMO (morte per qualsiasi causa, nuova necessità di somministrazione di farmaci ino-vasopressori o nuova necessità di supporto meccanico di circolo, o nuova ospedalizzazione per scompenso cardiaco/shock cardiovascolare) sono

stati confrontati con quelli dei pazienti che non hanno sviluppato alcun outcome. Sono stati inoltre eseguite una sub-analisi sullo sviluppo di eventi cardiaci e una analisi esplorativa ROC.

Risultati: nell'arco di 3 anni sono stati screenati 92 pazienti supportati da VA ECMO. 21 pazienti sono stati reclutati e di questi 19 sono stati analizzati. 5 pazienti hanno sviluppato l'outcome clinico composito di interesse (3 specificatamente un outcome cardiaco, 2 hanno avuto delle complicanze da shock settico). Al più basso flusso di supporto VA ECMO, la mediana della frazione di eiezione (EF) del gruppo di pazienti positivi per outcome clinico era 23.9% (IQR 15.4), invece era pari al 45.8% (IQR 18.7) nei pazienti negativi per outcome clinico ($p = 0.06$). Considerando invece la funzione del ventricolo destro (RV), lo strain e i parametri emodinamici, non sono state riscontrate differenze significative tra i pazienti positivi per outcome clinici e quelli negativi. Il volume telediastolico indicizzato (iEDV) era significativamente più basso nei pazienti negativi per eventi clinici rispetto ai pazienti positivi (rispettivamente 44.7 ml m^{-2} IQR 17.9; 70.4 ml m^{-2} IQR 43.0, $p < 0.01$). Analizzando i pazienti reclutati sulla base dello sviluppo di eventi cardiaci, la mediana dell'iEDV è rimasta significativamente più alta nel gruppo di pazienti positivi per outcome cardiaco (112.5 ml $min^{-1}m^{-2}$ IQR 47.1; $p = 0.01$). Il valore mediano dello strain circonferenziale era -5.6% (IQR 1.0) e quello della EF 23.0% (IQR 4.2) per i pazienti positivi per outcome cardiaco. Mentre, il valore mediano dello strain circonferenziale era -15.5% (IQR 6.5) e quello della EF era 45.8% (IQR 15.6) per i pazienti liberi da eventi cardiaci. Organizzando graficamente i risultati ecocardiografici è stato possibile osservare che, rispetto ai pazienti liberi da eventi, il valore assoluto della EF e dello strain circonferenziale è minore nei pazienti con outcome cardiaci, mentre è minore lo strain longitudinale della parete libera del RV nei pazienti positivi per outcome clinici. All'analisi ROC, i migliori cut point per discriminare i pazienti che sviluppavano un outcome da quelli liberi da outcome erano per l'EF 26.32% (AUC 0.79), per l'integrale velocità tempo al tratto di efflusso del ventricolo sinistro 14.36cm, (AUC 0.78), per l'indice cardiaco 2.5ml min^{-1} m^{-2} (AUC 0.71) e per lo strain longitudinale della parete libera del RV -12.0%. (AUC 0.75).

Conclusioni: la decisione di svezzare un paziente da VA ECMO è complessa e richiede una valutazione multiparametrica (ecocardiografica, emodinamica, respiratoria). Sulla base della nostra analisi, degli elevati valori di iEDV e delle basse EF predispongono allo sviluppo di outcome clinici, tuttavia anche valori bassi di strain circonferenziale e di strain longitudinale della parete libera del RV possono essere indicativi di sviluppo di outcome cardiaci e clinici rispettivamente. Dal nostro punto di vista, è fondamentale distinguere le ragioni per cui un VA ECMO viene impiantato e le cause di fallimento dello svezzamento da VA ECMO per identificare correttamente, in uno studio futuro, i predittori di successo per lo svezzamento da VA ECMO.

INDEX

LIST OF ACRONYMS

AF atrial fibrillation

AUC area under the curve

AUROC area under the receiver operating characteristics curve

CABG coronary artery bypass graft

CI cardiac index or confidence interval

CO cardiac output

CO_2 carbon dioxide

CVP central venous pressure

DC dendritic cell

ELSO Extracorporeal Life Support Organization

FiO_2 fraction of inspired oxygen

ICU intensive care unit

IU international unit

IVC inferior vena cava

IHD ischaemic heart disease

IJ internal jugular

LA left atrium

LV left ventricle

LVAD left ventricular assist device

LVETc corrected left ventricular ejection time

MAP mean arterial pressure

MCS mechanical circulatory support

MDSC myeloid suppressive cell

OR odds ratio

PaO_2 partial pressure of oxygen

PAWP pulmonary arterial wedge pressure

PEEP Positive end-expiratory pressure

RA right atrium

ROC receiver operating curve

RV right ventricle

RVAD right ventricular assist device
RVSP right ventricular systolic pressure
SvO_2 mixed venous oxygen saturation
TAH total artificial heart
TAPSE tricuspid annular plane systolic excursion
TOE transoesophageal echocardiogram
TTE transthoracic echocardiogram
VAD ventricular assist device
VA ECMO Venoarterial extracorporeal membrane oxygenation
cVA ECMO central VA ECMO
pVA ECMO peripheral VA ECMO
VV ECMO Venovenous extracorporeal membrane oxygenation

VA ECMO, HOW IT WORKS

Venoarterial extracorporeal membrane oxygenation (VA ECMO) is a type of temporary mechanical circulatory support (MCS) and extracorporeal gas exchange device for acute cardiovascular and respiratory failure(1). It withdraws desaturated blood from venous system, it oxygenates and removes carbon dioxide (CO_2), and then it pumps the blood with a non-pulsatile flow in the patient's arterial circulation. In this way, VA ECMO provides life-saving support and can be used as bridge to decision, recovery, heart transplant (HT) or durable assist device (left ventricular assist device (LVAD)/total artificial heart (TAH)). More specifically, VA-ECMOs consist of a venous (inflow, drainage) cannula, a pump, an oxygenator, and an arterial (outflow, return) cannula. VA ECMO can be peripheral or central according to the vascular accesses used. Central VA ECMOs (cVA ECMO) are implanted in operating theatres, often in post-cardiotomy patients who failed to come off bypass(2). The drainage cannula is placed in the right atrium (RA) and the return cannula is placed into the ascending aorta; these cannulas can be tunnelled to allow chest closure(3).

In peripheral VA ECMOs (pVA ECMO), the drainage cannula is positioned through the femoral vein, at the level of the intrahepatic inferior vena cava (IVC) and sometimes its tip is advanced in the RA. Typically this cannula has multiple holes (multistage cannula) to allow a more complete drainage. Rarely, the right internal jugular vein may be used to insert a single lumen or a two-stage bicaval cannula(3). The arterial cannula is a short 17-21 Fr cannula inserted in the femoral artery with the tip in the common iliac artery. Another option is to use an end-to-side Dacron graft to the right subclavian or axillary artery(4). pVA ECMO can be implanted outside operating theatre (at the bedside, in the catheterization laboratory or even out of hospital), this is an advantage, especially in case of patient's haemodynamic instability.

REASONS FOR VA ECMO

VA ECMO is a strategy to gain time and stability, it does not solve the condition causing the cardiovascular shock. The essence of VA ECMO strategy is summarised in the term bridge(5):

- **Bridge to decision:** VA ECMO is used to determine the reversibility of the organ damage after the cardiovascular shock event or to decide the next action;
- **Bridge to recovery:** VA ECMO is used to give time to the cardiovascular system to recover its function by treating the underlying condition (antibiotics in septic cardiomyopathy, myocardial revascularization in case of acute coronary syndrome, heparin in case of massive pulmonary embolism, etc.) or just supporting the heart (levosimendan infusion in case of myocarditis, myocardial stunning, etc);
- **Bridge to transplantation or durable mechanical circulatory support:** VA ECMO is used to achieve a temporary stability to allow HT or LVAD/TAH implantation.

Typical indications(6) for VA ECMO implantation are:

- Cardiac arrest (extracorporeal cardiopulmonary resuscitation);

- Cardiogenic shock due to:
    - Acute myocardial infarction;
    - Acute myocarditis;
    - Progression of cardiomyopathy;
    - Acute right ventricular (RV) failure due to pulmonary embolism;
    - Progression of RV failure due to pulmonary disease;
    - Progression of congenital heart disease;
    - Primary graft failure and acute allograft rejection after HT;
    - Overdose of cardiotoxic drugs;
    - Septic cardiomyopathy;
    - Refractory ventricular tachycardia;
    - Failure to wean off cardiopulmonary bypass;
- Circulatory support for high-risk invasive procedures.

VA ECMO contraindications can be divided into absolute and relative(6). Absolute contraindications are severe irreversible noncardiac organ failure threatening survival (e.g., severe brain injury or metastatic cancer), irreversible cardiac failure in patients who are not candidate for HT or durable MCS device, severe aortic regurgitation, and aortic dissection.
Relative contraindications to VA ECMO are: severe coagulopathy or contraindication to anticoagulation, limited vascular access (extreme obesity, amputated limbs, severe peripheral artery disease).

MORBIDITY IN VA ECMO

VA ECMO is a powerful resource to save lives, however, its use is associated with severe, life-threatening complications which influence the patients' prognosis strongly. The major VA ECMO complications are reported below.

LIMB ISCHAEMIA AND OTHER VASCULAR COMPLICATIONS

Limb ischemia is a serious complication of femoral arterial cannulation (pVA ECMO). It can be due to anatomical reasons (size, stenosis, calcifications,

previous surgeries, obesity) and patient's clinical conditions (low-flow states, vasoconstriction, etc.)(7). The incidence of limb ischaemia varies from 13% to 25%(8). The use of anterograde perfusion catheter reduces the risk of limb ischaemia because it allows the blood to flow to the distal extremity(9). Compartment syndrome is a severe complication of limb ischaemia which may require emergency fasciotomy or limb amputation.
Other possible vascular complications of pVA ECMO support are formation of arteriovenous fistula, pseudoaneurysm, arterial dissection, hematoma(10).

HARLEQUIN SYNDROME

It is a complication of pVA ECMO. In pVA ECMO a retrograde oxygenated blood flow perfuses mainly lower extremities and abdominal viscera, especially when the left ventricle contractility recovers sufficiently. Indeed, the blood oxygenated by the lungs is pumped by the heart towards the upper part of the body (head, upper limbs, heart). As a result, a differential hypoxia may be determined, and less oxygenated blood goes to the brain and the heart. To treat this complication, it must be optimised lung ventilation and when it is not sufficient, another VA ECMO cannula has to be inserted in the RA to deliver some oxygenated blood (VA-V access)(11).

THROMBOEMBOLIC COMPLICATIONS

Thromboembolic complications are consequences of blood-VA ECMO circuit surface interaction. Thanks to the improvement of biocompatible materials, thromboembolic complications have reduced.
Micro thrombosis of the oxygenator is the most frequent thromboembolic complication(12). A rare condition which may happen in case of VA ECMO is heparin-induced thrombotic thrombocytopenia. This condition is characterised by the formation of multiple arterial thrombi and severe thrombocytopenia. In this case a switch from heparin to argatroban is recommended(13).

BLEEDING

Bleeding is a frequent complication of VA ECMO support and it is not only related to vessel injury or sternotomy (cVA ECMO), but it is also due to systemic anticoagulation and consumption coagulopathy. Systemic anticoagulation is used to prevent VA ECMO circuit thrombosis and thromboembolism(8). During anticoagulation, the routine intensive care procedures may become a potential trigger for bleeding (bronchoscopy, urinary catheter insertion, etc.). Further, patients on VA ECMO support can develop some levels of disseminated intravascular coagulation (DIC) and acquired von Willebrand disease(14). This is due to the contact of the blood with the VA ECMO circuit surface which activates the coagulation cascade causing consumption coagulopathy(15). The rate of bleeding is around 10-30% in the postcardiotomy patients(16,17).

NEUROLOGICAL COMPLICATIONS

Neurological complications in VA ECMO patients are central nervous system haemorrhage (~2%), infarction (~4%)(18), and seizures with cerebral oedema (~2%)(18). Respectively 1 in 4 cerebral ischemia patients and 1 in 10 cerebral haemorrhage patients survives(19). Pre-ECMO factors may increase the risk of intracranial haemorrhage. For example, cardiac arrest causing anoxic brain injury or infarction can predispose to intracranial haemorrhagic transformation(18,20,21). Sepsis and influenza are others predisposing factors reported in several studies(22,23). Platelet dysfunction and consumption due to renal failure and renal replacement therapy increase the risk of intracranial haemorrhage(21,24). The cause of ischaemic stroke is multifactorial: hypoperfusion due to haemodynamic instability or thromboembolic events (clot or bubbles may be infused into the arterial VA ECMO circuit)(25). Ischemic stroke is frequent in extracorporeal cardiopulmonary resuscitation (7% of resuscitations)(26).

INFECTIONS

Immunocompromised status, VA ECMO cannulas, vascular catheters, invasive mechanical ventilation, surgical wounds, continuous renal replacement therapy, and patients' comorbidities make infections frequent complications in VA ECMO patients(27) (prevalence between 9 and 65%)(27-30). Several studies reported a strong association between a prolonged duration of VA ECMO support and infections(27,31). Infections are typically in the respiratory and urinary systems, often associated with sepsis. Less common but clinically relevant are cannula access infections(28,31).

Severe infections are associated with a significantly increased mortality and morbidity(30,31). Furthermore, the diagnosis of infection during VA ECMO support is challenging, since its signs may be masked by the effects of VA ECMO support on inflammatory and immune systems(32).

Another important point is the effect of VA ECMO circuit on antibiotics: VA ECMO membrane seems to sequestrate and altering the pharmacokinetic of several drugs, and VA ECMO circuit increases the volume of distribution(33,34).

IMMUNOLOGIC ALTERATIONS

VA-ECMO has been suspected to cause an alteration in the release of pro-inflammatory cytokines and in the function of T-cells, neutrophils, dendritic cells (DC), monocytes and myeloid suppressive cells(35).

In their research study Frerou et al. found an increased number of immature neutrophils during the first 24 hours post VA ECMO initiation(36). Immature neutrophils are characterised by impaired phagocytosis and bactericidal activities(37). Furthermore, Frerou et al. detected a transitory reduction in the HLA-DR expression on DC and a reduction in the circulating mDC CD141pos, which are the main subset of dendritic cells involved in infection response(38).

Another important finding was that VA ECMO induced both myeloid suppressive cells (MDSC) expansion and T-cell dysfunction. MDSC are cells which supress CD4pos and CD8pos T-cell activation and function and can induce T-cell apoptosis(39,40). This impairs the immune system function significantly.

Focusing on cytokines expression, Frerou et al. found an increase in the IL-10 (immunosuppressive interleukin)(41) and IL-6, IL-8 and TNF-α (pro-inflammatory cytokines)(35) levels on VA ECMO patients, but a reduction in IL-7 levels. IL-7 is responsible for T-cells function(41).

LV DISTENSION AND THROMBOSIS; THE NECESSITY OF VENTING

Other problems are LV distension, intraventricular blood stasis, thrombosis, and pulmonary oedema(42). According to the different site of cannulation, the VA ECMO flow may be additive or competitive to the native cardiac stroke volume (SV). As pVA ECMO (femoral-femoral cannulation) generates a retrograde blood flow into the aorta, and consequently it increases the afterload, it may cause an important reduction in the LV SV. By contrast, axillary cannulation (for the arterial return cannula) or central VA ECMO cannulation generate a flow in the same direction of LV SV, having a lower impact on LV afterload(43,44). These are the reasons why LV distension is more likely in case of pVA ECMO. Echocardiography is used in intensive care unit (ICU) to assess aortic valve opening, LV and RV dimensions, valves and to exclude thrombosis(45). In VA ECMO patients it is important to avoid an excessive increase in LV afterload, however, that is secondary to guarantee an optimal systemic perfusion. Furthermore, it is important to consider that an increased LV afterload is responsible for LV wall stress and oxygen demand which can be potentially harmful for the heart(46). In order to avoid LV distension and its complications, LV venting system is applied. There are several venting strategies which can be used(47). Inotropes, vasodilators, atrial septostomy, left atrium (LA) venting or surgical LV venting, Impella, intra-aortic balloon pump (IABP). These strategies may be classified into those increasing the inotropism of the heart, those reducing the afterload and those decreasing the preload. Inotropes increase cardiac inotropism favouring the increase in the SV and the opening of the aortic valve, however they cause a rise in the myocardial oxygen consumption which may have detrimental effect especially in case of myocardial infarction.

IABP and vasodilators are the strategies used to reduce the afterload. IABP has been demonstrated to be effective in reducing pulmonary artery occlusion pressure(48), and pulmonary oedema in patients on VA ECMO(49). However, its positive effect on the survival of VA ECMO patients is controversial(50,51). The most of the strategies acting on the preload has the drawback of the aortic valve closure, which may cause the thrombosis of the ascending aorta. Atrial septostomy creates a left-to-right shunt which decompresses the LA and indirectly the LV by the VA ECMO drainage cannula(52). This decompression is the consequence of a reduction in the preload.

LA venting by cannula connected to VA ECMO circuit is similar to atrial septostomy(53,54). In this case, the blood is aspirated by the LA venting cannula. Both atrial septostomy and LA venting cannula may be responsible for a persistent interatrial shunting after VA ECMO removal. Actually, a drainage cannula can be added in almost every part of the heart (pulmonary artery, pulmonary veins (55), LA, LV) in order to unload the LV by reducing the preload. LV venting by a cannula allows a direct LV unloading. The LV venting cannula is placed into the LV apex surgically and then the cannula is connected to the VA ECMO circuit(56). The cannula needs to be removed surgically and generally this option is used in case of VA ECMO as a bridge to HT or LVAD. Percutaneous LV venting by a transaortic pigtail catheter connected to VA ECMO circuit is another option to unload the LV(57). In this case the pigtail catheter is inserted percutaneously into the LV going through the aortic valve. The degree of unload depends on the pigtail size. Impella is a sort of combination between a LVAD and the aspirating transaortic pigtail catheter. It is an axial flow pump which may be inserted surgically (Impella 5, Impella 5.5) or percutaneously (Impella CP, Impella 2.5) across the aortic valve. By pumping the blood from the LV to the aorta, Impella decreases LV distension, blood stasis, and pulmonary congestion, and increases systemic blood flow. The combination of Impella and VA ECMO (so called ECMELLA or ECPELLA) has been proven to be associated with a decreased in mortality even though it is associated with a rise in the complications rate(58).Particularly interesting are the findings of the beneficial effect of LV unloading by Impella on infarct size on animal model(59). At present, among

intensivists, there is a consensus in the use of venting on selected patients undergoing VA ECMO support. Even though randomized control trials are missing, observational studies have shown a reduction in mortality in VA ECMO patients appropriately vented(60).

MORTALITY PREDICTORS AND PROGNOSTIC SCORES

According to Extracoroporeal Life Support Organisation (ELSO) registry report (April 2021) in 2020 there were 32307 cardiac VA ECMO runs(61). Only 44% of these VA ECMO patients survived to discharge or transfer. 29% of the 10115 extracorporeal pulmonary resuscitation (ECPR) patients survived. Considering these figures, it appears evident that VA ECMO patients are critically ill patients who are exposed to a high risk of complications and mortality. Furthermore, VA ECMO is particularly demanding in terms of resource utilization and hospital costs. So it is particularly important to appropriately select patients who have better prognosis in order to avoid any suffering prolongation and to use hospital resources effectively.

Several studies (the most retrospective single centre analysis) have been conducted to find mortality predictors in patients supported by VA ECMO. A retrospective analysis from Karolinska Hospital on 181 refractory cardiogenic shock patients supported by VA ECMO found on univariate regression analysis that age, ischemic heart disease (IHD), multiorgan failure, left ventricular ejection fraction (EF), mean arterial pressure (MAP), pre ECMO arterial lactate, international normalized ratio, and number of inotropes and vasopressors were significantly associated to 90-day mortality. Arterial lactate (odds ratio [OR] per unit: 1.14; 95% confidence interval [CI]: 1.06 to 1.23; $p < 0.001$), number of ino-vasopressors (OR per agent: 1.58; 95% CI: 1.13 to 2.21; $p = 0.008$), and IHD (OR: 2.90; 95% CI: 1.31 to 6.39; $p = 0.008$) were predictors of 90-day mortality on multivariable logistic regression analysis(62). In another study focusing on VA ECMO for cardiac arrest patients, Torre et al. found that lactate peak level ≥ 8.0mmol/L and time from cardiac arrest to VA ECMO ≥ 30min were significantly associated to 30-day mortality(63). From these studies we can infer that the timing of VA ECMO implantation is crucial, since a more profound status of cardiogenic

shock (hyperlactatemia and escalation of ino-vasopressors) is associated with a worse prognosis.

In their research study, Huang et al. found that pre-implantation hypoalbuminemia and shorter VA ECMO duration were strongly associated with mortality (multivariate analysis by logistic regression)(64). The authors hypothesised that hypoalbuminemia was a sign of hepatic synthetic dysfunction due to an advanced heart failure, instead a shorter VA ECMO duration was due to an early death on MCS. Stroke, limb ischemia, preimplantation creatinine > 100mmol/L were other mortality predictors found on univariate logistic regression analysis. Focusing on VA ECMO duration, according to an analysis of ELSO registry (2002 to 2012)(65), VA ECMO survival rate per day increased until day 4 of a VA ECMO run (multivariate regression analysis with adjustment for the covariates available OR 1.53 95% CI 1.37–1.71, $p < 0.001$), then progressively decreased until day 12 (OR 0.86, 95% CI 0.81-0.91, $p < 0.001$). After day 12 of VA ECMO run, there was no significant change. The significant covariates were diagnostic category, such as the cause of shock, ($p = 0.008$), age ($p \leq 0.001$), pH ($p = 0.005$), MAP ($p < 0.001$), and time from endotracheal intubation to initiation of VA ECMO ($p = 0.049$). Better survival was in myocarditis (64.4%) and post-heart transplantation (57.1%). Conversely poorer survival was in the categories: other medical cardiac disease (38.4%) and other surgical cardiac disease (35.9%). Another factor influencing patients' prognosis is age. In a retrospective single centre study on 355 VA-ECMO patients, Salna and colleagues(66) found that age over 72 years was an independent predictor of in-hospital mortality (OR 2.71 CI 1.22-6.00 $p = 0.014$) together with baseline lactate level (OR 1.09 CI 1.02-1.15 $p = 0.011$), coronary artery disease (OR 1.67 CI 0.94-2.98 $p = 0.081$), acute decompensate heart failure (OR 4.27 CI 1.69-9.95 $p = 0.001$). From this short reviews of studies, it is possible to understand that several factors may influence VA ECMO patients' outcome. In order to clarify this complex scenario, and help the clinician to appropriately identify the patients with more survival chances, predictor scores have been developed. These scores are based on pre-ECMO parameters.

One of the most important prognostic score for VA ECMO patients is SAVE score. It was developed from the international ELSO registry, involving 3846

patients affected by cardiogenic shock treated with VA ECMO. Multivariable modelling performed on these patients identified chronic renal failure, longer duration of mechanical ventilation prior VA ECMO, pre-ECMO acute organ dysfunction and cardiac arrest, congenital heart disease, lower pulse pressure, and lower serum bicarbonate as risk factors associated with hospital mortality. On the basis of these results, the 12 variables considered in the SAVE score were identified. Scores range from -35 to 17, and they are divided in 5 risk classes(67) (Table 1). A SAVE score of zero is approximately equivalent to 50% survival. The SAVE-score's area under the receiver operating characteristics curve (AUROC) was 0.68 (95%CI 0.64-0.71). External validation of the SAVE-score in an Australian population of 161 patients showed excellent discrimination with AUROC 0.90 (95% CI 0.85-0.95). However, the limitations of the SAVE score are the following: it comprises high and low volumes VA ECMO centres, and complete physiologic data was available in only 23% of patients. So when it was tested on a high volume North American VA ECMO centre, the SAVE score performed more poorly (AUROC curve 0.77)(68). It is worth of consideration that the population of VA ECMO patients is particularly heterogenous (cause of cardiogenic shock, age, sex, comorbidities, etc.), thus it is reasonable to develop specific risk scores for particular subgroups of VA ECMO patients. Consequently, REMEMBER score and ENCOURAGE score have been developed.
REMEMBER score is a predictive score for in-hospital mortality to be applied on patients who suffered from cardiogenic shock after isolated coronary artery bypass graft (CABG). This score was developed from a cohort of 166 CABG patients supported by VA ECMO at the Beijing Anzhen Hospital from 2004 to 2017(69). The REMEMBER score divides this specific population of patients in 4 risk classes on the basis of six pre-ECMO parameters: older age, left main coronary artery disease, inotropic score > 75, CK-MB > 130IU/L, serum creatinine > 150umol/L, and platelet count < 100 × 109/L (Table 1). The AUROC for the REMEMBER score is 0.85 (95% CI 0.79-0.91). Limitations of this score are the following: a single-centre design, a small sample size, a long recruitment period (the standards of care may be changed), the use of a non-fixed-time mortality outcome, the lack of external validation. ENCOURAGE score is another

mortality risk score for patients who underwent VA ECMO support in consequence of cardiogenic shock following a myocardial infarction(70). This risk score was developed from 138 patients admitted to two French ICUs from 2008 to 2013. It was constructed from seven pre-VA ECMO parameters identified by multivariable logistic regression analyses (age >60, female sex, body mass index (BMI) >25 kg/m2, Glasgow coma score <6, creatinine >150µmol/L, lactate levels, and prothrombin activity <50 %). The AUROC of this score is 0.84 (95 % CI 0.77-0.91) (Table 1).

Table 1. Pre-ECMO prognostic scores

SAVE score			**REMEMBER score**			**ENCOURAGE score**	
Hospital survival by risk class			Predicted hospital mortality at each score level.			Six months after VA ECMO, survival probabilities	
Score	**Risk class**	**Survival (%)**	**Score**	**Risk class**	**Mortality (%)**	**Risk class**	**Survival (%)**
>5	I	75	0–13	I	13%	0-12	80
1–5	II	58	14–19	II	55%	13-18	58
24 to 0	III	42	20–25	III	70%	19-22	25
29 to 25	IV	30	> 25	IV	94%	23-27	20
≤-10	V	18				≥28	7

SAVE score, REMEMBER score and ENCOURAGE score are scoring systems based on pre-ECMO implantation parameters. However, VA ECMO patients are extremely unstable, so their condition and mortality risk may change during MCS. To address this problem, the PREDICT score has been developed (71). This score was derived from a cohort of 205 VA ECMO patients (51% received VA ECMO because of resuscitation and 43% due to severe shock). Two prediction models at 6 and 12-hour have been designed based on lactate level, pH and standard

bicarbonate concentration. On external validation, the 6-hour and 12-hour PREDICT VA-ECMO scores showed a AUCs of 0.718 and 0.735 respectively.

VA ECMO WEANING STRATEGIES

Premature VA ECMO removal (weaning) may result in hemodynamic/respiratory deterioration and put at risk the patient's life. However, unnecessarily prolonging VA ECMO support may be responsible for patient's complications and death. In consequence of that, identifying the correct time window for VA ECMO weaning is essential for the patient's safety.

There are several VA ECMO weaning protocols reported in scientific literature (Table 2). Although each protocol has its own peculiar characteristics, the most of them combine haemodynamic, echocardiographic and respiratory parameters. The combination of these parameters reflects the complexity and variety of conditions which can be treated with VA ECMO support. "Bypassing heart and lungs" VA ECMO gives a complete support to the whole body, providing both oxygenation and circulatory support. Indeed, VA ECMO can be used in case of LV failure, RV failure, distributive shock, arrhythmic storm, and in case of respiratory failure (VV ECMO is more indicated in the last case)(6).

Table 2. VA ECMO weaning protocol cited in Scientific literature

Author or Name of the Protocol	VA ECMO weaning protocol
Aziz TA, et al, 2010(72)	VA ECMO removal is considered if: - VA ECMO flow < 2.5L/min - Cardiac index (CI) >2.4L/min/m^2, - Mean arterial pressure (MAP) > 60mmHg, - Pulmonary arterial wedge pressure (PAWP) < 18mmHg, - Central venous pressure (CVP) <18 mmHg.
Aissaoui, et al, 2011(73)	A VA ECMO weaning trial is considered when the patient is considered hemodynamically stable: MAP

	> 60mmHg while receiving no or low-dose vasoactive agents and a pulsatile arterial waveform maintained for at least 24h, and when pulmonary blood oxygenation is not compromised. The VA ECMO flow is decreased to 66% for 10-15min, then to 33% and/or to a minimum of 1-1.5 L/min for 10-15min. VA ECMO removal is considered if at VA ECMO flow of 1-1.5L/min - MAP ≥ 60mmHg - LV EF ≥ 20-25% and - Left ventricle outflow tract velocity time integral (LVOT VTI) ≥10cm under minimal VA ECMO support
Cavarocchi NC, et al, 2013(74)	VA ECMO trial consists of the following phases: 1) Baseline assessment of RV and LV function with full VA ECMO flow. 2) Reduction of VA ECMO flow from full to half flow gradually in increments of 0.5L/min and assessment of LV and RV function by transoesophageal echocardiogram (TOE). If distention occurs, it is necessary to return to full flow and stop VA ECMO weaning trial. 3) Volume load (10mL/kg) over 20min, with half VA ECMO flow, and assessment of RV and LV function by TOE over at least 1 hour. 4) Administration of inotrope (dobutamine and/or milrinone), reduction of VA ECMO flow to minimum

	(1-1.5L/min), and assessment of LV and RV function at least for 1h. After VA ECMO weaning trial: - If biventricular failure persists, consider TAH placement end-of-life discussion. - If one of the ventricle recovers, but the other does not consider VAD (LVAD for LV dysfunction, RVAD for RV dysfunction). - If both LV and RV functions are recovered, consider VA ECMO decannulation.
Pappalardo F, et al, 2015(75)	Stepwise reduction of pump flow (0.5 L every 6-24h), if the Inotropic Score is ≤ 10 under serial echocardiographic assessment. In case the VA ECMO flow < 2L/min, after a short period of observation, circuit is clamped and VA ECMO cannulas are removed.
Huang KC et al, 2018(76)	VA-ECMO flow is reduced to 0.5L/min and maintained for a 5min observation period. If the patient tolerates that, the cardiovascular surgeon clamps both arterial and venous cannulas. A new 3-min observation period off VA-ECMO support is waited. If the patient tolerates this period, the VA ECMO cannulas are removed. Any hemodynamic instability aborts the VA ECMO removal.
Ling L, et al, 2018(77)	1) VA ECMO arterial flow probe direction is reversed and the pump speed is reduced to achieve a retrograde flow of 0.5 to 1.0L/min. 2) The sweep gas flow is turned off. 3) Haemodynamic and echocardiographic assessments are performed.

	After 1h, the patient is considered ready for VA ECMO removal if: - MAP ≥ 60mmHg, - vasopressor inotropic equivalent is less than 30 - a base deficit is less than 7 - Fractional inspiratory oxygen (FiO_2) requirement ≤60% with arterial saturation ≥90%.
Mazankowski Alberta Heart Institute VA-ECMO Liberation Protocol(78)	VA ECMO removal is considered if: - physiological stability is achieved: - reversal of end-organ dysfunction, - acceptable hemodynamic parameters. VA ECMO flows are reduced by increment of 0.5 L/min in several hour, to a minimum of 2 L/min - Mixed venous oxygen saturation (SvO_2) targets of 60-70% should be maintained, - lactate should remain < 2mmol/L, - Partial pressure oxygen (PaO_2) should remain 100-190mmHg, Target ventilator settings during weaning are: 1) FiO_2 ≤ 50%; 2) Plateau pressure < 25cmH_20; and driving pressure < 15cmH_20; 3) Positive end-expiratory pressure (PEEP) ≤ 12cmH_20 TOE is performed during VA ECMO weaning to evaluate cardiac performance and valvular function.

	Inotropes and vasoactive agents are adjusted to optimize haemodynamics during the TOE.
Thomas M, et al, 2020(79)	VA ECMO removal is considered if: - VA ECMO flow max 2L/min; - PEEP < 10mbar; - FiO_2 < 50%; - LV EF ≥ moderately impair; - No high grade of valve dysfunction; - Stable lung function; Defined regimen of nitric oxide (NO)/catecholamines.
Tohme J, et al, 2021(80)	VA ECMO removal is considered if: - haemodynamically stable patient; - MAP ≥65mmHg with low doses of vasoactive agents; - pulsatile arterial waveform; - LV EF >25%-30%; - LVOT VTI >12cm, - PaO_2/FiO_2 ratio >200, with FiO_2 delivered by the extracorporeal circuit ≤30% and that delivered by the ventilator circuit ≤60%.

At the basis of every VA ECMO weaning protocol there is a gradual reduction of VA ECMO support to the lowest possible flow (1±0.5 L/min) and the evaluation of haemodynamic, echocardiographic and respiratory parameters. Some protocols consider the possibility of clamping the VA ECMO circuit for some minutes in order to assess the patient's body reaction to the temporary complete suspension of MCS(76,77). However, such a manoeuvre threatens the VA ECMO circuit integrity(81). In the majority of VA ECMO weaning protocols, a Mean arterial pressure (MAP) ≥ 60mmHg at the lowest flow of VA ECMO support is necessary to consider VA ECMO decannulation. ELSO guidelines recommend at least 24h

of pulsatility on invasive blood pressure monitoring(81). In fact, pulsatility is an index of cardiac contractility(75) Some protocols advise to have a Pulmonary arterial wedge pressure (PAWP) < 18mmHg, and central venous pressure (CVP) <18mmHg at the lowest flow of support(72). Respiratory and blood gases values are also important parameters to be considered when a patient is liberated from VA ECMO (which is also a respiratory support). A Partial pressure oxygen/Fraction inspiratory oxygen (PaO_2/ FiO_2) ratio >200, with FiO_2 delivered by the extracorporeal circuit ≤30% and that delivered by the ventilator circuit ≤60% are necessary to be reached before VA ECMO weaning. Some centres to check whether a patients can be weaned from VA ECMO performed a pump-controlled retrograde trial off which consists in reducing to the minimum the VA ECMO support and turning to zero the sweep gas so that the only oxygenation of the blood is provided by the patient's lungs(77).

Another important point to be considered at the moment of VA ECMO weaning is the amount of inotropes and vasopressors administered. The lower is the amount of ino- vasopressor support used the higher is the probability to successfully wean the patient from VA ECMO(75).

Echocardiography is a valuable tool in intensive care because it allows to assess the cardiac recovery and the reaction of the heart to the haemodynamic changes due to VA ECMO flow variation(45).

The most used echocardiographic parameters to guide VA ECMO weaning are left ventricular outflow tract velocity time integral (LVOT VTI) and EF, which reflect the cardiac output (CO) and the LV systolic function respectively. Some protocols recommend to assess right ventricular outflow tract (RVOT) VTI too. However, ELSO guidelines highlights that RV is always unloaded to some degree, so that the real RV function and output cannot be evaluated until the VA ECMO removal(74,81).

According to ELSO guidelines, an LVOT VTI of at least 10-12cm and an EF of 20-25% are sufficient to consider VA ECMO weaning. These values are taken from Aissaoui et al's experience, who found that a LVOT VTI of 12cm, LV EF of 20-25% and lateral mitral annular s' tissue Doppler imaging (TDI) > 6cm/sec were predictors of successful weaning from VA ECMO (Table 3)(73).

Many efforts have been made to find other, more effective and standardised echocardiographic parameters to guide VA ECMO weaning (Table 3). In a retrospective study on 46 patients liberated from VA ECMO, Huang et al(76) found that RV EF ≤ 24.6% was associated with poor prognosis within 30 days (hazard ratio, 15.86; 95% CI 3.56-70.73; p < 0.001). RV EF was calculated by 3D echocardiogram, which is a time consuming technique needing a high level of expertise, a very good image qualities. These characteristics make RV EF difficult to be used in intensive care where time is essential, echocardiography is generally performed by intensivists and the quality of the images is generally poor. In another study, Sawada et al(82) found that the values of fractional shortening, corrected left ventricular ejection time (LVETc), LVOT VTI, and LVETc divided by PAWP were higher in the VA ECMO weaned patients who survived at least 30 days from VA ECMO removal. On multivariable analysis, LVETc⁄PAWP (cut off 15.9) was a significant independent predictor of successful weaning (LVETc⁄PAWP, OR 0.82, 95% CI 0.71–0.94, *P* = 0.005). A possible explanation for LVETc/PAWP as predictor of weaning is that LVETc is correlated with CO(83-85) and PAWP is correlated with pulmonary congestion(86). So, the higher is the LVETc⁄PAWP the better is the CO and the lower is pulmonary congestion, which is the best option to wean a patient from VA ECMO. Another promising parameter to guide VA ECMO weaning is the ratio between tricuspid annular S' and right ventricular systolic pressure (RV S'/ RVSP)(87). This is a measure of the coupling between RV contractile function (tricuspid annular S') and pulmonary circulation (RVSP calculated by adding estimated right atrial pressure to tricuspid regurgitation jet maximum velocity). According to their analysis, Kim et al found that, at full VA ECMO flow, RV free wall longitudinal strain, RVSP, tricuspid annular S'/RVSP, tricuspid annular plane systolic excursion (TAPSE)/RVSP and the absolute value of RV free wall longitudinal strain/RVSP were significantly different between patients successfully weaned from VA ECMO support and patients who failed VA ECMO weaning. Conversely, the conventional parameters used to guide VA ECMO weaning (LV EF, LVOT VTI, lateral mitral annular S') failed to show any significant difference. On ROC analysis, the tricuspid annular S'/RVSP > 0.33

showed a better performance than conventional echocardiographic parameters to predict successful VA ECMO weaning (AUC = 0.692, 95% CI: 0.574-0.809, p = 0.005).

Table 3. Studies reporting echocardiographic predictors of successful weaning from VA ECMO.

Study/ Authors	Population	Weaning Ratio	Categories	Parameters	Cut off
Aissaoui et al, 2011(73)	51	20/51	TTE	LVOT VTI LV EF Lateral S' TDI	> 10 cm > 20-25% > 6 cm/sec
Cavarocchi et al, 2013(74)	21	6/21	TOE	LV and RV function	qualitative
Huang et al, 2018(76)	46	38/46	3D-TTE	RV EF	> 24.6%
Sawada et al., 2020(82)	50	24/50	TTE / Swan Ganz catheter	LVETc⁄PAWP	>15.9
Kim et al, 2021(87)	79	50/79	TTE	tricuspid annular S'/RVSP	> 0.33

SPECKLE TRACKING

Speckle tracking is an imaging technique that allows a quantitative evaluation of global and regional myocardial function. This technique is based on the analysis of the spatial dislocation of speckles (spots generated by the interaction between the ultrasound beam and myocardial fibres) on routine 2D echocardiographic

images(88). Currently speckle tracking is still a post hoc analysis, performed on the achieved 2D echocardiographic images through dedicated software. Strain is a measurement of the degree of deformation of the analysed myocardial portion in relation to its initial dimensions. The higher is the contractility of the myocardium, the more negative is the strain value. Longitudinal strain represents myocardial deformation directed from the base to the apex. Global longitudinal strain has been validated as a quantitative index for global LV function(89). The same measurement can be applied to the speckle-tracking echocardiographic analysis of longitudinal myocardial deformation of the RV. Radial strain represents radially directed myocardial deformation. Circumferential strain represents LV myocardial fibers shortening along the circular perimeter observed on a short-axis view. It is possible to obtain a global circumferential and radial strain value likewise for longitudinal strain(90). Advantages of speckle tracking measurement technique are: the ability to discriminate active myocardial contraction from passive displacement of dysfunctional region of myocardium (tethering), the independency from the angulation of the ultrasound (US) beam and the use of Lagrangian strain instead of natural strain. In Lagrangian strain a single reference length is taken as landmark and all the others measurements are compared against it. In contrast, natural strain employs a reference length which changes over time, as the object deforms(91). Disadvantages of speckle tracking technique are intervendor variability, necessity of high quality of echocardiographic images, instability of speckle tracking patterns. Intervendor variability is principally related to post-processing algorithms. Instability of speckle tracking patterns depends in part on the displacement out of plan of the speckles and in part on the physiological changes of living structures and changes on the interrogation angle between tissue and ultrasound beam.

STUDY RATIONALE

During a VA ECMO weaning procedure echocardiographic measurements (including LVOT VTI, EF, measures of ventricular cavity size, etc.) and haemodynamic measurements (including MAP, HR, etc.) are assessed as the loading conditions of the heart are altered (VA ECMO flow change)(73).

Echocardiographic measurements are made either via transthoracic or transoesophageal echocardiography (the decision of one imaging over the other will depend upon local expertise and experience, availability of specialist equipment and patient factors such as recent cardiac surgery, which will make transthoracic imaging challenging). The aim of this pilot study was to identify echocardiographic parameters able to discriminate patients who could tolerate discontinuation of VA ECMO support and patients who could not. Speckle tracking is an image analysis technique which is able to quantify the active contraction of myocardium, with low inter-observer variability(88,90). Speckle tracking is able to detect small changes in active deformation of the heart (strain) in both systole and diastole, and it appears to be more sensitive than traditional echocardiographic and Doppler measurements in detecting patients with impaired myocardial function. Furthermore, previous studies on different loading conditions have proven a variation of strain values consequently to the variations of loading conditions. In other words, speckle tracking appears to be able to assess the myocardial contractile reserve(92,93). Speckle tracking analysis requires images of sufficient quality (good myocardial definition) and a frame rate between 60 to 80 frames per second(91).

In both the institutions involved in the research study, standard-of-care is for all VA ECMO weaning procedures to be guided by TOE. TOE exams acquire high quality images which are well suited for post hoc analysis of strain by speckle tracking. Modification of the frame rate during acquisition of images has negligible impact on the quality of images, does not raise costs and does not delay the procedure. Post hoc strain analysis introduces no risks beyond those related to a standard TOE examination. Furthermore, since it is a post-hoc analysis, speckle tracking does not influence the VA ECMO weaning procedure, which is conducted on the basis of conventional echocardiographic and haemodynamic findings.

A prospective analysis of clinical outcomes after VA ECMO removal could provide information about the best strategy of VA ECMO weaning procedure.

The risks related to TOE are low: the reported incidence of major morbidity related to diagnostic TOE is 0.2% (1 in 500), with the incidence of oesophageal perforation and the incidence of major bleeding both <0.01%(94).

AIM OF THE STUDY

By this pilot study we wanted to investigate whether parameters obtained with speckle tracking analysis immediately before VA ECMO liberation could improve the identification of VA ECMO patients who may undergo a safe weaning from VA ECMO reducing the number of clinical adverse outcomes in the first 3 months after liberation. The protocol was approved by both the IRB of the Royal Brompton and Harefield NHS Foundation Trust, and the Ethical Committee of Azienda Ospedaliera Universitaria Integrata di Verona. The protocol was registered in ClinicalTrial.gov

PRIMARY OBJECTIVE

Purpose: to assess whether strain obtained through speckle tracking (LV longitudinal and circumferential strain, RV free wall longitudinal strain) could give additional information in identifying patients who develop adverse outcomes within 3 (± 1) months post successfully VA ECMO removal (liberation not for palliation).

In order to achieve this purpose, strain analysis was performed on the recorded images of the TOE performed during the last VA ECMO weaning trial procedure of patients defined ready for VA ECMO liberation. VA ECMO liberation was based according to conventional echocardiographic-haemodynamic parameters and clinical judgment during patients' VA ECMO weaning procedure. It was evaluated whether the population experiencing the composite outcome of interest (death for any reasons within 3 (± 1) months from VA ECMO liberation, hospital admission for a new episode of cardiogenic shock or heart failure within 3 (± 1) months from VA ECMO liberation, need for new mechanical circulatory support or new necessity of high doses of ino-vasopressors within 3 (± 1) months from VA ECMO liberation) and the population not experiencing this composite

outcome had different values of strain (LV longitudinal and circumferential, and RV free wall longitudinal strain) during the weaning procedure.

SECONDARY OBJECTIVES

Purpose:

- to assess the reliability of conventional echocardiographic parameters (id est EF, TAPSE, RV fractional area change (FAC), LVOT VTI, etc.) in the identification of patients who develop adverse clinical outcomes within 3 (± 1) months post successfully VA ECMO removal (liberation not for palliation).
- to assess whether strain obtained through speckle tracking (LV longitudinal and circumferential strain, RV free wall longitudinal strain) could give additional information in identifying patients who develop specifically adverse cardiac outcomes within 3 (± 1) months post successfully VA ECMO removal (liberation not for palliation).

POPULATION

The population consisted of consecutive patients receiving VA ECMO support for cardiovascular shock who underwent an echocardiogram as part of the weaning process and a decision was made to attempt weaning (liberation) from VA ECMO. The decision to wean the patient from VA ECMO was made by the attending clinician who was aware of all echocardiographic measurements with the exception of the strain results (this is the standard-of-care at the patients recruiting institutions). The study investigator who performed strain analysis was not involved in the decision to wean the patient from VA ECMO.

All patients receiving VA ECMO in ICU who underwent an echocardiogram as part of the weaning procedure were screened. Patients were considered enrolled if a decision was made to attempt liberation from VA ECMO following the weaning trial. When a screened patient underwent a weaning trial and was deemed unsuitable for attempting liberation from VA ECMO, their recruitment was deferred pending further weaning trial procedures. If the patient was subsequently

deemed suitable for attempting liberation from VA ECMO - following a subsequent weaning trial procedure, they were enrolled.
Enrolled patients dropped out from the study in case the doctors taking care of the patient decided:

- That the patient underwent heart transplant (HT) or VAD implantation without VA ECMO liberation;
- To combine VA ECMO with Impella support;
- To switch from VA ECMO to Impella support;
- Patient's death before VA ECMO weaning procedure/VA ECMO liberation.

INCLUSION CRITERIA

- ≥18 years.
- Receiving VA ECMO mechanical circulatory support for cardiovascular shock and planning to undergo an echocardiogram as part of a weaning trial procedure.
- Informed consent for study participation provided by the patient (where able to provide consent) or by the next of kin, acting as personal consultee (according to the personal consultee's advice the patient would not have any objection to taking part in the research study), when the patient was unable to provide consent.

EXCLUSION CRITERIA

- Contraindication to TOE .
- Patients who were expected to require heart transplantation, LVAD or RVAD or biventricular VAD within 30 days from admission.
- Patients who were not expected to survive weaning from VA ECMO (liberation from VA ECMO exclusively for purpose of palliation).
- Contemporary presence of Impella device during VA ECMO weaning procedure.
- Patients in atrial fibrillation or in a different rhythm from sinus or paced at the moment of VA ECMO weaning procedure.

- Heart transplanted patients.

WITHDRAWING CRITERIA DURING THE STUDY

1) Enrolled patients dropped out from the study in case the doctors taking care of the patients decided:
    - To perform HT or VAD implantation without VA ECMO liberation;
    - To combine VA ECMO with Impella support;
    - To switch from VA ECMO to Impella support;
    - Poor quality of the echocardiographic images;
    - Patient's death before VA ECMO weaning procedure.
2) Withdraw of the patient from the study.

    The patient or the personal consultee (in case the patient was unable to provide consent) were free to withdraw the patient from the study at any time, without giving any reason, without their medical care or legal rights being affected.

ECHOCARDIOGRAPHIC PROCEDURE AND SPECKLE TRACKING ANALYSIS

During the VA ECMO weaning procedure guided by TOE, the ultrasound machine was set with stable ECG trace, frame rate of 70 ± 5Hz and at least 5 consecutive beats were recorded. According to the recruiting hospitals clinical practice, the following views (when possible) were performed at each modification of VA ECMO flow, dose of administrated drugs and other variation of coexistent types of mechanical support (Ex. IABP ratio, etc.):

1) Midesophageal 4 chamber view (mandatory);
2) Transgastric view at the level of papillary muscles (mandatory);
3) Midesophageal long axis view;
4) Midesophageal 2 chamber view.

The following views were those recommended to be performed in order to achieve LVOT VTI and RVOT VTI Doppler, although any other view which

allowed achieving a good alignment with the Doppler flow could be used. Images of LVOT VTI, RVOT VTI, were achieved at each modification of VA ECMO flow, dose of administrated drugs and other variation of coexistent types of mechanical support (Ex. IABP ratio, etc.):

5) Transgastric view which allowed to see mid left ventricle, mid right ventricle, RVOT, tricuspid valve, pulmonary valve;
6) Transgastric view (0-20°) in order to see left ventricle, right ventricle, LVOT, aortic valve, aortic root, mitral valve;
7) Transgastric view (120-140°) in order to see left ventricle, right ventricle, LVOT, aortic valve, aortic root, mitral valve;
8) Midesophageal 4 chamber view which allowed to see left and right atriums, interatrial septum, left and right ventricles, interventricular septum, mitral and tricuspid valves.

Clinical parameters, LVOT and RVOT VTI measurements, VA ECMO setting changes, etc. were recorded on the following table (Table 4). These recordings were part of a normal VA ECMO weaning trial procedure.

Table 4. Table to report data during VA ECMO weaning.

STUDY ID: Number of tentative of VA ECMO weaning: _____ Date (dd/mmm/yyyy): _____			ECMO Flow at baseline (L/min)	ECMO flow at 1 stage of weaning (L/min)	ECMO flow at 2 stage of weaning (L/min)	ECMO flow at 4 stage of weaning (L/min)	ECMO flow at 5 stage of weaning (L/min)	ECMO flow at 6 stage of weaning (L/min)
ECHO	**Midesophageal 4 chamber view~**							
	Midesophageal long axis view~							
	Midesophageal 2 chamber view~							
	Transgastric view of LV at the level of papillary muscles~							
	Transgastric view which allows to see mid left ventricle, mid right ventricle, RVOT, tricuspid valve, pulmonary valve~							
	Transgastric view (0-20) in order to see LV, RV, LVOT, aortic valve, aortic root, mitral valve~ or **Transgastric view (120-140) in order to see LV, RV, LVOT, aortic valve, aortic root, mitral valve~**							
	LVOT VTI	Cm						
	RVOT VTI	Cm						
CLINICAL	MAP	mmHg						
	P systolic	mmHg						
	P diastolic	mmHg						
	HR	Bpm						
	RAP	mmHg						
DRUGS	Adrenaline	(mcg/Kg/min)						
	Noradrenaline	(mcg/Kg/min)						
	Milrinone	(mcg/Kg/min)						
	Dobutamine							
	Vasopressin	Unit/min						
	iNOS	ppm						
	Dopamine	(mcg/Kg/min)						
MCS	IABP ratio	(STOP, NO, 1:1, 1:2, etc.)						
	Number of previous administration of levosimendan (number of vials) =							
	Date of administration of levosimendan (dd/mmm/yyyy)							
	Other							
	Comments							
~ Make a checkmark when the image is acquired								

Enrolled patients deemed suitable for VA ECMO decannulation (liberation) following a VA ECMO weaning trial procedure had images recorded as part of the weaning procedure. These images were analysed using speckle tracking software. During every weaning procedure a TOE examination with the characteristics described above was performed.

TOOLS

Echocardiogroaphy machine: Philips echocardiographic machines which were able to perform a TOE study and acquire the images in a format compatible with QLAB speckle tracking analysis. Examples of these echocardiographic machines are Epiq 7, Affiniti 70 and IE 33.

QLAB software (aCMQA.I.): Strain was measured within a Region of Interest (ROI) so that the strain of the selected tissue could be evaluated. First, the system automatically created a ROI template based on the selected view type and an analysis of the image. The user could also manually specify a ROI template by identifying three points. The system then automatically identified the endocardial boundary and created a ROI that extended from the endocardial boundary a fixed distance outward toward the epicardium. This ROI was divided into seven sub-regions to measure the regional longitudinal strains. aCMQA.I. used Lagrangian strain calculation. Each sub-region of the ROI was divided into blocks, which allowed the speckle structure in each block to be tracked. The measured deformation for each sub-region was a weighted combination of the displacements from each of the blocks. The weighting gave greater weight to endocardial blocks than to epicardial blocks. The strain was then calculated from the per-region deformation at the endocardial border. aCMQA.I. was also used to measure circumferential strain using a short-axis view. In this function, the user initiated the ROI generation process by placing a circle on the image area and adjusting it to the desired size. This ROI was divided into six sub-regions for mid short-axis display of strain results. After approval of the ROI, the strain was calculated over the cardiac cycle. The strain results were shown as a parametric image loop and the tissue motion could be displayed by playing this loop. aCMQA.I. automatically evaluated the quality of the speckle tracking for each of the ROI segments. When determining the maximum strain, aCMQA.I. gave the user complete control over the interval within which the maximum strain was measured. Common choices were the measurement of maximum strain within only the systolic period. Circumferential strain was calculated as the change in

circumference of each of region as compared to the relaxed circumference of that region. As with the measurement of longitudinal strain, the measured deformation was weighted from the endocardium to the epicardium, with the endocardium being given greater weight. The strain was calculated from the per-region deformation at the endocardial border. The strain waveforms from each of the regions could be combined into a global strain waveform. The aCMQA.I. consolidation process used a weighted average of the regional strains. The weighting factors were based on the rest length of each of the regions. By using a weighted average, each region's contribution to the global value is based on the size of the region.

SCREENING ASSESSMENTS

- Diagnosis on admission
- Age
- Inclusion criteria
- Exclusion criteria
- Cause of cardiovascular shock
- Admission date
- Admission in ICU date
- History of heart failure before the admission
- History of percutaneous coronary intervention (PCI) before the admission
- History of cardiac surgery before the admission

BASELINE ASSESSMENTS

Clinical data

- Date of birth (DD/MM/YYYY)
- Date VA ECMO weaning procedure
- Number of attempt of VA ECMO weaning procedure performed previously
- Baseline VA ECMO flow (L/min)
- Baseline Noradrenaline (mcg/Kg/min)
- Baseline Milrinone (mcg/Kg/min)

- Baseline Adrenaline (mcg/Kg/min)
- Baseline Dobutamine (mcg/Kg/min)
- Baseline Vasopressin (unit/min)
- Baseline Dopamine (mcg/Kg/min)
- Baseline iNOS (ppm)
- IABP Ratio
- MAP (mmHg)
- Systolic pressure (Psyst) (mmHg)
- Diastolic pressure (Pdiast) (mmHg)
- Heart rate (HR) (bpm)
- Right atrial pressure (RAP) (mmHg)

Baseline Echocardiographic data

- LVOT VTI (cm)
- LVOT diameter (mm)
- RVOT VTI (cm)
- RVOT diameter (mm)
- LV End diastolic volume (EDV) (mL)
- LV End systolic volume (ESV) (mL)
- Septal S' tissue Doppler imaging (TDI) (cm/sec)
- Lateral S' TDI (cm/sec)
- RV End diastolic area (EDA) (cm^2)
- RV End systolic area ESA (cm^2)
- RV S' TDI (cm/sec)
- TAPSE (mm)
- Baseline LV Circumferential strain at the level of papillary muscles
    - Peak systolic strain
- Baseline LV Longitudinal strain in Midoesophageal 4 chamber view
    - Peak systolic strain
- Baseline RV Longitudinal strain in Midoesophageal 4 chamber view
    - Peak systolic strain
    - Free wall peak systolic strain

3 cardiac cycles were analysed and an average of the conventional echocardiographic parameters and strain (calculated on Midoesophageal 4 chamber view and Transgastric view at the level of papillary muscles) values respectively obtained at echocardiographic and speckle tracking analysis were statistically analysed at the end of the study.

Clinical data

- VA ECMO flow (L/min)
- Noradrenaline (mcg/Kg/min)
- Milrinone (mcg/Kg/min)
- Adrenaline (mcg/Kg/min)
- Dobutamine (mcg/Kg/min)
- Vasopressin (unit/min)
- Dopamine (mcg/Kg/min)
- iNOS (ppm)
- IABP Ratio
- MAP (mmHg)
- Psyst (mmHg)
- Pdiast (mmHg)
- Herat rate (bpm)
- RAP (mmHg)

Echocardiographic data (at each stage of VA ECMO weaning procedure)

- LVOT VTI (cm)
- LVOT diameter (mm)
- RVOT VTI (cm)
- RVOT diameter (mm)
- LV EDV (mL)
- LV ESV (mL)
- Septal S' TDI (cm/sec)
- Lateral S' TDI (cm/sec)

- RV EDA (cm^2)
- RV ESA (cm^2)
- RV S' TDI (cm/sec)
- TAPSE (mm)
- LV Circumferential strain at the level of papillary muscles at each stage of VA ECMO weaning procedure
    - Peak systolic strain
- LV Longitudinal strain in 4 chamber view at each stage of VA ECMO weaning procedure
    - Peak systolic strain
- RV Longitudinal strain at the level of 4 chamber view at each stage of VA ECMO weaning procedure
    - Peak systolic strain
    - Free wall peak systolic strain

3 cardiac cycles have been analysed and an average of the conventional echocardiographic parameters and strain values respectively obtained at echocardiographic and speckle tracking analysis were statistically analysed at the end of the study.

At each stage of the VA ECMO weaning procedure (Baseline included), such as at every change of parameters like VA ECMO flow or administered drug or IABP ratio, the aforementioned data were recorded.
In terms of speckle tracking analysis and other conventional echocardiographic parameters not immediately used in the VA ECMO weaning process, they were measured after the end of the successful VA ECMO weaning procedure (post-hoc analysis).

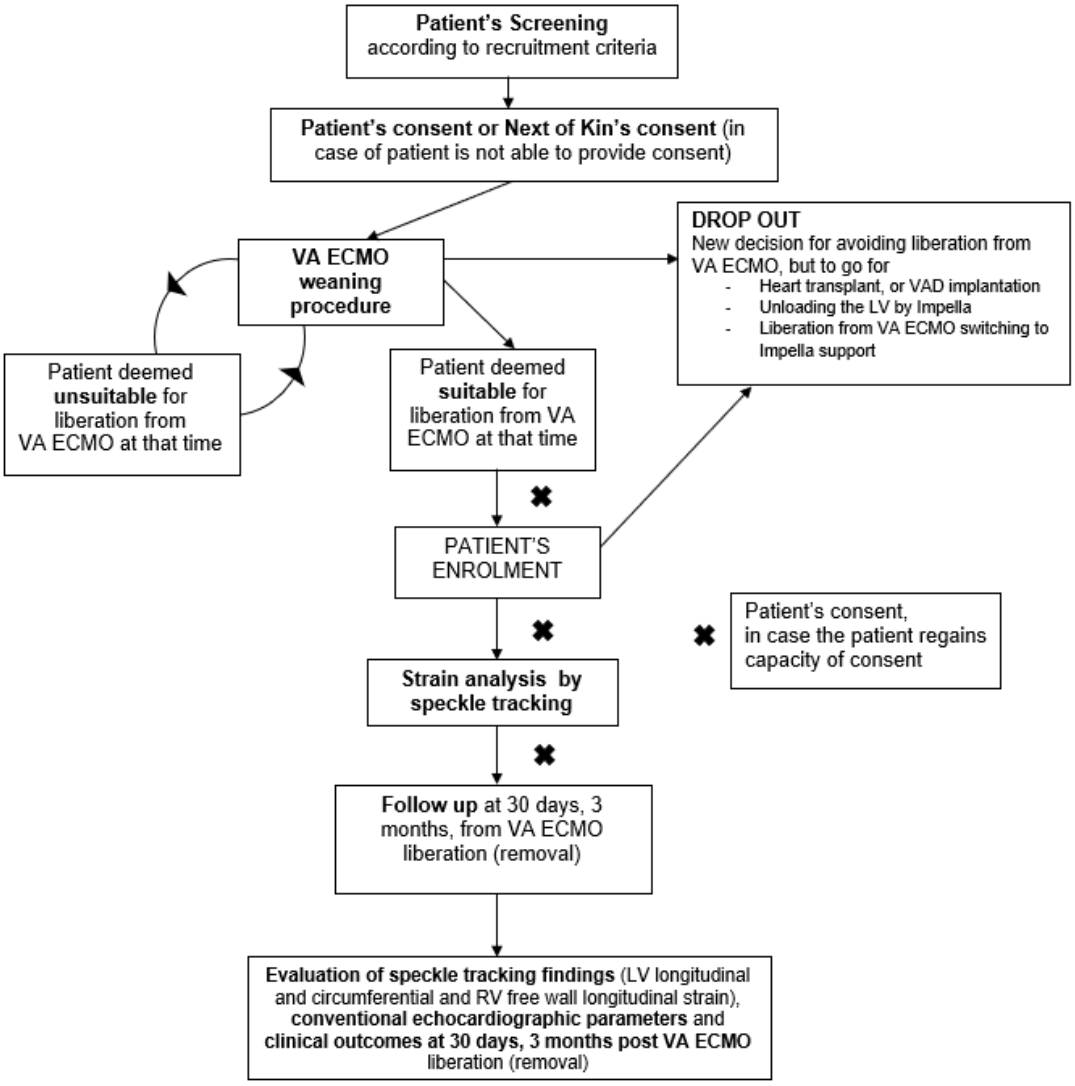
SUBSEQUENT ASSESSMENTS

Clinical data

- Date of VA ECMO liberation (removal)

- Patient status at 30 days, 3 (± 1) months after VA ECMO liberation (Alive, Dead, lost to follow up, dropped out, reason for dropping out)
- Death cause (if applicable)
- Days of hospitalization (date of discharge from the hospital)
- Days in ICU
- Necessity of new Mechanical Support device within hospitalization (IABP excluded)
- Necessity of new Mechanical Support device within 30 days, 3 (± 1) months (IABP excluded)
- Mortality within 30 days, 3 (± 1) months from VA ECMO liberation
- New necessity of high doses of ino-vasopressors within 30 days, 3 (± 1) months from VA ECMO liberation (specify the reason: cardiac or not)
- Hospitalization due to Heart failure within 30 days, 3 (± 1) months from VA ECMO liberation
- Hospitalization due to cardiogenic shock within 30 days, 3 (± 1) months from VA ECMO liberation
- Necessity of LVAD or HT within 30 days, 3 (± 1) months from VA ECMO liberation
- Necessity of RVAD implantation within 30 days, 3 (± 1) months from VA ECMO liberation

FLOWCHART


Patient's Screening
according to recruitment criteria
Patient's consent or Next of Kin's consent (in case of patient is not able to provide consent)
VA ECMO weaning procedure
DROP OUT
New decision for avoiding liberation from VA ECMO, but to go for
- Heart transplant, or VAD implantation
- Unloading the LV by Impella
- Liberation from VA ECMO switching to Impella support
Patient deemed unsuitable for liberation from VA ECMO at that time
Patient deemed suitable for liberation from VA ECMO at that time
PATIENT'S ENROLMENT
Patient's consent, in case the patient regains capacity of consent
Strain analysis by speckle tracking
Follow up at 30 days, 3 months, from VA ECMO liberation (removal)
Evaluation of speckle tracking findings (LV longitudinal and circumferential and RV free wall longitudinal strain), conventional echocardiographic parameters and clinical outcomes at 30 days, 3 months post VA ECMO liberation (removal)

PRIMARY ENDPOINT

Clinical outcome was a composite outcome of the following endpoints of interest:

- death for any reason within 3 (±1) months from VA ECMO liberation;
- the necessity of new MCS (VA-ECMO, Impella, LVAD, RVAD) or new necessity of high doses of ino-vasopressors within 3 months from VA ECMO liberation
    - Whether after VA ECMO liberation the patient still needed IABP support, this situation was not considered necessity of new MCS. Necessity of new MCS was defined when IABP or RVAD or LVAD or VA ECMO or Impella were needed again after their removal.
- HT within 3 (±1) months from VA ECMO liberation;
- readmission to hospital for cardiogenic shock/heart failure within 3 (±1) months from VA ECMO liberation.

SECONDARY ENDPOINT

Cardiac outcome was a composite outcome of the following endpoints of interest:

- death for cardiac reason within 3 (±1) months from VA ECMO liberation;
- the necessity of new MCS (VA-ECMO, Impella, LVAD, RVAD) or new necessity of high doses of ino-vasopressors for cardiac reasons within 3 months from VA ECMO liberation
    - Whether after VA ECMO liberation the patient still needed IABP support, this situation was not considered necessity of new MCS. Necessity of new MCS was defined when IABP or RVAD or LVAD or VA ECMO or Impella were needed again after their removal;
- HT within 3 (±1) months from VA ECMO liberation;
- readmission to hospital for cardiogenic shock/heart failure within 3 (±1) months from VA ECMO liberation.

STATISTICS

Patients who underwent VA ECMO liberation and experienced a clinical outcome within 3±1 months from decannulation (death for any reasons, new necessity of MCS, heart transplant/VAD implantation, new necessity of high dose of ino-vasopressors, new hospitalization for cardiogenic shock/heart failure) were compared with outcome-free patients.
A sub-analysis of the group of patients developing clinical outcomes was made focusing specifically on cardiac outcomes (death for cardiac reasons, new necessity of MCS for heart failure/cardiogenic shock, heart transplant/VAD implantation, new necessity of high dose of ino-vasopressors for cardiac reason, new hospitalisation for heart failure/cardiogenic shock).

Descriptive and bivariable analyses

The variables considered, in addition to the socio-demographic ones, were mainly echocardiographic and haemodynamic: speckle tracking parameters (LV longitudinal strain, RV free wall longitudinal strain, LV circumferential strain), EF, TAPSE, RV S' TDI, RV FAC, LVOT VTI, iSV, CI, MAP, HR, etc.
All variables were summarized by median and interquartile range (IQR) because they deviated from normality. Categorical variables were expressed as percentages. The skewness of the variables has been estimated by Shapiro-Wilk tests. Regarding continuous variables, differences between groups were estimated using two-samples independent t-test and Wilcoxon signed-rank test in case of non-normal distributions. The distribution of categorical variables was compared by chi-squared test or Fisher's exact test.

ROC analyses

To evaluate the ability of the a priori classification of patients who will experience a clinical outcome, ROC (Receiver Operating Characteristic) curves of the variable of interest were developed and the area under the curve (AUC) calculated. Hypothetical cutoffs were also identified on the basis of the necessary clinical sensitivity. The AUC values were interpreted as per the literature(95)

according to the following criterion: 0.6-0.7 = acceptable; 0.7-0.8 = good; 0.8-0.9 excellent; 0.9-1 optimal. Regarding the variables EF, LVOT VTI, iSV, CI, since the value is on average higher for outcome-free patients, the non-event of the outcome was considered as an event.

Mixed effects modelling

Finally, for the variables of interest, the trajectories relating to the three different VA ECMO support flows (flow 1, 2 and 3) stratified for the outcome-free group and the group that experienced a clinical outcome were estimated. In this regard, linear mixed-effect models have been developed. The group variable (outcome-free, clinical-outcome), the flow level and the interaction between the two factors were included as predictors. The patient ID was introduced as a random factor to take into account the clustered structure of the data. The result of interest of these models was the statistical significance of the interaction terms that could be interpreted as a different response of the patients of one group compared to the other at different levels of VA ECMO flow. The graphs of the trajectories predicted by the models were reported for the variables with statistically significant interactions.

RESULTS

In the period between November 2018 and August 2021 (2018-2019 Royal Brompton and Harefield NHS Foundation Trust; 2019-2021 Azienda Ospedaliera Universitaria Integrata Verona) a total of 92 patients have been screened. Table 5 shows the reasons for not recruiting them and the percentage.

Table 5. Reason for screening failure.

	Palliation	Presence of Impella	HT/VAD/TAH implantation expected within	Quality of Images	Permanent Atrial Fibrillation	VA ECMO post Heart Transplant	No consent achieved
n	28	16	4	4	4	6	9
(%)	(39.4)	(22.5)	(5.6)	(5.6)	(5.6)	(8.5)	(12.7)

In the end, 21 patients were liberated from VA ECMO and included in the study. Two patients were excluded because of the quality of the images which could not allow any reliable analysis (dropped outs). The causes of cardiovascular shock were myocarditis (2 patients), acute coronary artery syndrome (8 patients), post-cardiotomy (5 patients), aortic prosthesis dysfunction (1 patient), vasoplegia post lung transplant (1 patient) and septic cardiomyopathy (2 patients). Five of them experienced a clinical outcome (death for any reason, new hospitalisation for heart failure/cardiovascular shock or new necessity of high doses of ino-vasopressors, or necessity of MCS, HT or VAD implantation within 3±1 months from VA ECMO removal), and 3 of those had a cardiovascular outcome (death for cardiac reason, new hospitalisation for heart failure/cardiogenic shock or new necessity of high doses of ino-vasopressors due to heart failure, or new necessity of MCS, HT or VAD implantation within 3±1 months from VA ECMO removal). Twelve patients were supported by pVA ECMO, the remaining by cVA ECMO. The patients developing clinical outcomes were prevalently supported by pVA ECMO (1 postcardiotomy patient supported by cVA ECMO and 4 pVA ECMO). During the VA ECMO weaning trial, 15.8% of patients had an intra-aortic balloon pump

(IABP) and 36.8% had a paced rhythm. Among patients developing cardiac outcomes only 1 was unloaded by IABP. Seven patients (36.8%) received a levosimendan infusion before VA ECMO weaning. The median flow before starting a VA ECMO weaning trial was 2.8L/min (IQR 1.2), the lowest flow before VA ECMO decannulation was 1.0L/min (IQR 0.5). Data at an intermediate flow of VA ECMO support was available in 9 patients.

Splitting the recruited patients into two groups (patients experiencing a clinical outcome and patients free from clinical outcomes), the groups were comparable in terms of ino-vasopressor infusion, IABP use, type of VA ECMO, patients' age and clinical history, and cause of cardiovascular shock (Table 6).

Table 6: General population's characteristics.

	CLINICAL OUTCOME – (n = 14)	**CLINICAL OUTCOME + (n = 5)**	**p Value**
Sex, n (%)			
Male	9 (64.3)	4 (80.0)	1.000*
Female	5 (35.7)	1 (20.0)	
Age (years), median (IQR)	44.5 (16.0)	54.0 (4.0)	0.3531**
BMI (Kg/m²), median (IQR)	25.8 (6.8)	25.7 (6.6)	0.6434**
pVA ECMO, n (%)			
Central	6 (42.9)	1 (20.0)	0.603*
Peripheral	8 (57.1)	4 (80.0)	
Heart failure history, n (%)			
No	10 (71.4)	4 (80.0)	1.000*
Yes	4 (28.6)	1 (20.0)	
PCI history, n (%)			
No	13 (92.9)	4 (80.0)	0.468*

Yes	1 (7.1)	1 (20.0)	
Cardiac surgery history, n (%) **No** **Yes**	 12 (85.7) 2 (14.3)	 5 (100) 0 (0.0)	1.000*
Days from decannulation to ICU discharge (median; IQR)	14.5 (13.0)	16.0 (38.0)	0.4579**
Days from decannulation to Hospital discharge (median; IQR)	34.0 (26.0)	51.0 (55.0)	0.3125**
IABP, n (%) **No** **Yes**	 10 (71.4) 4 (28.6)	 4 (80.0) 1 (20.0)	1.000*
Levosimendan administration, n (%) **No** **Yes**	 10 (71.4) 4 (28.6)	 2 (40.0) 3 (60.0)	0.305*
Paced Rhythm, n (%) **No** **Yes**	 9 (64.3) 5 (35.7)	 3 (60.0) 2 (40.0)	1.000*

*Fisher test ; **Wilcoxon rank sum test

As reported in Table 7 and Figure 1, at the lowest flow of VA ECMO support (1 L min^{-1}, interquartile range (IQR) 0.5), the median EF was 23.9% (IQR 15.4) in the clinical outcome + patients and 45.8% (IQR 18.7) in clinical outcome – patients ($p = 0.06$). Considering RV function, no significant difference was found between clinical outcome + and clinical outcome – patients (FAC 38.0% IQR 4.7 vs 38.9

IQR 15.1, p = 0.83; TAPSE 1.4cm IQR 0.3 vs 1.2cm IQR 0.7, p = 0.35; RV s' 10.5cm/sec IQR 4.1 vs 5.6cm/sec IQR 4.8, p = 0.07). Focusing on strain parameters, for clinical outcome + patients the median LV longitudinal strain was -10.3% (IQR 2.9); RV FW longitudinal strain -11.0% (IQR 2.3); LV circumferential strain -11.6% (IQR 12.0). Conversely, for clinical outcome – patients the median LV longitudinal strain was -10.3% (IQR 5.8); RV FW longitudinal strain -16.1% (IQR 7.9); LV circumferential strain -14.5% (8.5). No significant differences were found when MAP, Psyst and CI were considered. The indexed end diastolic volume (iEDV) was significantly lower in patient free from clinical outcomes compared to the others (respectively 44.7 ml m^{-2} IQR 17.9; 70.4 ml m^{-2} IQR 43.0, $p < 0.01$), instead the RAP was higher (13.0mmHg IQR 3.5; 10.0mmHg IQR 1, p = 0.03).

The median CI was 2.3ml $min^{-1}m^{-2}$ (IQR 0.7) in the group experiencing clinical outcomes and 3.3ml $min^{-1}m^{-2}$ (IQR 1.1) in free from clinical outcome patients.

Table 7. Haemodynamic and echocardiographic values at the lowest flow of VA ECMO support of patients developing clinical outcomes and of free from outcome patients.

VA ECMO minimum flow	clinical outcome – (n =14)	clinical outcome + (n = 5)	p-Value**
MAP (mmHg), median (IQR)	70.0 (19.0)	78.0 (8.0)	0.52
HR (bpm), median (IQR)	95.5 (19.0)	87.0 (3.0)	0.38
RAP (mmHg), median (IQR)	13.0 (3.5)	10.0 (1.0)	0.03
TAPSE (cm), median (IQR)	1.2 (0.7)	1.4 (0.3)	0.35
RV s' (cm/sec), median (IQR)	5.6 (4.8)	10.5 (4.1)	0.07
FAC (%), median (IQR)	38.9 (15.1)	38.0 (4.7)	0.83
EF (%), median (IQR)	45.8 (18.7)	23.9 (15.4)	0.06
LVOT VTI (cm), median (IQR)	16.2 (7.4)	13.9 (3.1)	0.07
RVOT VTI (cm), median (IQR)	12.4 (2.5)	14.0 (8.7)	0.85

iSV (ml m^{-2}), median (IQR)	32.4 (12.9)	26.7 (2.8)	0.23
CI (L min^{-1} m^{-2}), median (IQR)	3.3 (1.1)	2.3 (0.7)	0.16
iEDV (ml m^{-2}), median (IQR)	44.7 (17.9)	70.4 (43.0)	<0.01
iRVDA (cm^2 m^{-2}), median (IQR)	10.1 (3.2)	8.6 (2.3)	0.39
LV longitudinal strain (%), median (IQR)	-10.3 (5.8)	-10.3 (2.9)	0.78
RV free wall longitudinal strain (%), median (IQR)	-16.1 (7.9)	-11.0 (2.3)	0.11
RV longitudinal strain (%), median (IQR)	-16.2 (8.3)	-11.5 (1.3)	0.14
LV circumferential strain (%), median (IQR)	-14.5 (8.5)	-11.6 (12.0)	0.40

**Wilcoxon Rank Sum Test

Analysing the recruited patients on the basis of cardiac outcome development (Table 8 and Figure 1), the median iEDV remained significantly lower in patients free from cardiac outcomes (46.6 ml $min^{-1}m^{-2}$ IQR 27.8) compared to the others (112.5 ml $min^{-1}m^{-2}$ IQR 47.1) (p = 0.01). Furthermore, the median circumferential strain was -5.6% (IQR 1.0) and the EF 23.0% (IQR 4.2) in the group of patients experiencing cardiac outcomes. Conversely, circumferential strain was -15.5% (IQR 6.5) and EF was 45.8% (IQR 15.6) for patients free from cardiac outcomes.

Table 8. Haemodynamic and echocardiographic values at the lowest flow of VA ECMO support of patients developing cardiac outcomes and of free from cardiac outcome patients.

VA ECMO minimum flow	**cardiac outcome – (n = 16)**	**cardiac outcome + (n = 3)**	**p-Value****
MAP (mmHg), median (IQR)	70.0 (20.0)	81.0 (10.0)	0.09
HR (bpm), median (IQR)	92.0 (17.0)	85.0 (30.0)	0.61
RAP (mmHg), median (IQR)	12.5 (4.0)	10.0 (2.0)	0.21
TAPSE (cm), median (IQR)	1.2 (0.8)	1.4 (0.1)	0.32

RV s' (cm/sec), median (IQR)	6.9 (5.8)	12.4 (7.7)	0.20
FAC (%), median (IQR)	40.5 (17.1)	36.4 (6.2)	0.26
EF (%), median (IQR)	45.8 (15.6)	23.0 (4.2)	<0.01
LVOT VTI (cm), median (IQR)	16.0 (7.9)	13.9 (4.8)	0.21
iSV (ml m^{-2}), median (IQR)	30.2 (11.6)	28.7 (23.0)	0.82
CI (L min^{-1} m^{-2}), median (IQR)	3.2 (1.3)	2.9 (2.0)	0.91
iEDV (ml m^{-2}), median (IQR)	46.6 (27.8)	112.5 (47.1)	0.01
iRVDA (cm^2 m^{-2}), median (IQR)	9.9 (2.7)	8.6 (12.1)	1.00
LV longitudinal strain (%), median (IQR)	-10.3 (6.6)	-10.3 (4.8)	0.43
RV free wall longitudinal strain (%), median (IQR)	-15.4 (8.7)	-11.0 (3.0)	0.17
RV longitudinal strain (%), median (IQR)	-15.1 (8.0)	-11.5 (1.3)	0.38
LV circumferential strain (%), median (IQR)	-15.5 (6.5)	-5.6 (1.0)	0.02

**Wilcoxon Rank Sum Test

Figure 1. EF, iEDV, FAC and Strain values at the lowest flow of VA ECMO support.

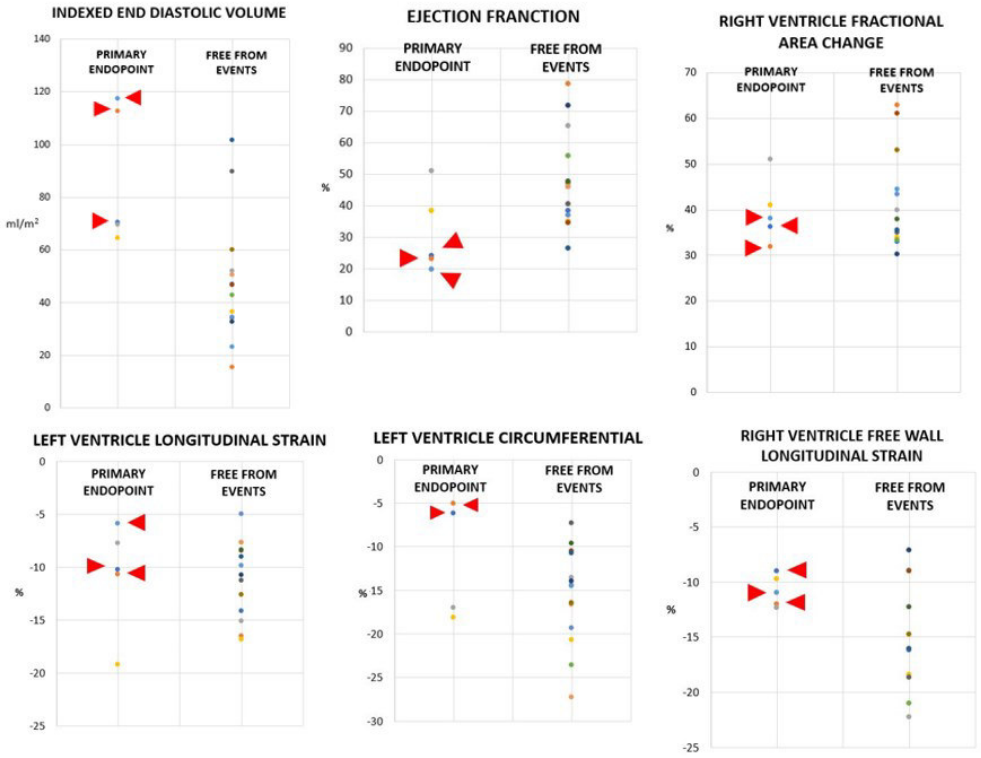


Figure Legend: Primary endpoint: patients developing clinical outcomes after VA ECMO removal. Free from events: patients who did not experience any outcome after VA ECMO removal. Red Triangles: patients developing cardiac outcomes.

At the highest flow of support before VA ECMO weaning trial (2.8 L min^{-1} IQR 1.2), the median CI in patients free from clinical outcomes was 2.5 L $min^{-1}m^{-2}$ (IQR 1.7), while it was 2.1 L min^{-1}-m^{-2} (IQR 1.2) in clinical outcome + patients and 2.3 L $min^{-1}m^{-2}$ (IQR 2.1) in the subgroup of cardiac outcome + patients (Table 9 and 10). Focusing on conventional echocardiographic parameters, iEDV was significantly higher (89.5 ml m^{-2}, IQR 54.5 vs , 43.8 ml m^{-2}, IQR18.1, p = 0.02) in the clinical outcome + group and the EF was significantly lower (22.6%, IQR10.3 vs 46.6% IQR 21.6, p = 0.03).

Table 9. Haemodynamic and echocardiographic values at the highest flow of VA ECMO support of patients developing a clinical outcome and of free from outcome patients.

VA ECMO highest flow	clinical outcome – (n=11)	clinical outcome + (n=4)	p-Value**
MAP (mmHg), median (IQR)	83.0 (31.0)	75.5 (14.5)	0.40
P Syst (mmHg), median (IQR)	123.0 (51.0)	115.5 (51.0)	1.00
HR (bpm), median (IQR)	91 (39.0)	87 (16.5)	0.51
RAP (mmHg), median (IQR)	10.5 (2.0)	7.5 (2.0)	0.02
TAPSE (cm), median (IQR)	0.73 (1.0)	1.5 (0.5)	0.13
RV s' (cm/sec), median (IQR)	6.23 (4.8)	9.0 (0.0)	0.35
FAC (%), median (IQR)	43.3 (13.2)	38.8 (11.4)	0.64
EF (%), median (IQR)	46.6 (21.6)	22.6 (10.3)	0.03
LVOT VTI (cm), median (IQR)	12.8 (6.2)	10.6 (2.9)	0.24
iSV (ml), median (IQR)	24.6 (10.1)	22.9 (20.6)	0.90
CI (L min^{-1} m^{-2}), median (IQR)	2.5 (1.7)	2.1 (1.2)	0.90
iEDV (ml m^{-2}), median (IQR)	43.8 (18.1)	89.5 (54.5)	0.02
iRVDA (cm^2 m^{-2}) , median (IQR)	9.5 (2.1)	8.5 (5.8)	0.76
LV longitudinal strain (%), median (IQR)	-10.7 (3.7)	-10.4 (7. 9)	0.82
RV free wall longitudinal strain (%), median (IQR)	-12.9 (6.1)	-13.6 (5.8)	0.41
RV longitudinal strain (%), median (IQR)	-13.6 (5.7)	-13.3 (7.4)	0.93
LV circumferential strain (%), median (IQR)	-10.8 (10.7)	-6.7 (7.2)	0.10

**Wilcoxon Rank Sum Test

Table 10. Haemodynamic and echocardiographic values at the highest flow of VA ECMO support of patients developing cardiac outcomes and of free from cardiac outcome patients.

VA ECMO highest flow	cardiac outcome – (n= 12)	cardiac outcome + (n = 3)	p-Value**
MAP (mmHg), median (IQR)	79.0 (31.0)	76.0 (3.0)	0.94
P Syst (mmHg), median (IQR)	110.5 (46.0)	132.0 (39.0)	0.39
HR (bpm), median (IQR)	91.0 (29.5)	86.0 (29.0)	0.66
RAP (mmHg), median (IQR)	10.0 (4.0)	8.0 (1.0)	0.12
TAPSE (cm), median (IQR)	0.8 (0.9)	1.4 (0. 9)	0.22
FAC (%), median (IQR)	43.2 (13.2)	41.0 (18.3)	0.87
EF (%), median (IQR)	43.4 (20.8)	20.3 (10.5)	0.03
LVOT VTI (cm), median (IQR)	12.4 (4.7)	10.7 (5.5)	0.47
iSV (ml m^{-2}), median (IQR)	24.5 (10.8)	28.7 (29.1)	0.39
CI (L min^{-1} m^{-2}), median (IQR)	2.4 (1.3)	2.3 (2.1)	0.77
iEDV (ml m^{-2}), median (IQR)	45.5 (16. 6)	111.8 (50.5)	0.01
iRVDA (cm^2 m^{-2}), median (IQR)	8.8 (3.1)	9.4 (8.7)	0.61
LV longitudinal strain (%), median (IQR)	-10.7 (3.7)	-10.4 (7. 9)	0.82
RV free wall longitudinal strain (%), median (IQR)	-12.9 (6.1)	-13.6 (5.8)	0.41
RV longitudinal strain (%), median (IQR)	-13.6 (5.7)	-13.3 (5.8)	0.93
LV circumferential strain (%), median (IQR)	-10.8 (9.5)	-5.2 (3.1)	0.03

**Wilcoxon Rank Sum Test

The effect of VA ECMO flow on echocardiographic parameters and CI in patients who had both the data in high flow (similar values) and low flow of VA ECMO support have been graphically compared (Figure 2 and 3). By decreasing the VA

ECMO flow support, the CI of cVA ECMO and pVA ECMO patients increased. The EF of cVA ECMO patients increased with the reduction of VA ECMO support, while it did not change significantly in pVA ECMO patients (even when a sub-analysis for cardiac outcome development was performed). Circumferential and longitudinal strain did not show any specific trend associated with the reduction of VA ECMO flow (Figure 3).

Figure 2. Ejection Fraction and Cardiac Index of each patients having both VA ECMO high flow and low flow data.

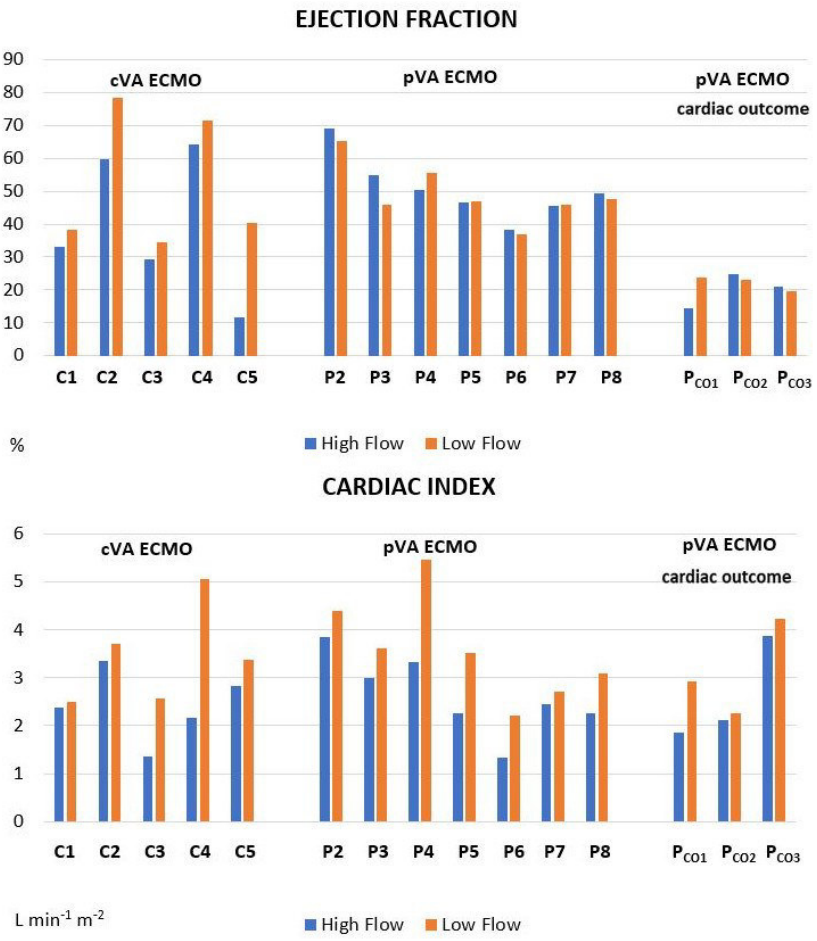


Figure Legend: High Flow: VA ECMO flow before the weaning trial; Low flow: VA ECMO flow = 1±0.5L min^{-1}; C = central; P = peripheral; P_{CO} = peripheral developing cardiac outcome.

Figure 3. LV and RV Free wall Longitudinal strain and LV Circumferential strain of each patients having both VA ECMO high flow and low flow data.

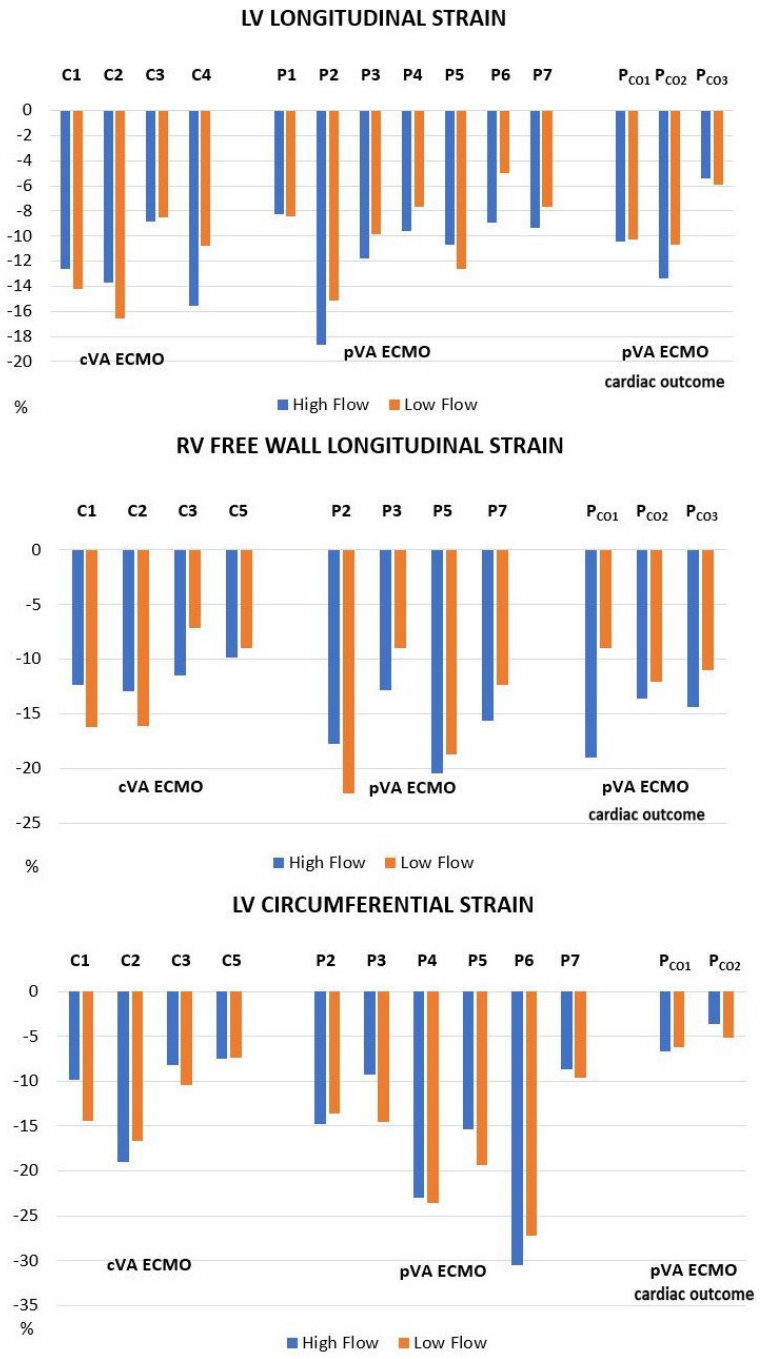


Figure Legend: High Flow: VA ECMO flow before the weaning trial; Low flow: VA ECMO flow = 1±0.5L min^{-1}; C = central; P = peripheral; P_{CO} = peripheral developing cardiac outcome.

When mixed effects model was applied to all the patients with all the flow values of VA ECMO support (flow 1 = low flow, flow 2 = mid flow, flow 3 = high flow), it highlighted a statistically significant interaction between the flow level and the patient group (clinical outcome + and clinical outcome – group) for the following variables: CI, iSV and RV Free wall longitudinal strain (Figure 4). In particular, the difference in CI at flow 3 from baseline was 0.82 points higher in those who experienced the clinical outcome than in outcome free patients (p = 0.029). Similarly, the iSV value from baseline was 7.89 points higher in the clinical outcome group (p = 0.049). On the contrary, in case of RV Free wall longitudinal strain the difference between flow 3 and flow 1 was lowered by 4.42 points in the group with outcome (p = 0.042).

Figure 4. Effect of flow change on iSV, Cardiac index and RV Free wall longitudinal strain with the application of mixed effects model.

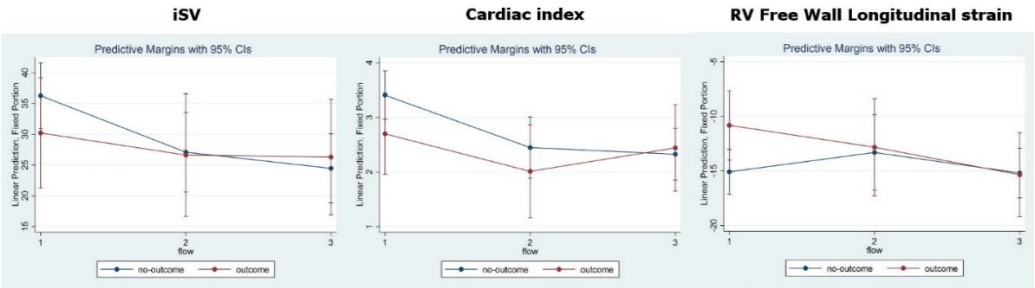


Figure Legend: flow 1 = low flow, flow 2 = mid flow, flow 3 = high flow.

A ROC analysis was performed to identify the best cut point value of different echocardiographic parameters to discriminate patients developing clinical outcomes from those free from outcomes at the lowest flow of VA ECMO support. For EF, the best cut point was 26.32% (AUC 0.79), for LVOT VTI the best cut point found was 14.36cm, with an AUC 0.78, for CI was 2.5ml min^{-1} m^{-2} (AUC 0.71). Considering strain parameters, only peak systolic RV free wall longitudinal strain had an acceptable AUROC (0.75) with the cut off of -12.0% (Appendix). Because of the paucity of data the 95% confidence interval of all AUROCs were relatively large.

DISCUSSION

For an intensivist, VA ECMO weaning trial is of crucial importance for the VA ECMO patients' management. Premature VA ECMO liberation without sufficient ventricular recovery may cause a cardiovascular shock relapse. However, an unjustified delay in VA ECMO removal exposes the patients to the risk of complications and death. Furthermore, prolonging VA ECMO support on a patient without any ventricular recovery (VA ECMO support dependent) and who is not a candidate for VAD, biventricular assist device or HT is ethically questionable. For these reasons, several studies have been performed to find predictors to identify patients who can tolerate VA ECMO removal or not.
This pilot study is one of the few studies analysing specifically VA ECMO weaned patients (21 recruited patients out of 92 patients screened). In other studies patients who were considered not able to tolerate VA ECMO removal were considered VA ECMO weaning failure and directly transplanted or VAD implanted(73,82,87). This consideration is particularly important when we consider LVOT VTI as predictor of VA ECMO weaning. In our pilot study, the median LVOT VTI for patients free from cardiac outcomes and for patients developing a cardiac outcome was 16.0cm (IQR 7.9), and 13.9cm (IQR 4.8) respectively. No significant difference was found between the two groups. This is in contrast with what was found in a previous study(73). The discrepancy may be explained by two considerations: firstly, our research study is based on the scientific knowledge so far achieved, so that, only few of the recruited patients have been liberated from VA ECMO when LVOT VTI was lower than 10cm. Indeed, patients were generally liberated from VA ECMO when they had a CI ≥ 2.2L min^{-1} m^{-2}. This is also due to the fact that the two recruiting hospitals involved in the study usually based the decision to wean a patient on multiple haemodynamic parameters: especially CI, MAP and blood gasses. The acritical application of LVOT VTI ≥ 10 cm or MAP ≥ 60mmHg, may be misleading since a MAP ≥ 60mmHg may be consequent to an excessive vasoconstriction, and iSV depends on patient's body surface area, LVOT area and LV ejection time and LVOT VTI.

Secondly, our pilot study considered only patients who underwent a proper VA ECMO weaning. The patients who were judged VA ECMO dependent by the clinical team and directly bridged to HT or VAD were excluded (5.6% of the screened patients). So patients with particularly low LVOT VTI, iSV and CI were excluded from the analysis. Indeed, the median CI of patients free from cardiac outcomes was 3.2 L min^{-1} m^{-2} (IQR 1.3), and 2.9 L min^{-1} m^{-2} (IQR 2.0) for patients developing a cardiac outcome. Focusing on MAP, despite it didn't reach the statistical significance, patients who developed cardiac outcomes had higher MAP median value compared to patients who didn't experience cardiac outcomes. This is in part in contrast with what it could be thought intuitively, because low cardiac output may correspond to low arterial pressure. However, a possible explanation is that a higher level of vasoconstriction occurs in patients with low cardiac output. This may be consequent to both a higher use of vasopressors used by intensivists at the moment of VA ECMO weaning or a physiological reaction to low peripheral perfusion. It is important to highlight that the MAP IQR of cardiac outcome + and – patients are wide, so that this observation may be just the result of chance.

Focusing on conventional echocardiographic parameters, the patients developing clinical outcomes (3 had a cardiac failure relapse, one died for septic shock, and one needed high dose of ino-vasopressors post VA ECMO removal for septic shock) presented an iEDV significantly higher than those free from outcomes (Figure 1). This finding may have several explanations. First, it is possible that more dilated ventricles correspond to a more severe and chronic LV dysfunction. Second, the higher is iEDV value the flatter is the slope of the Frank Starling curve (graph of EDV plotted against SV)(96). Of note, Frank Starling curve is still valid in case of VA ECMO support(97). Therefore, patients experiencing a clinical outcome could be those not able to increase their CO by Frank Starling principle or those that may have a less favourable pressure volume area (PVA) and so able to generate less stroke work(42,98). These hypotheses are supported by the findings on mixed random effect model (Figure 4). Indeed, the response to the reduction of the VA ECMO flow support, was different for clinical outcome + patients and clinical outcome – patients. The group developing clinical outcomes

was the group with a lower increase in iSV and CI, and almost no change in RV free wall longitudinal strain.

A possible objection to this explanation could be that not all the patients experiencing an outcome had a particularly high iEDV on the basis of chambers quantification references value(99). However, it is worth of consideration that one of the limitations of TOE assessment is the underestimation of cardiac chamber volumes due to a foreshortening problem.

Our pilot study is limited by the low number of cardiovascular events (only 3 out of 19 analysed patients) or major clinical events (5 in total), however some interesting findings can be observed.

EF, LV longitudinal and circumferential strain failed to be significantly different between the group of patients experiencing clinical outcomes and the group of those not experiencing any outcomes (Table 7). However, especially for EF and circumferential strain it is possible to observe how the absolute values of these parameters are lower in patients experiencing cardiac outcomes. This aspect is less evident, but still recognisable in LV longitudinal strain values (Figure 1). This is in line with what observed by Aissaoui et al(73) and recommended in ELSO guidelines(81). In contrast with Huang et al(76), RV function parameters (TAPSE, RV s', FAC, RV free wall longitudinal strain) did not show any significant difference in the compared groups of patients. However, if we graphically organise RV free wall longitudinal strain values and FAC, we can observe lower values of strain in patients developing clinical outcomes (Figure 1). It is worth noticing that patients developing clinical outcomes were patients experiencing cardiac outcomes or being complicated by septic shock. RV is particularly vulnerable to septic shock, and RV dysfunction has been associated with higher mortality in patients affected by sepsis(100). This may be the reason why RV free wall longitudinal strain and FAC values at the lowest flow of VA ECMO support are similar between patients experiencing specifically cardiac outcomes and patients complicated by septic shock. It is also important to underline that, despite the best possible alignment of TDI signal and M-mode was obtained and the lowest foreshortening was tried to achieve for strain analysis, TOE is not the best imaging technique to be used to assess the RV systolic

function. This may explain some of the discrepancies between our findings (especially the conventional echocardiographic ones) and those of Huang et al (3D transthoracic echocardiogram)(76). It is important to mention that RV s' values in our analysis appears to be the more affected by bad alignment, indeed their values do not follow the trend of other RV function parameters.

The peculiar trend of circumferential strain in cardiac outcomes + patients and clinical outcome + patients complicated by septic shock (Figure 1) and the knowledge on the impact of sepsis on RV lead to another consideration. The focus of all the research studies conducted so far was to find an algorithm or some variables who are satisfactory predictors of VA ECMO weaning success or failure. However, the causes of cardiovascular shock are different, thus the predictors of weaning success or failure may be different. This does not mean that the variables so far identified are not valid, but it could explain why they are not 100% reliable. Extremely simplifying a CI < 2.2L min^{-1} m^{-2} may be due to both pulmonary embolism, RV myocardial infarction, LV myocardial infarction and myocarditis. A VA ECMO is useful in all the aforementioned cases; however, the treatments of these conditions are different as well as the prognosis. In our study the cardiac outcome + patients were a case of myocarditis and two cases of severe ischaemic heart disease with biventricular involvement. This may explain the low values of RV free wall longitudinal strain which were comparable to the values of the clinical outcome + patients complicated by septic shock.

Critically analysing the results of ROC analysis performed considering clinical outcomes, an acceptable AUROC was found for EF, CI and RV free wall longitudinal strain (AUC between 0.7 and 0.8, see Appendix). However, the best cut point for EF was 26.32% and CI 2.5L $min^{-1}m^{-2}$. These cut points are not useful from a clinical point of view, since patients with EF of 26.32% and with CI 2.5 L $min^{-1}m^{-2}$ live without a MCS and so the VA ECMO weaning is not in question in patients with these values. More useful is the cut point -12.0% for RV free wall longitudinal strain since less is known on speckle tracking parameters in the MCS setting, because the technique is relatively recent. The cut point found for RV free wall longitudinal strain is in line with what has been previously found(76,87).

Focusing on the effect of VA ECMO flow change on haemodynamic and echocardiographic parameters, other interesting trends may be observed (Figures from 2 to 4). Not surprisingly, CI increased with the reduction of VA ECMO support (Figure 2 and 4). This is due to a reduction in afterload and to an increase in preload and it has been described previously in scientific literature(42). In contrast, it was not detected any peculiar trend in EF (Figure 2) and LV strain parameters (Figure 3). The absence of consistent increasing tendency in absolute value of strain may be related to the fact that strain assesses contractility and not volume (CI = stroke volume x HR/body surface area; EF = SV / EDV x 100). So that the preload and afterload changes may not determine an important increase in contractility. Another possible explanation is that the increment in preload and the decrease in afterload determine a variation on EDV which altered the position of the heart in the Frank Starling curve. It was not possible a precise evaluation EDV variation because of the limited images recorded during the VA ECMO weaning trial (Midoesophageal 4 chamber and Transgastric view at the level of papillary muscles). The absence of rise in EF with the reduction of VA ECMO support is more challenging to be explained. In a normal heart, an increase in preload and/or a reduction in afterload determine a rise in CI. However, this rise in CI does not correspond to a significative increase in EF(101). Furthermore, some of the recruited patients had some degree of mitral regurgitation. Mitral regurgitation is responsible for a reduction in the forward stroke volume and in an overestimation of EF(102). With the reduction of VA ECMO flow support, there is a reduction in peripheral resistances and an increase in preload. These load changes, due to VA ECMO flow variations, may determine a reduction in the degree of mitral regurgitation with an increase in the forward stroke volume which can be hidden and overlooked using the EF formula.

LIMITATIONS

This research study has several limitations. First of all, the small sample size. This study was developed as a pilot study. Its purpose was to generate new hypothesis and speculations, and assess the feasibility of a larger study. Due to its particular design (only patients properly weaned from VA ECMO, no patients on atrial

fibrillation, etc.) and, not to be forgotten the Covid 19 outbreak, the sample size was relatively small, and this affected the quality of statistics unavoidably. The decision to analyse together patients supported by cVA ECMO and patients supported by pVA ECMO was strictly connected to the small sample size. The two groups of patients are different for several reasons. For example, cVA ECMO patients are generally surgical patients who underwent cardiac surgery and failed the weaning from the Heart Lung Machine. pVA ECMO are not implanted in cardiac surgery patients usually. Furthermore, as previously explained, the effect of central and peripheral VA ECMOs on afterload is different. Anyway, in the most of the published studies on VA ECMO weaning (73-75) and in the majority of the scientific literature on VA ECMO physiology(1,42) the differentiation between cVA ECMO and pVA ECMO was not made.

Second, the number of clinical events was small and they were limited to cardiac events or death/escalating dose of ino-vasopressors due to septic shock. As a consequence of that, we can make speculations only on these two particular subgroups of patients which have particularly different haemodynamics(103). The septic shock is a vasodilatory shock, with low peripheral vascular resistances and higher CI, instead the cardiac events are new episodes of cardiogenic shock (low cardiac output, high peripheral vascular resistances). Patients developing septic shock may have an underlying septic conditions, which expressed itself completely after the VA ECMO removal. Consequently, the VA ECMO weaning trial could be partly influenced by the underlying sepsis. Third, the study was developed to be feasible in an intensive care setting. VA ECMO patients have generally low quality transthoracic echocardiographic views, are haemodynamically unstable and have high bleeding risk. For these reasons, the images recorded in the study were limited to those used by intensivists during the weaning procedure (Midesophageal 4 chamber view, Transgastric view at the level of papillary muscles, Midesophageal long axis view, and those to assess LVOT VTI and RVOT VTI). These views allowed to assess biventricular function and SV, however regional wall motion abnormalities could be overlooked, thus overestimation or underestimation of strain and EF were possible. However, a comprehensive TOE study causes delays in the weaning procedure, threatening

the patient's life or VA ECMO circuit integrity. Fourth, it was not possible to record all data for each patients due to imaging qualities, different VA ECMO flows during the weaning trial, different haemodynamic conditions and intensive care/surgical requirements.

CONCLUSIONS

From this pilot study we could draw the following conclusions and insights:

1) Despite its limitations, EF is still a valuable echocardiographic parameter to distinguish VA ECMO patients who are going to tolerate VA ECMO weaning and patients who are not.
2) iEDV is a conventional echocardiographic parameter which allows to identify patients already on the plateau of the Frank Starling curve and with a unfavourable oxygen consumption/stroke work production, consequently more keen on developing adverse clinical outcomes.
3) The increment in CI, iSV and RV longitudinal strain (absolute value) with the reduction of VA ECMO flow is indicative of VA ECMO weaning success.
4) In a future study, in order to identify reliable predictors of VA ECMO weaning success it is fundamental to identify the primary cause and mechanism responsible for the cardiovascular shock and the cause of weaning failure.
    - Sub-analysis based on the cause of shock may clarify the picture;
    - Cardiac outcomes should be the principal focus for weaning failure. Other outcomes are useful in terms of prognosis assessment, but they may confound the interpretation of results.
5) LV circumferential strain and RV free wall longitudinal strain appear to be interesting parameters indicative of weaning success. However, it is necessary to prolong the pilot study recruitment to confirm their potentialities and utility.
6) This study provided the experience necessary to calculate an adequate sample size for a validation trial.

PERSPECTIVES FOR A FUTURE VALIDATION TRIAL

Delivering this pilot study offered the opportunity to find several critical issues which have become crunching food for thoughts.

1) Were the inclusion/exclusion criteria too stringent? Should they be confirmed?

Yes they were, but they had/have to be. The idea of this study was developed by observing intensivists in the process of the decision making. It is obvious that some patients cannot tolerate VA ECMO removal: with the decrease of the VA ECMO support, lactate level increases, arterial pressure drops significantly and oxygen saturation worsen. For these patients, there are no doubts, they must undergo HT, VAD implantation or a decision for palliation must be made. In this study what we wanted to address were the grey zones: patients who were judged able to be liberated from VA ECMO, but they unexpectedly had a relapse. The decision to exclude patients who were on super urgent list for VAD/HT was due to that, as weaning in these patients is not taken into consideration, consequently they do not undergo a proper VA ECMO weaning trial. Another important point was the decision to exclude patients in atrial fibrillation during VA ECMO weaning trial. Atrial fibrillation (AF) causes variability in stroke volume and cardiac contractility(104). Since we wanted to use speckle tracking to assess myocardial contractility, we decided to avoid AF as confounder on our analysis. Further, it was decided to avoid to recruit primary graft dysfunction HT patients because it was judged a too peculiar population which could bias our data(105). This was a correct thought, which should also be extended to other specific populations such as Grown-Up Congenital Heart Disease who are patients who may have some particularly altered anatomy and circulation (for example Fontan circulation)(106).

2) Is this pilot study obsolete in the era of ECMELLA (VA ECMO + Impella as venting system)?

This pilot study was developed when the ECMELLA concept was still not diffused. The decision to exclude patients with both Impella and VA ECMO was due to the fact that Impella is a venting system acting on the preload, which determines an important alteration in pressure-volume loops and areas(42), adding another variable to be considered and hindering the assessment of the contractile reserve which depends on the preload(97). Indeed, in a study on an animal model, Impella determined a reduction EDV and contractility assessed by strain (circumferential and longitudinal strain became more positive)(107). The effect of Impella on strain may be applied to all the venting system acting on preload, and consequently this has to be considered in a future validation trial. However, at present, the use of ECMELLA is spreading. Indeed 17% of the screened VA ECMO patients had Impella as venting system. The beneficial effects of Impella as venting system are: intraventricular thrombosis preventions, myocardial oxygen consumption reduction and coronary perfusion(59,60). The combination of Impella and VA ECMO appears to reduce mortality in VA ECMO patients(58,108), however randomised controlled trials are needed. Considering the advantages of Impella alone (non-inferiority of Impella compared to VA ECMO in particular populations) or the ECMELLA combination, it is expected a rise in the use of Impella device, especially in some conditions like cardiogenic shock due to myocarditis or myocardial infarction, where the reduction of myocardial oxygen consumption and the improvement of coronary perfusion are beneficial(98,109). However, VA ECMO and Impella offer two different types of support. VA ECMO gives circulatory (adapt to distributive shock, RV and or LV failure) and respiratory support. Conversely, Impella is specifically a circulatory support which provides a constant cardiac output which varies depending on the model (Impella 2.5; Impella CP, Impella 5). If Impella is used, it is particularly important to identify whether there is a LV failure (Impella 2.5; Impella CP, Impella 5) or RV failure (Impella RP) and the entity of the cardiogenic shock. It has also been described the combination of two Impellas one for the RV and one

for the LV (BIPELLA)(110). Considering the variety of causes of cardiovascular shock and the different patients' characteristics, the choice of the MCS device should be tailored on patient's requirements. In some patients VA ECMO alone or VA ECMO combined with IABP is still the best choice. Furthermore, in some countries the ECMELLA solution may be too expensive. In conclusion, a multicentre study focusing on predictors of VA ECMO weaning success designed with the inclusion/exclusion criteria of this pilot study is not obsolete.

3) This pilot study is the stepping stone for the development of a multicentre study.

From graphical analysis of the data, circumferential strain and RV free wall longitudinal strain appear to be two interesting parameters, but their potentialities could not be properly assessed so far because of a low number of events (1 cardiac outcomes every 6 VA ECMO weaned patients). If the parameters will confirm their potentialities (by recruiting some more patients) a proper sample size calculation for a multicentre study may be performed. For the variable circumferential strain, 108 patients in the "outcome free group" and 18 patients in the "cardiac outcome group" (allocation ratio: 1/6) are required to have a 90% chance of detecting, as significant at the 5% level, a difference of 5% between "outcome free group" and "cardiac outcome group". For the variable RV free wall longitudinal strain, 120 patients in the "outcome free group" and 20 patients in the "cardiac outcome group" (allocation ratio: 1/6) are required to have a 90% chance of detecting, as significant at the 5% level, a difference of 4% between "outcome free group" and "cardiac outcome group". Since exploratory studies suggest that outcomes have a skewed distribution, group sizes will be divided by 0.864 to allow for non-parametric analysis as suggested by Randle et al (111).

4) Improvement of the RV assessment in a future trial.

As mentioned above, RV free wall longitudinal strain is a promising parameter to be further investigated. RV assessment is becoming the topic of recent research

studies, and cardiologists and cardiac intensivists are focusing more and more their attention on this cardiac chamber(76,87). In reference to the study of this cardiac chamber, compared to transthoracic echocardiogram (TTE), TOE is not the best imaging method to assess RV, and VA ECMO patients generally have poor quality transthoracic images. Although this caveat, the preliminary results of the present study suggest that a more systematic study of the RV may give useful prognostic information. Thus in a future validation study, the RV should be analysed by combining TOE with good quality TTE images (when possible) at the moment of VA ECMO weaning trial. In this way, an additional analysis of the transthoracic parameters (conventional and speckle tracking) recorded could be performed. It is worth noting that TOE method was chosen in this pilot study and should remain (in a future validation study) the primary method to assess the heart function, because it is the only imaging method which guarantees the quality of images necessary to perform speckle tracking in almost all the patients. TTE was excluded as primary method of heart evaluation in this pilot study because it would have raised much more the number of dropouts due to quality of images and logistical problems. Indeed, VA ECMO weaning trial with both TTE and TOE can be performed only in ICU, where it is possible to switch ultrasound probe easily (this is almost impossible in operating theatre, where sterility of the surgical field is mandatory).

REFERENCES

1. Rao P, Khalpey Z, Smith R, Burkhoff D, Kociol RD. Venoarterial Extracorporeal Membrane Oxygenation for Cardiogenic Shock and Cardiac Arrest. Circ Heart Fail 2018;11:e004905.

2. Biancari F, Perrotti A, Dalen M et al. Meta-Analysis of the Outcome After Postcardiotomy Venoarterial Extracorporeal Membrane Oxygenation in Adult Patients. J Cardiothorac Vasc Anesth 2018;32:1175-1182.

3. Jayaraman AL, Cormican D, Shah P, Ramakrishna H. Cannulation strategies in adult veno-arterial and veno-venous extracorporeal membrane oxygenation: Techniques, limitations, and special considerations. Ann Card Anaesth 2017;20:S11-S18.

4. Babu AN. Techniques for Venoarterial Extracorporeal Membrane Oxygenation Support and Conversion to Temporary Left Ventricular Assist Device. Operative Techniques in Thoracic and Cardiovascular Surgery 2014;19:365-379.
5. McDonagh TA, Metra M, Adamo M et al. 2021 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure. Eur Heart J 2021;42:3599-3726.
6. Guglin M, Zucker MJ, Bazan VM et al. Venoarterial ECMO for Adults: JACC Scientific Expert Panel. J Am Coll Cardiol 2019;73:698-716.
7. Barbone A, Malvindi PG, Sorabella RA et al. 6 months of "temporary" support by Levitronix left ventricular assist device. Artif Organs 2012;36:639-42.
8. Lafç G, Budak AB, Yener A, Cicek OF. Use of extracorporeal membrane oxygenation in adults. Heart Lung Circ 2014;23:10-23.
9. Juo YY, Skancke M, Sanaiha Y, Mantha A, Jimenez JC, Benharash P. Efficacy of Distal Perfusion Cannulae in Preventing Limb Ischemia During Extracorporeal Membrane Oxygenation: A Systematic Review and Meta-Analysis. Artif Organs 2017;41:E263-E273.
10. Bisdas T, Beutel G, Warnecke G et al. Vascular complications in patients undergoing femoral cannulation for extracorporeal membrane oxygenation support. Ann Thorac Surg 2011;92:626-31.
11. Alexis-Ruiz A, Ghadimi K, Raiten J et al. Hypoxia and Complications of Oxygenation in Extracorporeal Membrane Oxygenation. J Cardiothorac Vasc Anesth 2019;33:1375-1381.
12. Zanatta P, Forti A, Bosco E et al. Microembolic signals and strategy to prevent gas embolism during extracorporeal membrane oxygenation. J Cardiothorac Surg 2010;5:5.
13. Cornell T, Wyrick P, Fleming G et al. A case series describing the use of argatroban in patients on extracorporeal circulation. ASAIO J 2007;53:460-3.
14. Tamura T, Horiuchi H, Obayashi Y et al. Acquired von Willebrand syndrome in patients treated with veno-arterial extracorporeal membrane oxygenation. Cardiovasc Interv Ther 2019;34:358-363.
15. Oezpeker C, Zittermann A, Baurichter D et al. Changes in Von Willebrand factor profile predicts clinical outcomes in patients on mechanical circulatory support. J Card Surg 2018;33:693-702.
16. Thiagarajan RR, Barbaro RP, Rycus PT et al. Extracorporeal Life Support Organization Registry International Report 2016. ASAIO J 2017;63:60-67.

17.Bartlett RH, Gattinoni L. Current status of extracorporeal life support (ECMO) for cardiopulmonary failure. Minerva Anestesiol 2010;76:534-40.
18.Lorusso R, Barili F, Mauro MD et al. In-Hospital Neurologic Complications in Adult Patients Undergoing Venoarterial Extracorporeal Membrane Oxygenation: Results From the Extracorporeal Life Support Organization Registry. Crit Care Med 2016;44:e964-72.
19.Camboni D, Schmid C. Neurologic and pulmonary complications in ECLS. 2017:575-581.
20.Lorusso R, Gelsomino S, Parise O et al. Neurologic Injury in Adults Supported With Veno-Venous Extracorporeal Membrane Oxygenation for Respiratory Failure: Findings From the Extracorporeal Life Support Organization Database. Crit Care Med 2017;45:1389-1397.
21.Fletcher-Sandersjoo A, Bartek J, Jr., Thelin EP et al. Predictors of intracranial hemorrhage in adult patients on extracorporeal membrane oxygenation: an observational cohort study. J Intensive Care 2017;5:27.
22.Martucci G, Lo Re V, Arcadipane A. Neurological injuries and extracorporeal membrane oxygenation: the challenge of the new ECMO era. Neurol Sci 2016;37:1133-6.
23.Chow FC, Edlow BL, Frosch MP, Copen WA, Greer DM. Outcome in patients with H1N1 influenza and cerebrovascular injury treated with extracorporeal membrane oxygenation. Neurocrit Care 2011;15:156-60.
24.Lockie CJA, Gillon SA, Barrett NA et al. Severe Respiratory Failure, Extracorporeal Membrane Oxygenation, and Intracranial Hemorrhage. Crit Care Med 2017;45:1642-1649.
25.Mehta A, Ibsen LM. Neurologic complications and neurodevelopmental outcome with extracorporeal life support. World J Crit Care Med 2013;2:40-7.
26.Nasr DM RA. Neurologic complications of extracorporeal membrane oxygenation. J Clin Neurol 2015;11:383–9.
27.Schmidt M, Brechot N, Hariri S et al. Nosocomial infections in adult cardiogenic shock patients supported by venoarterial extracorporeal membrane oxygenation. Clin Infect Dis 2012;55:1633-41.
28.Hsu MS, Chiu KM, Huang YT, Kao KL, Chu SH, Liao CH. Risk factors for nosocomial infection during extracorporeal membrane oxygenation. J Hosp Infect 2009;73:210-6.

29.Burket JS, Bartlett RH, Vander Hyde K, Chenoweth CE. Nosocomial infections in adult patients undergoing extracorporeal membrane oxygenation. Clin Infect Dis 1999;28:828-33.
30.Aubron C, Cheng AC, Pilcher D et al. Infections acquired by adults who receive extracorporeal membrane oxygenation: risk factors and outcome. Infect Control Hosp Epidemiol 2013;34:24-30.
31.Sun HY, Ko WJ, Tsai PR et al. Infections occurring during extracorporeal membrane oxygenation use in adult patients. J Thorac Cardiovasc Surg 2010;140:1125-32 e2.
32.Grasselli G, Scaravilli V, Di Bella S et al. Nosocomial Infections During Extracorporeal Membrane Oxygenation: Incidence, Etiology, and Impact on Patients' Outcome. Crit Care Med 2017;45:1726-1733.
33.Touchard C, Aubry A, Eloy P et al. Predictors of insufficient peak amikacin concentration in critically ill patients on extracorporeal membrane oxygenation. Crit Care 2018;22:199.
34.Hahn J, Choi JH, Chang MJ. Pharmacokinetic changes of antibiotic, antiviral, antituberculosis and antifungal agents during extracorporeal membrane oxygenation in critically ill adult patients. J Clin Pharm Ther 2017;42:661-671.
35.Millar JE, Fanning JP, McDonald CI, McAuley DF, Fraser JF. The inflammatory response to extracorporeal membrane oxygenation (ECMO): a review of the pathophysiology. Crit Care 2016;20:387.
36.Frerou A, Lesouhaitier M, Gregoire M et al. Venoarterial extracorporeal membrane oxygenation induces early immune alterations. Crit Care 2021;25:9.
37.Spaan AN, Surewaard BG, Nijland R, van Strijp JA. Neutrophils versus Staphylococcus aureus: a biological tug of war. Annu Rev Microbiol 2013;67:629-50.
38.Jongbloed SL, Kassianos AJ, McDonald KJ et al. Human CD141+ (BDCA-3)+ dendritic cells (DCs) represent a unique myeloid DC subset that cross-presents necrotic cell antigens. J Exp Med 2010;207:1247-60.
39.Gey A, Tadie JM, Caumont-Prim A et al. Granulocytic myeloid-derived suppressor cells inversely correlate with plasma arginine and overall survival in critically ill patients. Clin Exp Immunol 2015;180:280-8.
40.Gabrilovich DI, Nagaraj S. Myeloid-derived suppressor cells as regulators of the immune system. Nat Rev Immunol 2009;9:162-74.
41.Grutz G. New insights into the molecular mechanism of interleukin-10-mediated immunosuppression. J Leukoc Biol 2005;77:3-15.

42.Burkhoff D, Sayer G, Doshi D, Uriel N. Hemodynamics of Mechanical Circulatory Support. J Am Coll Cardiol 2015;66:2663-2674.
43.Andrei S, Tran-Dinh A, Provenchere S et al. A quantified description of the interactions between the native cardiovascular system and femoro-femoral versus femoro-axillary extracorporeal life support using descending thoracic aorta velocity time integral. Artif Organs 2019;43:647-655.
44.Gu K, Zhang Y, Gao B, Chang Y, Zeng Y. Hemodynamic Differences Between Central ECMO and Peripheral ECMO: A Primary CFD Study. Med Sci Monit 2016;22:717-26.
45.Platts DG, Sedgwick JF, Burstow DJ, Mullany DV, Fraser JF. The role of echocardiography in the management of patients supported by extracorporeal membrane oxygenation. J Am Soc Echocardiogr 2012;25:131-41.
46.Boulate D, Luyt CE, Pozzi M et al. Acute lung injury after mechanical circulatory support implantation in patients on extracorporeal life support: an unrecognized problem. Eur J Cardiothorac Surg 2013;44:544-9; discussion 549-50.
47.Cevasco M, Takayama H, Ando M, Garan AR, Naka Y, Takeda K. Left ventricular distension and venting strategies for patients on venoarterial extracorporeal membrane oxygenation. J Thorac Dis 2019;11:1676-1683.
48.Petroni T, Harrois A, Amour J et al. Intra-aortic balloon pump effects on macrocirculation and microcirculation in cardiogenic shock patients supported by venoarterial extracorporeal membrane oxygenation*. Crit Care Med 2014;42:2075-82.
49.Gass A, Palaniswamy C, Aronow WS et al. Peripheral venoarterial extracorporeal membrane oxygenation in combination with intra-aortic balloon counterpulsation in patients with cardiovascular compromise. Cardiology 2014;129:137-43.
50.Vallabhajosyula S, O'Horo JC, Antharam P et al. Concomitant Intra-Aortic Balloon Pump Use in Cardiogenic Shock Requiring Veno-Arterial Extracorporeal Membrane Oxygenation. Circ Cardiovasc Interv 2018;11:e006930.
51.Li Y, Yan S, Gao S et al. Effect of an intra-aortic balloon pump with venoarterial extracorporeal membrane oxygenation on mortality of patients with cardiogenic shock: a systematic review and meta-analysis†. Eur J Cardiothorac Surg 2019;55:395-404.
52.Alhussein M, Osten M, Horlick E et al. Percutaneous left atrial decompression in adults with refractory cardiogenic shock supported with veno-arterial extracorporeal membrane oxygenation. J Card Surg 2017;32:396-401.

53.Aiyagari RM, Rocchini AP, Remenapp RT, Graziano JN. Decompression of the left atrium during extracorporeal membrane oxygenation using a transseptal cannula incorporated into the circuit. Crit Care Med 2006;34:2603-6.
54.Hlavacek AM, Atz AM, Bradley SM, Bandisode VM. Left atrial decompression by percutaneous cannula placement while on extracorporeal membrane oxygenation. J Thorac Cardiovasc Surg 2005;130:595-6.
55.Weymann A, Schmack B, Sabashnikov A et al. Central extracorporeal life support with left ventricular decompression for the treatment of refractory cardiogenic shock and lung failure. J Cardiothorac Surg 2014;9:60.
56.Rao P, Mosier J, Malo J et al. Peripheral VA-ECMO with direct biventricular decompression for refractory cardiogenic shock. Perfusion 2018;33:493-495.
57.Fumagalli R, Bombino M, Borelli M et al. Percutaneous bridge to heart transplantation by venoarterial ECMO and transaortic left ventricular venting. Int J Artif Organs 2004;27:410-3.
58.Schrage B, Becher PM, Bernhardt A et al. Left Ventricular Unloading Is Associated With Lower Mortality in Patients With Cardiogenic Shock Treated With Venoarterial Extracorporeal Membrane Oxygenation: Results From an International, Multicenter Cohort Study. Circulation 2020;142:2095-2106.
59.Watanabe S, Fish K, Kovacic JC et al. Left Ventricular Unloading Using an Impella CP Improves Coronary Flow and Infarct Zone Perfusion in Ischemic Heart Failure. J Am Heart Assoc 2018;7.
60.Russo JJ, Aleksova N, Pitcher I et al. Left Ventricular Unloading During Extracorporeal Membrane Oxygenation in Patients With Cardiogenic Shock. J Am Coll Cardiol 2019;73:654-662.
61.https://www.elso.org/Registry/Statistics.aspx.
62.Fux T, Holm M, Corbascio M, van der Linden J, Lund LH. Pre-Implant Outcome Predictors in Patients With Refractory Cardiogenic Shock Supported With VA-ECMO. J Am Coll Cardiol 2017;70:2094-2096.
63.Torre T, Toto F, Klersy C et al. Early predictors of mortality in refractory cardiogenic shock following acute coronary syndrome treated with extracorporeal membrane oxygenator. J Artif Organs 2021;24:327-335.
64.Huang M, Ong BH, Hoo AEE et al. Prognostic Factors for Survival After Extracorporeal Membrane Oxygenation for Cardiogenic Shock. ASAIO J 2020;66:141-145.

65.Smith M, Vukomanovic A, Brodie D, Thiagarajan R, Rycus P, Buscher H. Duration of veno-arterial extracorporeal life support (VA ECMO) and outcome: an analysis of the Extracorporeal Life Support Organization (ELSO) registry. Crit Care 2017;21:45.
66.Salna M, Takeda K, Kurlansky P et al. The influence of advanced age on venous-arterial extracorporeal membrane oxygenation outcomes. Eur J Cardiothorac Surg 2018;53:1151-1157.
67.Schmidt M, Burrell A, Roberts L et al. Predicting survival after ECMO for refractory cardiogenic shock: the survival after veno-arterial-ECMO (SAVE)-score. Eur Heart J 2015;36:2246-56.
68.Amin F, Lombardi J, Alhussein M et al. Predicting Survival After VA-ECMO for Refractory Cardiogenic Shock: Validating the SAVE Score. CJC Open 2021;3:71-81.
69.Wang L, Yang F, Wang X et al. Predicting mortality in patients undergoing VA-ECMO after coronary artery bypass grafting: the REMEMBER score. Crit Care 2019;23:11.
70.Muller G, Flecher E, Lebreton G et al. The ENCOURAGE mortality risk score and analysis of long-term outcomes after VA-ECMO for acute myocardial infarction with cardiogenic shock. Intensive Care Med 2016;42:370-378.
71.Wengenmayer T, Duerschmied D, Graf E et al. Development and validation of a prognostic model for survival in patients treated with venoarterial extracorporeal membrane oxygenation: the PREDICT VA-ECMO score. Eur Heart J Acute Cardiovasc Care 2019;8:350-359.
72.Aziz TA, Singh G, Popjes E et al. Initial experience with CentriMag extracorporal membrane oxygenation for support of critically ill patients with refractory cardiogenic shock. J Heart Lung Transplant 2010;29:66-71.
73.Aissaoui N, Luyt CE, Leprince P et al. Predictors of successful extracorporeal membrane oxygenation (ECMO) weaning after assistance for refractory cardiogenic shock. Intensive Care Med 2011;37:1738-45.
74.Cavarocchi NC, Pitcher HT, Yang Q et al. Weaning of extracorporeal membrane oxygenation using continuous hemodynamic transesophageal echocardiography. J Thorac Cardiovasc Surg 2013;146:1474-9.
75.Pappalardo F, Pieri M, Arnaez Corada B et al. Timing and Strategy for Weaning From Venoarterial ECMO are Complex Issues. J Cardiothorac Vasc Anesth 2015;29:906-11.
76.Huang KC, Lin LY, Chen YS, Lai CH, Hwang JJ, Lin LC. Three-Dimensional Echocardiography-Derived Right Ventricular Ejection Fraction Correlates with Success

of Decannulation and Prognosis in Patients Stabilized by Venoarterial Extracorporeal Life Support. J Am Soc Echocardiogr 2018;31:169-179.
77.Ling L, Chan KM. Weaning adult patients with cardiogenic shock on veno-arterial extracorporeal membrane oxygenation by pump-controlled retrograde trial off. Perfusion 2018;33:339-345.
78.Singh G, Hudson D, Shaw A. Medical Optimization and Liberation of Adult Patients From VA-ECMO. Can J Cardiol 2020;36:280-290.
79.Thomas M, Kreibich M, Beyersdorf F, Benk C, Maier S, Trummer G. Standardized Weaning from Temporary Extracorporeal Life Support in Cardiovascular Patients. Thorac Cardiovasc Surg 2020;68:425-432.
80.Tohme J, Piat C, Aissat N et al. Weaning-Related Shock in Patients With ECMO: Incidence, Mortality, and Predisposing Factors. J Cardiothorac Vasc Anesth 2021;35:41-47.
81.Richardson ASC, Tonna JE, Nanjayya V et al. Extracorporeal Cardiopulmonary Resuscitation in Adults. Interim Guideline Consensus Statement From the Extracorporeal Life Support Organization. ASAIO J 2021;67:221-228.
82.Sawada K, Kawakami S, Murata S et al. Predicting Parameters for Successful Weaning from Veno-Arterial Extracorporeal Membrane Oxygenation in Cardiogenic Shock. ESC Heart Fail 2021;8:471-480.
83.Lewis RP, Rittogers SE, Froester WF, Boudoulas H. A critical review of the systolic time intervals. Circulation 1977;56:146-58.
84.Denardo SJ, Nandyala R, Freeman GL, Pierce GL, Nichols WW. Pulse wave analysis of the aortic pressure waveform in severe left ventricular systolic dysfunction. Circ Heart Fail 2010;3:149-56.
85.Haiden A, Eber B, Weber T. U-shaped relationship of left ventricular ejection time index and all-cause mortality. Am J Hypertens 2014;27:702-9.
86.Ma TS, Paniagua D, Denktas AE et al. Usefulness of the Sum of Pulmonary Capillary Wedge Pressure and Right Atrial Pressure as a Congestion Index that Prognosticates Heart Failure Survival (from the Evaluation Study of Congestive Heart Failure and Pulmonary Artery Catheterization Effectiveness Trial). Am J Cardiol 2016;118:854-859.
87.Kim D, Park Y, Choi KH et al. Prognostic Implication of RV Coupling to Pulmonary Circulation for Successful Weaning From Extracorporeal Membrane Oxygenation. JACC Cardiovasc Imaging 2021;14:1523-1531.

88.Collier P, Phelan D, Klein A. A Test in Context: Myocardial Strain Measured by Speckle-Tracking Echocardiography. J Am Coll Cardiol 2017;69:1043-1056.
89.Brown J, Jenkins C, Marwick TH. Use of myocardial strain to assess global left ventricular function: a comparison with cardiac magnetic resonance and 3-dimensional echocardiography. . Am Heart J 2009;157:102.e1-5.
90.Mondillo S, Galderisi M, Mele D et al. Speckle-tracking echocardiography: a new technique for assessing myocardial function. J Ultrasound Med 2011;30:71-83.
91.Voigt JU, Pedrizzetti G, Lysyansky P et al. Definitions for a common standard for 2D speckle tracking echocardiography: consensus document of the EACVI/ASE/Industry Task Force to standardize deformation imaging. J Am Soc Echocardiogr 2015;28:183-93.
92.Grund FF, Kristensen CB, Myhr KA, Vejlstrup N, Hassager C, Mogelvang R. Layer-Specific Strain Is Preload Dependent: Comparison between Speckle-Tracking Echocardiography and Cardiac Magnetic Resonance Feature-Tracking. J Am Soc Echocardiogr 2021;34:377-387.
93.Nafati C, Gardette M, Leone M et al. Use of speckle-tracking strain in preload-dependent patients, need for cautious interpretation! Ann Intensive Care 2018;8:29.
94.Hahn RT, Abraham T, Adams MS et al. Guidelines for performing a comprehensive transesophageal echocardiographic examination: recommendations from the American Society of Echocardiography and the Society of Cardiovascular Anesthesiologists. J Am Soc Echocardiogr 2013;26:921-64.
95.Mandrekar JN. Receiver operating characteristic curve in diagnostic test assessment. J Thorac Oncol 2010;5:1315-6.
96.Jacob R, Dierberger B, Kissling G. Functional significance of the Frank-Starling mechanism under physiological and pathophysiological conditions. Eur Heart J 1992;13 Suppl E:7-14.
97.Dickstein ML. The Starling Relationship and Veno-Arterial ECMO: Ventricular Distension Explained. ASAIO J 2018;64:497-501.
98.Curran J, Burkhoff D, Kloner RA. Beyond Reperfusion: Acute Ventricular Unloading and Cardioprotection During Myocardial Infarction. J Cardiovasc Transl Res 2019;12:95-106.
99.Lang RM, Badano LP, Mor-Avi V et al. Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. Eur Heart J Cardiovasc Imaging 2015;16:233-70.

100.Lanspa MJ, Cirulis MM, Wiley BM et al. Right Ventricular Dysfunction in Early Sepsis and Septic Shock. Chest 2021;159:1055-1063.
101.Konstam MA, Abboud FM. Ejection Fraction: Misunderstood and Overrated (Changing the Paradigm in Categorizing Heart Failure). Circulation 2017;135:717-719.
102.Dupuis M, Mahjoub H, Clavel MA et al. Forward Left Ventricular Ejection Fraction: A Simple Risk Marker in Patients With Primary Mitral Regurgitation. J Am Heart Assoc 2017;6.
103.Baran DA, Grines CL, Bailey S et al. SCAI clinical expert consensus statement on the classification of cardiogenic shock: This document was endorsed by the American College of Cardiology (ACC), the American Heart Association (AHA), the Society of Critical Care Medicine (SCCM), and the Society of Thoracic Surgeons (STS) in April 2019. Catheter Cardiovasc Interv 2019;94:29-37.
104.Brookes CI, White PA, Staples M et al. Myocardial contractility is not constant during spontaneous atrial fibrillation in patients. Circulation 1998;98:1762-8.
105.Singh SSA, Dalzell JR, Berry C, Al-Attar N. Primary graft dysfunction after heart transplantation: a thorn amongst the roses. Heart Fail Rev 2019;24:805-820.
106.Gewillig M, Brown SC. The Fontan circulation after 45 years: update in physiology. Heart 2016;102:1081-6.
107.Hammoudi N, Watanabe S, Bikou O et al. Speckle-Tracking Echocardiographic Strain Analysis Reliably Estimates Degree of Acute LV Unloading During Mechanical LV Support by Impella. J Cardiovasc Transl Res 2019;12:135-141.
108.Pappalardo F, Schulte C, Pieri M et al. Concomitant implantation of Impella((R)) on top of veno-arterial extracorporeal membrane oxygenation may improve survival of patients with cardiogenic shock. Eur J Heart Fail 2017;19:404-412.
109.Tschope C, Van Linthout S, Klein O et al. Mechanical Unloading by Fulminant Myocarditis: LV-IMPELLA, ECMELLA, BI-PELLA, and PROPELLA Concepts. J Cardiovasc Transl Res 2019;12:116-123.
110.Kuchibhotla S, Esposito ML, Breton C et al. Acute Biventricular Mechanical Circulatory Support for Cardiogenic Shock. J Am Heart Assoc 2017;6.
111.Randles RH, Wolfe DA. Introduction to the Theory of Nonparametric Statistics. New York: John Wiley & Sons 1979.

APPENDIX

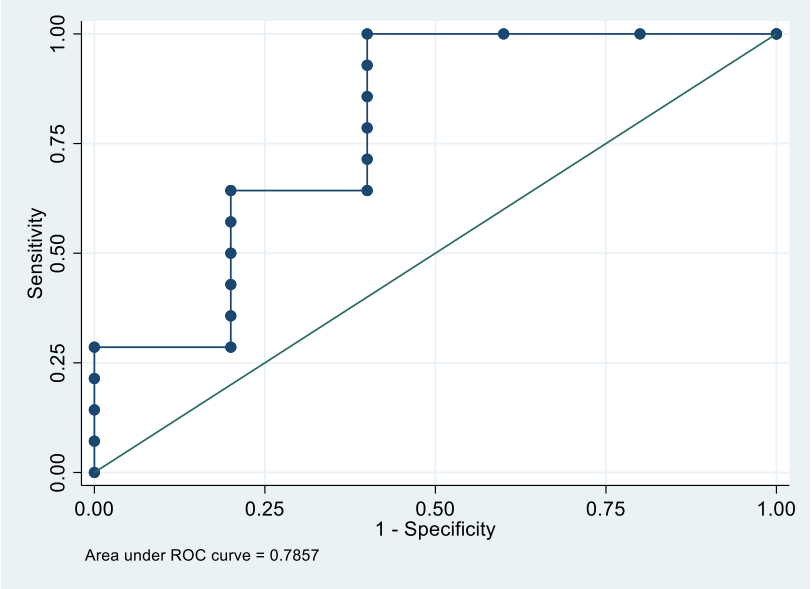
Outcome in variables Ejection Fraction, LVOT VTI, indexed stroke volume, cardiac index: **no-occurrence of clinical outcome.**

Outcome in variables systolic peak LV longitudinal strain, systolic peak RV free wall longitudinal strain, systolic peak circumferential strain: **occurrence of clinical outcome.**

Ejection fraction detailed report					
Cutpoint	**Sensitivity**	**Specificity**	**Classified**	**Likelihood ratio +**	**Likelihood ratio -**
(≥19.70..)	100.00%	0.00%	73.68%	1.0000	
(≥22.98..)	100.00%	20.00%	78.95%	1.2500	0.0000
(≥23.88..)	100.00%	40.00%	84.21%	1.6667	0.0000
(≥26.32..)	100.00%	60.00%	89.47%	2.5000	0.0000
(≥34.5574)	92.86%	60.00%	84.21%	2.3214	0.1190
(≥34.82..)	85.71%	60.00%	78.95%	2.1429	0.2381
(≥36.7835)	78.57%	60.00%	73.68%	1.9643	0.3571
(≥38.27..)	71.43%	60.00%	68.42%	1.7857	0.4762
(≥38.3675)	64.29%	60.00%	63.16%	1.6071	0.5952
(≥40.4086)	64.29%	80.00%	68.42%	3.2143	0.4464
(≥45.76..)	57.14%	80.00%	63.16%	2.8571	0.5357
(≥45.86..)	50.00%	80.00%	57.89%	2.5000	0.6250
(≥47.00..)	42.86%	80.00%	52.63%	2.1429	0.7143
(≥47.48..)	35.71%	80.00%	47.37%	1.7857	0.8036
(≥50.8221)	28.57%	80.00%	42.11%	1.4286	0.8929
(≥55.52..)	28.57%	100.00%	47.37%		0.7143
(≥65.0935)	21.43%	100.00%	42.11%		0.7857
(≥ 71.49..)	14.29%	100.00%	36.84%		0.8571
(≥78.50..)	7.14%	100.00%	31.58%		0.9286

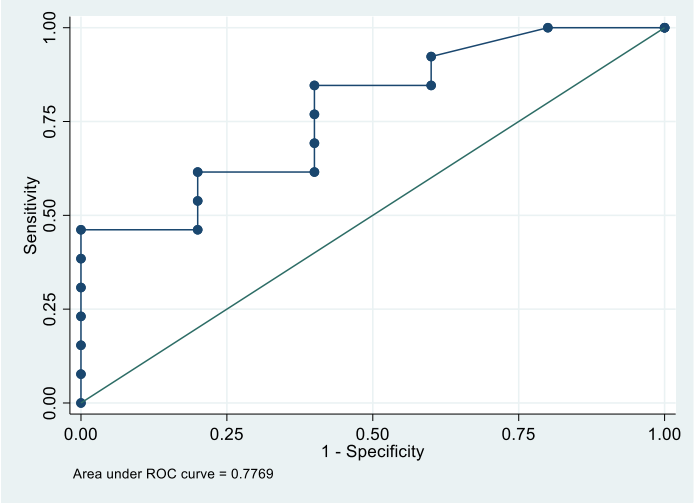
(> 78.50..)	0.00%	100.00%	26.32%		1.0000

ROC		Asymptotic Normal		
Observations	**Area**	**Standard Error**	**95% Confidence Interval**	
19	0.7857	0.1496	0.49257	1.00000



LVOT VTI detailed report					
Cutpoint	Sensitivity	Specificity	Classified	Likelihood ratio +	Likelihood ratio -
(≥11.53..)	100.00%	0.00%	72.22%	1.0000	
(≥12.83..)	100.00%	20.00%	77.78%	1.2500	0.0000
(≥13.1)	92.31%	40.00%	77.78%	1.5385	0.1923
(≥13.86..)	84.62%	40.00%	72.22%	1.4103	0.3846
(≥14.36..)	84.62%	60.00%	77.78%	2.1154	0.2564
(≥14.83..)	76.92%	60.00%	72.22%	1.9231	0.3846
(≥15.8)	69.23%	60.00%	66.67%	1.7308	0.5128
(≥15.93..)	61.54%	60.00%	61.11%	1.5385	0.6410
(≥16)	61.54%	80.00%	66.67%	3.0769	0.4808
(≥16.2)	53.85%	80.00%	61.11%	2.6923	0.5769
(≥16.3)	46.15%	80.00%	55.56%	2.3077	0.6731
(≥19.96..)	46.15%	100.00%	61.11%		0.5385
(≥21.03..)	38.46%	100.00%	55.56%		0.6154
(≥22.23..)	30.77%	100.00%	50.00%		0.6923
(≥22.5)	23.08%	100.00%	44.44%		0.7692
(≥26.26..)	15.38%	100.00%	38.89%		0.8462
(≥34.73..)	7.69%	100.00%	33.33%		0.9231
(>34.73..)	0.00%	100.00%	27.78%		1.0000

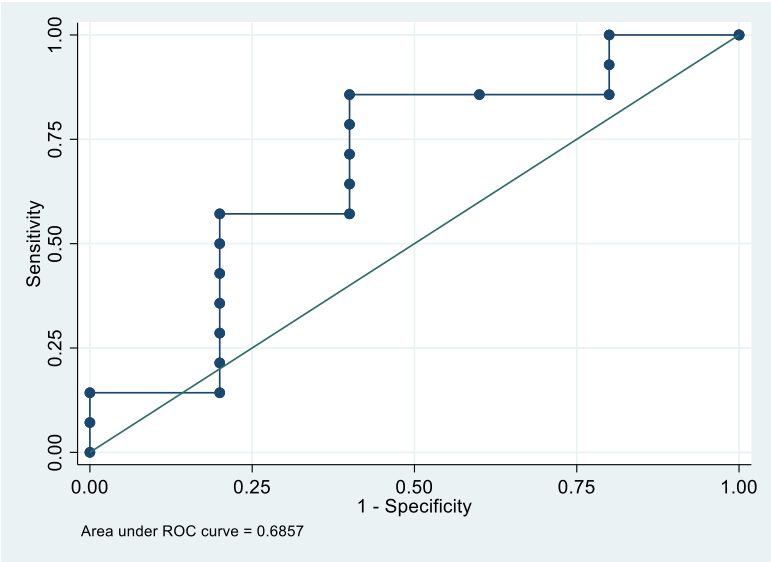
ROC		Asymptotic Normal		
Observations	Area	Standard Error	95% Confidence Interval	
18	0.7769	0.1249	0.53211	1.00000



Indexed Stroke Volume detailed report					
Cutpoint	**Sensitivity**	**Specificity**	**Classified**	**Likelihood ratio +**	**Likelihood ratio -**
(≥20.15..)	100.00%	0.00%	73.68%	1.0000	
(≥25.04..)	100.00%	20.00%	78.95%	1.2500	0.0000
(≥25.38..)	92.86%	20.00%	73.68%	1.1607	0.3571
(≥25.93..)	85.71%	20.00%	68.42%	1.0714	0.7143
(≥26.66..)	85.71%	40.00%	73.68%	1.4286	0.3571
(≥27.67..)	85.71%	60.00%	78.95%	2.1429	0.2381
(≥28.35..)	78.57%	60.00%	73.68%	1.9643	0.3571
(≥28.43)	71.43%	60.00%	68.42%	1.7857	0.4762
(≥28.48..)	64.29%	60.00%	63.16%	1.6071	0.5952
(≥28.6859)	57.14%	60.00%	57.89%	1.4286	0.7143
(≥31.89..)	57.14%	80.00%	63.16%	2.8571	0.5357
(≥32.84..)	50.00%	80.00%	57.89%	2.5000	0.6250
(≥35.11..)	42.86%	80.00%	52.63%	2.1429	0.7143
(≥35.54..)	35.71%	80.00%	47.37%	1.7857	0.8036
(≥41.25..)	28.57%	80.00%	42.11%	1.4286	0.8929
(≥42.94..)	21.43%	80.00%	36.84%	1.0714	0.9821

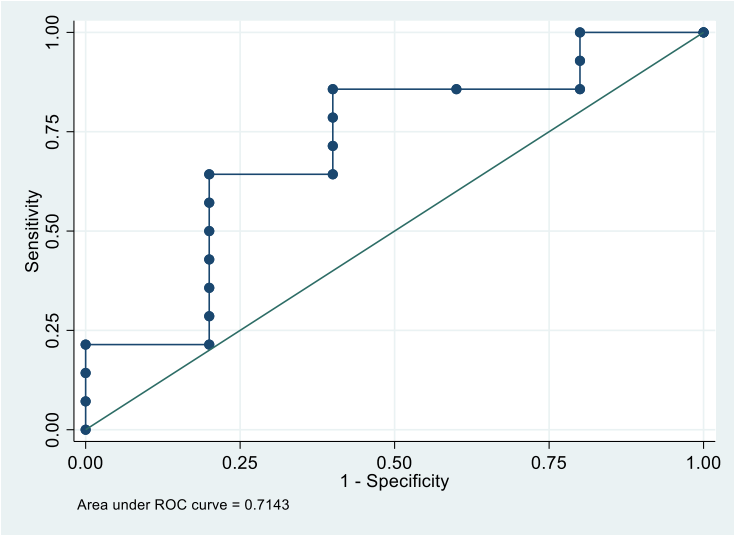
(≥49.61..)	14.29%	80.00%	31.58%	0.7143	1.0714
(≥56.3271)	14.29%	100.00%	36.84%		0.8571
(≥68.44..)	7.14%	100.00%	31.58%		0.9286
(>68.44..)	0.00%	100.00%	26.32%		1.0000

ROC		**Asymptotic Normal**		
Observations	**Area**	**Standard Error**	**95% Confidence Interval**	
19	0.6857	0.1659	0.360481	1.00000



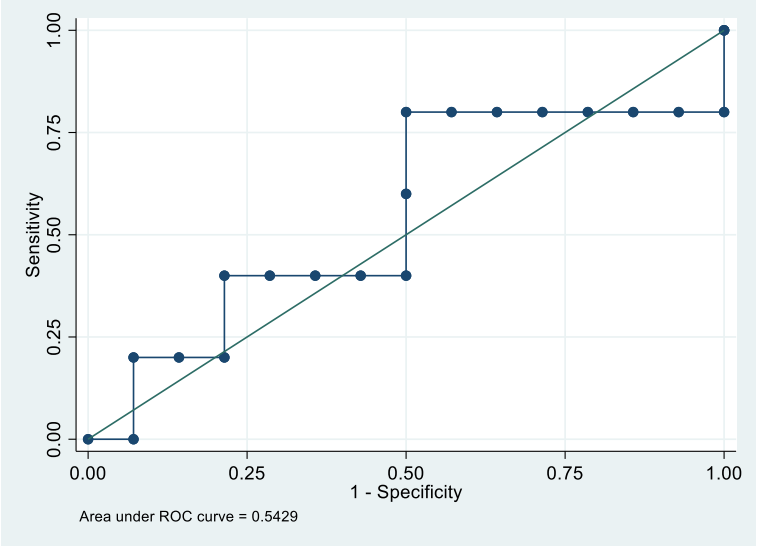
Cardiac Index detailed report					
Cutpoint	**Sensitivity**	**Specificity**	**Classified**	**Likelihood ratio +**	**Likelihood ratio -**
(≥1.786..)	100.00%	0.00%	73.68%	1.0000	
(≥2.184..)	100.00%	20.00%	78.95%	1.2500	0.0000
(≥2.205..)	92.86%	20.00%	73.68%	1.1607	0.3571
(≥2.264..)	85.71%	20.00%	68.42%	1.0714	0.7143
(≥2.310..)	85.71%	40.00%	73.68%	1.4286	0.3571
(≥2.504..)	85.71%	60.00%	78.95%	2.1429	0.2381
(≥2.56789)	78.57%	60.00%	73.68%	1.9643	0.3571
(≥2.714..)	71.43%	60.00%	68.42%	1.7857	0.4762
(≥2.91769)	64.29%	60.00%	63.16%	1.6071	0.5952
(≥3.101..)	64.29%	80.00%	68.42%	3.2143	0.4464
(≥3.270..)	57.14%	80.00%	63.16%	2.8571	0.5357
(≥3.371..)	50.00%	80.00%	57.89%	2.5000	0.6250
(≥3.527..)	42.86%	80.00%	52.63%	2.1429	0.7143
(≥3.62374)	35.71%	80.00%	47.37%	1.7857	0.8036
(≥3.702..)	28.57%	80.00%	42.11%	1.4286	0.8929
(≥4.219..)	21.43%	80.00%	36.84%	1.0714	0.9821
(≥4.403..)	21.43%	100.00%	42.11%		0.7857
(≥5.064..)	14.29%	100.00%	36.84%		0.8571
(≥5.465..)	7.14%	100.00%	31.58%		0.9286
(>5.465..)	0.00%	100.00%	26.32%		1.0000

	ROC	**Asymptotic Normal**		
Observations	**Area**	**Standard Error**	**95% Confidence Interval**	
19	0.7143	0.1536	0.41327	1.00000


Sensitivity
0.00
0.25
0.50
0.75
1.00
0.00
0.25
0.50
0.75
1.00
1 - Specificity
Area under ROC curve = 0.7143

Systolic peak longitudinal strain detailed report					
Cutpoint	Sensitivity	Specificity	Classified	LR +	LR -
(≥-19.216)	100.00%	0.00%	26.32%	1.0000	
(≥-16.8..)	80.00%	0.00%	21.05%	0.8000	
(≥-16.5..)	80.00%	7.14%	26.32%	0.8615	2.8000
(≥-15.1..)	80.00%	14.29%	31.58%	0.9333	1.4000
(≥-14.1..)	80.00%	21.43%	36.84%	1.0182	0.9333
(≥-12.6..)	80.00%	28.57%	42.11%	1.1200	0.7000
(≥-11.3..)	80.00%	35.71%	47.37%	1.2444	0.5600
(≥-10.805)	80.00%	42.86%	52.63%	1.4000	0.4667
(≥-10.7..)	80.00%	50.00%	57.89%	1.6000	0.4000
(≥-10.2..)	60.00%	50.00%	52.63%	1.2000	0.8000
(≥-9.8774)	40.00%	50.00%	47.37%	0.8000	1.2000
(≥-9.0454)	40.00%	57.14%	52.63%	0.9333	1.0500
(≥-8.4884)	40.00%	64.29%	57.89%	1.1200	0.9333
(≥-8.38..)	40.00%	71.43%	63.16%	1.4000	0.8400
(≥-7.77..)	40.00%	78.57%	68.42%	1.8667	0.7636
(≥-7.6913)	20.00%	78.57%	63.16%	0.9333	1.0182
(≥-7.6835)	20.00%	85.71%	68.42%	1.4000	0.9333
(≥-5.8978)	20.00%	92.86%	73.68%	2.8000	0.8615
(≥-5.02..)	0.00%	92.86%	68.42%	0.0000	1.0769
(>-5.02..)	0.00%	100.00%	73.68%		1.0000

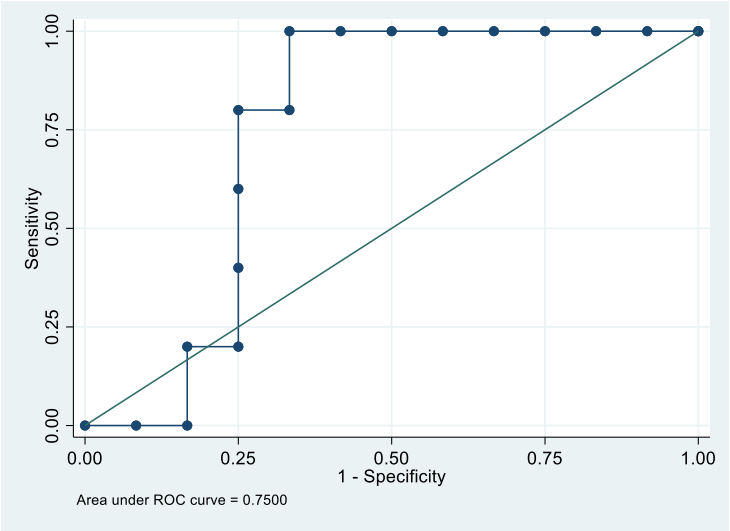
ROC		Asymptotic Normal		
Observations	Area	Standard Error	95% Confidence Interval	
19	0.5429	0.1767	0.19661	0.88910



Systolic peak RV free wall longitudinal strain detailed report					
Cutpoint	**Sensitivity**	**Specificity**	**Classified**	**Likelihood ratio +**	**Likelihood ratio -**
(≥-22.2..)	100.00%	0.00%	29.41%	1.0000	
(≥-21.0..)	100.00%	8.33%	35.29%	1.0909	0.0000
(≥-18.6..)	100.00%	16.67%	41.18%	1.2000	0.0000
(≥-18.4..)	100.00%	25.00%	47.06%	1.3333	0.0000
(≥-16.2..)	100.00%	33.33%	52.94%	1.5000	0.0000
(≥-16.1..)	100.00%	41.67%	58.82%	1.7143	0.0000
(≥-16.0..)	100.00%	50.00%	64.71%	2.0000	0.0000
(≥-14.7..)	100.00%	58.33%	70.59%	2.4000	0.0000
(≥-12.3..)	100.00%	66.67%	76.47%	3.0000	0.0000
(≥-12.312)	80.00%	66.67%	70.59%	2.4000	0.3000
(≥-12.0..)	80.00%	75.00%	76.47%	3.2000	0.2667
(≥-11.014)	60.00%	75.00%	70.59%	2.4000	0.5333
(≥-9.72..)	40.00%	75.00%	64.71%	1.6000	0.8000
(≥-9.01..)	20.00%	75.00%	58.82%	0.8000	1.0667
(≥-8.99..)	20.00%	83.33%	64.71%	1.2000	0.9600

(≥-8.98..)	0.00%	83.33%	58.82%	0.0000	1.2000
(≥-7.12..)	0.00%	91.67%	64.71%	0.0000	1.0909
(>-7.12..)	0.00%	100.00%	70.59%		1.0000

ROC		Asymptotic Normal		
Observations	**Area**	**Standard Error**	**95% Confidence Interval**	
17	0.7500	0.1238	0.50744	0.99256



Systolic peak circumferential strain detailed report					
Cutpoint	Sensitivity	Specificity	Classified	Likelihood ratio +	Likelihood ratio -
(≥-27.2..)	100.00%	0.00%	22.22%	1.0000	
(≥-23.6..)	100.00%	7.14%	27.78%	1.0769	0.0000
(≥-20.7..)	100.00%	14.29%	33.33%	1.1667	0.0000
(≥-19.3..)	100.00%	21.43%	38.89%	1.2727	0.0000
(≥-18.1..)	100.00%	28.57%	44.44%	1.4000	0.0000
(≥-17.0..)	75.00%	28.57%	38.89%	1.0500	0.8750
(≥-16.6..)	50.00%	28.57%	33.33%	0.7000	1.7500
(≥-16.4..)	50.00%	35.71%	38.89%	0.7778	1.4000
(≥-14.53)	50.00%	42.86%	44.44%	0.8750	1.1667
(≥-14.4..)	50.00%	50.00%	50.00%	1.0000	1.0000
(≥-13.945)	50.00%	57.14%	55.56%	1.1667	0.8750
(≥-13.578)	50.00%	64.29%	61.11%	1.4000	0.7778
(≥-10.7..)	50.00%	71.43%	66.67%	1.7500	0.7000
(≥-10.482)	50.00%	78.57%	72.22%	2.3333	0.6364
(≥-9.64)	50.00%	85.71%	77.78%	3.5000	0.5833
(≥-7.3382)	50.00%	92.86%	83.33%	7.0000	0.5385
(≥-6.1491)	50.00%	100.00%	88.89%		0.5000
(≥-5.1007)	25.00%	100.00%	83.33%		0.7500
(> -5.1007)	0.00%	100.00%	77.78%		1.0000

ROC		Asymptotic Normal		
Observations	**Area**	**Standard Error**	**95% Confidence Interval**	
18	0.6429	0.2155	0.22048	1.00000