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Brain death determination in patients with veno-arterial extracorporeal membrane oxygenation: A systematic study to address the Harlequin syndrome

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ABSTRACT

Purpose: The Harlequin syndrome may occur in patients treated with venoarterial extracorporal membrane oxygenation (VA-ECMO), in whom blood from the left ventricle and the ECMO system supply different parts of the body with different p_aCO_2-levels. The purpose of this study was to compare two variants of p_aCO_2-analysis to account for the Harlequin syndrome during apnea testing (AT) in brain death (BD) determination.
Materials and methods: Twenty-seven patients (median age 48 years, 26–76 years; male $n = 19$) with VA-ECMO treatment were included who underwent BD determination. In variant 1, simultaneous arterial blood gas (ABG) samples were drawn from the right and the left radial artery. In variant 2, simultaneous ABG samples were drawn from the right radial artery and the postoxygenator ECMO circuit. Differences in p_aCO_2-levels were analysed for both variants.
Results: At the start of AT, median p_aCO_2-difference between right and left radial artery (variant 1) was 0.90 mmHg (95%-confidence intervall [CI]: 0.7–1.3 mmHg). Median p_aCO_2-difference between right radial artery and postoxygenator ECMO circuit (variant 2) was 3.3 mmHg (95%-CI: 1.5–6.0 mmHg) and thereby significantly higher compared to variant 1 ($p = 0.001$). At the end of AT, p_aCO_2-difference according to variant 1 remained unchanged with 1.1 mmHg (95%-CI: 0.9–1.8 mmHg). In contrast, p_aCO_2-difference according to variant 2 increased to 9.9 mmHg (95%-CI: 3.5–19.2 mmHg; $p = 0.002$).
Conclusions: Simultaneous p_aCO_2-analysis from right and left distal arterial lines is the method of choice to reduce the risk of adverse effects (e.g. severe respiratory acidosis) while performing AT in VA-ECMO patients during BD determination.

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1. Introduction

Extra-corporeal life support (ECLS) has become an integral part of modern intensive medicine [1-3]. The widespread application of ECLS in recent years has also entailed new challenges for brain death (BD) determination [4-7]. This is most relevant for patients on veno-arterial extra-corporeal membrane oxygenation (VA-ECMO) [8-10]. VA-ECMO is a type of ECLS, which provides temporary mechanical circulatory support and extracorporeal gas exchange and has emerged as a salvage intervention in patients with cardiogenic shock and cardiac arrest refractory to standard therapies [11]. Within the last decade, the number of VA-ECMO treatments has increased markedly [12,13]. At the same time, the hospital mortality of patients treated with VA-ECMO has remained high with up to 66% [12]. Consistent with the underlying diagnoses, there is a relevant risk of BD among VA-ECMO patients, and the number of patients receiving BD diagnostics may continue to rise [4,14,15]. Therefore, it is essential to provide sound evidence for those diagnostic procedures within BD protocols that necessitate special regulations in VA-ECMO patients.

To determine BD, demonstration of absent spontaneous breathing by apnea testing (AT) is mandatory [16,17]. Performing AT during VA-ECMO requires special consideration of its effects on the distribution of blood flow and ABG tensions, depending of the site of ECMO cannulation and the extent of intrinsic cardiac function [8,9]. The Harlequin syndrome may occur in VA-ECMO-patients when left ventricular function starts to recover. Antegrade blood flow from the left ventricle may then collide with retrograde aortic perfusion delivered by the ECMO system, creating a “mixing cloud” [18,19]. If this “mixing cloud” is located in the aortic arch, the right arm and the right side of the head receive blood with higher p_aCO_2 and lower p_aO_2 from the left ventricle via the brachiocephalic trunk (Fig. 1). Meanwhile, the ECMO system supplies fully oxygenated and decarboxylated blood to the left arm and the left side of the head via the subclavian and common carotid arteries. In patients with maintained cerebral perfusion, this differential perfusion also affects the chemoreceptors of the medulla oblongata, which is supplied by the right and left vertebral arteries. To address the Harlequin syndrome, several guidelines have introduced recommendations on how to assess different p_aCO_2-levels in the right and left side of the body.

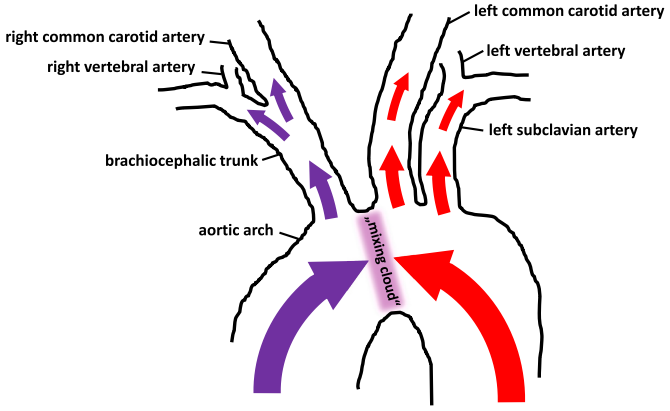

Fig. 1. Anatomical illustration of the Harlequin syndrome in the aortic arch. Arrows indicate intrinsic blood flow from the left ventricle (purple) and retrograde aortic perfusion delivered by the ECMO system (red), which collide in the aortic arch creating a “mixing cloud”. Consequently, the right arm and the right side of the head receive blood with higher p_aCO_2 and lower p_aO_2 from the left ventricle via the brachiocephalic trunk. Meanwhile, the ECMO system supplies fully oxygenated and decarboxylated blood to the left arm and the left side of the head via the subclavian and common carotid arteries. In patients with maintained cerebral perfusion, this differential perfusion also affects the chemoreceptors of the medulla oblongata, which is supplied by the right and left vertebral arteries. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

The main principle is to guide the AT by bilaterally simultaneous ABG analyses, which are drawn either from a right and left distal arterial line (variant 1) or from a right arterial line and the postoxygenator ECMO circuit (variant 2) [6,8,9,16,17,20]. To date, these recommendations are mainly based on pathophysiological considerations, while systematic validation is largely missing [8,21].

Here we report results of a comprehensive study testing the feasibility of simultaneous ABG analysis to guide AT. Moreover, we systematically compared ABG results from right and left distal arterial lines (variant 1) and ABG results from right arterial line and postoxygenator ECMO circuit (variant 2).

2. Materials and methods

The study was performed in five German university hospitals between 2020 and 2023. We retrospectively analysed patients who underwent BD determination while circulation was sustained by VA-ECMO. The clinical signs of BD (coma, loss of brain stem reflexes, central apnea) were ascertained by two physicians with extensive experience in the care of patients with severe brain injury. At least one investigator was a neurologist. AT was performed with the assistance of a critical care specialist trained in ECLS.

2.1. Apnoea testing

Before performing AT, arterial lines were placed as necessary to allow for simultaneous sampling from both radial arteries. At least five minutes of preoxygenation were provided by elevating the oxygen fraction of the mechanical ventilator and of the ECMO system to 100%. VA-ECMO flow rate and/or vasopressors were adjusted to maintain a mean arterial blood pressure of least 60 mmHg. Before AT was started, simultaneous ABG analyses were drawn from the right and left arterial lines (variant 1) or from the right arterial line and the postoxygenator ECMO circuit (variant 2). Ventilation and ECMO parameters were adjusted as necessary until p_aCO_2-levels between 35 and 45 mmHg were present in both samples. AT was then started by either (i) disconnecting the mechanical ventilator and providing intratracheal oxygen insufflation, (ii) setting the mechanical ventilator to continuous positive airway pressure mode with provision of 100% oxygen, or (iii) by providing 100% oxygen via a resuscitation bag with a positive end-expiratory pressure valve. The sweep gas flow rate of the ECMO system was then reduced to 0.5–1.0 L/min to increase p_aCO_2-levels. While assessing for spontaneous breathing, p_aCO_2 was monitored by repeated simultaneous ABG measures until p_aCO_2 increased to at least 60 mmHg in ABG samples from all sites. After completion of the AT, mechanical ventilation and ECMO were returned to their previous settings. BD was confirmed by an ancillary test.

2.2. Statistical analysis

The differences of individual p_aCO_2-levels between right and left arterial line (variant 1) and right arterial line and postoxygenator ECMO circuit (variant 2) were averaged across patients for the time AT was started and for the time AT was completed. Results are reported as median with 95%-confidence intervals (95%-CI) and ranges, or median with interquartile range (IQR) where appropiate. Mann–Whitney-*U* test was used to test for significant differences between variants 1 and 2 (with a significance level of 0.05), using IBM SPSS Statistics 29.0.1.0.

The study was approved by the institutional ethics review board *(Ethikkommission des Universitätsklinikums Freiburg, Germany, IRB-Nr. 22–1140)*. Informed consent was waived due to the retrospective nature of the data analysis. All procedures in this study were followed in accordance with the ethical standards of the 1964 Declaration of Helsinki.

3. Results

During the study period, 1.8% of all patients treated with VA-ECMO were introduced for BD determination. Twenty-seven patients (i.e., 1.4% of all VA-ECMO patients) received simultaneous p_aCO_2-analysis according to variant 1 or 2 during AT and consequently fulfilled the inclusion criteria of our study (median age 48 years, 26–76 years; male $n = 19$). All 27 patients were admitted due to cardiogenic shock or cardio-pulmonary resuscitation (CPR) requiring VA-ECMO. ECMO cannulation was performed via femoral arteries and veins in all patients, most often on the right side ($n = 22$; 81,5%). In five patients, VA-ECMO alone was not sufficient to stabilize hemodynamic parameters and additional percutaneous microaxial pumps (Impella® heart devices) were implanted to support ventricular function. The mean interval between admission and BD determination was 3.1 days (range 0–7 days). In the majority of patients (85.2%; $n = 23$) secondary brain injury (hypoxic encephalopathy) due to the initial cardiac or pulmonary emergency was the leading cause of BD. In three patients, intracerebral hemorrhage and subarachnoid hemorrhage (SAH) developed as complications of VA-ECMO implantation and led to BD. In the remaining patient, a primary SAH (grade V° according to World Federation of Neurological Surgeons [22]) led to neurogenic stunned myocardium and later BD. Table 1 summarizes demographic data, diagnosis, ECMO flow-rate, left ventricular ejection fraction, ancillary tests and the aetiologies leading to cardiogenic shock or CPR.

In 25 out of 27 patients, arterial lines were implanted in the right and left radial artery (variant 1). In five out of these 25 patients, simultaneous ABG samples were taken from both distal arterial lines and also from the postoxygenator ECMO circuit (combination of variants 1 and 2). In the two remaining patients, implantation of lines in the left distal arteries (radial or brachial artery) was not possible and ABG samples were taken simultaneously from right radial artery and postoxygenator ECMO circuit. Consequently, 25 data sets were analysed for variant 1 and seven data sets for variant 2.

In all patients ($n = 27$), simultaneous ABG analysis according to variant 1 or 2 or both was feasible to guide AT. There were no instances where AT had to be stopped due to cardiac, hemodynamic or respiratory instability and in no patient exogene CO_2 had to be supplied via the ECMO system to support hypercapnea. No negative results in AT were encountered, no patient started breathing during AT. BD was ultimately determined in all participants.

3.1. Simultaneous ABG analysis at the start of AT

At the start of AT, median p_aCO_2 of the right radial artery across all patients ($n = 27$) was 40.2 mmHg (IQR 37.1–42.1 mmHg, range 34.4–44.0 mmHg). With regard to variant 1 ($n = 25$), median baseline p_aCO_2 levels of right and left radial arteries were 40.3 mmHg and 39.5 mmHg (95%-CI: 38.9–41.1 and 38.6–40.9 mmHg; $p = 0.75$). Fig. 2 illustrates the initial distribution of individual p_aCO_2-differences according variant 1 and 2. The median difference at baseline between right and left radial p_aCO_2 was 0.90 mmHg (95%-CI: 0.7–1.3 mmHg, range 0.0–2.7 mmHg; Fig. 3). In variant 2 ($n = 7$), baseline p_aCO_2-levels in the right radial artery and in the postoxygenator ECMO circuit were 39.2 and 38.5 mmHg (95%-CI: 36.1–42.4 and 34.0–40.4 mmHg; $p = 0.40$). The median p_aCO_2-difference between right radial artery and postoxygenator ECMO circuit was 3.3 mmHg (95%-CI: 1.5–6.0 mmHg, range 0.7–7.5 mmHg; Fig. 3), significantly higher than in variant 1 ($p = 0.001$). With variant 1, higher baseline p_aCO_2-levels were found on the right side in 13 out of 25 patients, 11 patients had higher values on the left side, while the remaining patient showed equal p_aCO_2-levels on both sides (Fig. 2). With variant 2, all patients ($n = 7$) had higher p_aCO_2 in the right radial artery compared to the postoxygenator ECMO circuit (Fig. 2). In five patients, in whom ABG samples were taken simultaneously from all three sites, p_aCO_2-level was higher in the left radial artery than in the postoxygenator ECMO circuit (median p_aCO_2-difference 4.5 mmHg, range 0.1–7.2 mmHg). Here, median p_aCO_2-differences according to variant 1 and 2 were 1.0 and 3.4 mmHg (95%-CI: 0.1–2.8 and 0.3–7.8 mmHg).

Table 1
summarizes demographic data (sex, age), causes for VA-ECMO treatment, VA-ECMO flow rate, left ventricular ejection fraction (LVEF), cause of BD and mode of BD confirmation (type of ancillary test). *Abbreviations* used in the table are as follows: male (M), female (F), cardiopulmonary resuscitation (CPR), cardiogenic shock (CS), myocardial infarction (MI), pulmonary embolism (PE), ventricular fibrillation (VF), hypoxic encephalopathy (HE), intracerebral hemorrhage (ICH), subarachnoidal hemorrhage (SAE), perfusion scintigraphy (PS), somatosensory evoked potentials (SE), not assessed (n.a.).

Patient No.	Sex, age	Cause for VA-ECMO treatment	VA-ECMO flow rate	LVEF	Cause of BD	Ancillary test
1.	M, 51	CPR, asystolia, MI	3.5	10%	HE	EEG
2.	M, 39	CPR, PE	3.6	n.a.	HE	EEG
3.	M, 48	CPR, VF, MI	3.2	<5%	HE	EEG
4.	F, 76	CPR, VF, MI	3.5	n.a.	HE	EEG
5.	F, 38	CPR, PE	3.2	n.a.	HE	CT-angio
6.	M, 46	CPR, asystolia, MI	3.5	n.a.	HE	EEG
7.	M, 71	CPR, PE	3.8	<5%	HE	CT-Angio
8.	M, 46	CPR, asystolia	2.6	25%	HE	EEG
9.	M, 27	CPR, VF	3.8	<5%	HE	EEG, PS, TCD
10.	F, 32	CPR, drowning	4.5	n.a.	HE	EEG
11.	M, 52	CPR, MI	3.5	<5%	HE	EEG
12.	M, 48	CPR, MI	3.3	<5%	HE	EEG
13.	F, 34	CPR, VF	2.0	25%	HE	EEG
14.	M, 36	CPR, VF	2.5	35%	HE	EEG
15.	F, 75	CPR, cardiac arrest	3.6	n.a.	HE, ICH SAH	EEG
16.	M, 58	CPR, PE	4.0	35%	HE	EEG
17.	M, 58	CPR, asystolia	2.6	n.a.	HE	EEG, SEP, PS
18.	M, 26	CPR, asystolia, drowning	3.4	15%	HE	EEG
19.	M, 56	CPR, VF	3.4	<5%	HE	EEG
20.	M, 49	CS, MI	4.7	10%	HE	EEG
21.	M, 44	CPR, MI	3.2	n.a.	HE	EEG
22.	M, 53	CPR, stunned myocardium	5.6	5%	SAH	EEG
23.	F, 36	CPR, PE	4.3	n.a.	HE	EEG
24.	M, 57	CPR, PE	3.5	60%	ICH, SAH	EEG
25.	M, 43	CPR, MI	n.a.	n.a.	HE, ICH	EEG
26.	F, 69	CS, MI	2.0	10%	HE	2nd clin. Exam
27.	F, 28	CPR, PE	3.5	n.a.	HE, ICH	EEG

3.2. Simultaneous ABG analysis at the end of AT

At the end of AT, p_aCO_2 of the right radial artery across all patients was 67.1 mmHg (IQR 64.8–72.4 mmHg, range 60.4–94.6 mmHg). With variant 1, p_aCO_2-levels in the right and left radial arteries were 66.6 and 68.2 mmHg (95%-CI: 66.3–72.5 and 66.8–72.8 mmHg; $p = 0.81$). Fig. 2b illustrates the final distribution of all individual p_aCO_2-differences

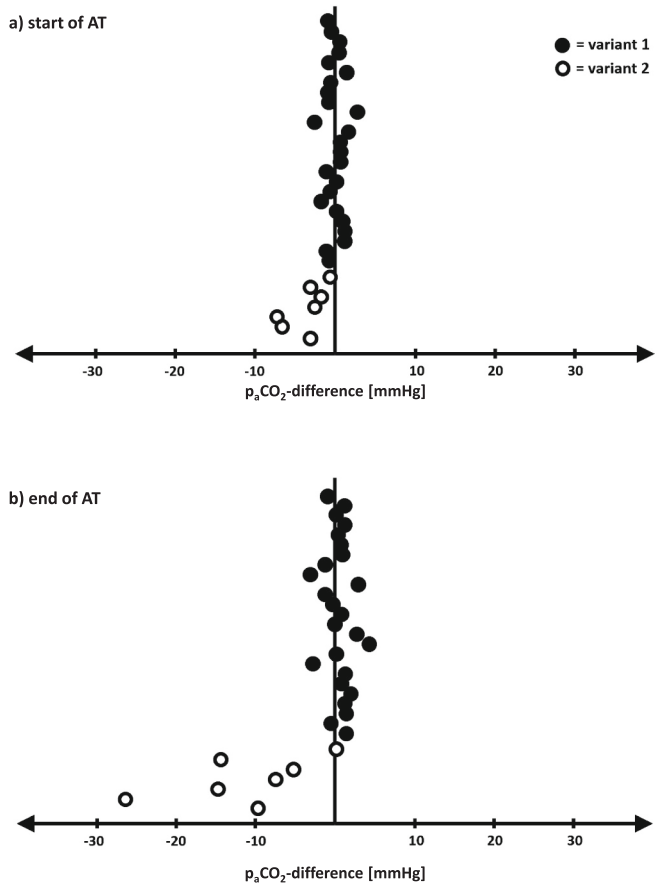

Fig. 2. a–b. Distribution of individual p_aCO_2-differences as illustrated by points (variant 1) and circles (variant 2) for each patient. Points and circles on the left side of the x-axis represent patients in whom p_aCO_2-values were higher on the right radial artery as compared to either the left radial artery (variant 1) or the postoxygenator ECMO circuit (variant 2), and vice versa. Fig. 2 illustrates individual p_aCO_2-differences at the start of apnea testing ($n = 25$ for variant 1 as illustrated by points; $n = 7$ for variant 2 as illustrated by circles). Fig. 2b illustrates individual p_aCO_2-differences at the end of apnea testing.

according variant 1 and 2. The median p_aCO_2-difference for variant 1 was 1.1 mmHg (95%-CI: 0.9–1.8 mmHg, range 0.1–4.2 mmHg; Fig. 3). With regard to variant 2, final p_aCO_2-levels in the right radial artery and postoxygenator ECMO circuit were 74.8 and 60.9 mmHg (95%-CI: 64.1–84.1 and 59.7–65.8 mmHg; $p = 0.04$). Here, the median p_aCO_2-difference was 9.9 mmHg (95%-CI: 3.5–19.2 mmHg, range 0.1–26.7 mmHg; Fig. 3), significantly higher than in variant 1 ($p = 0.002$). Sixteen out of 25 patients in whom variant 1 was applied had slightly higher p_aCO_2-levels on the left as compared to the right side (Fig. 2b). In variant 2, again all patients ($n = 7$) showed higher p_aCO_2-levels in the right radial artery compared to the postoxygenator ECMO (Fig. 2b). In five patients, in whom ABG samples were taken simultaneously from all three sites median p_aCO_2-difference between left radial artery and postoxygenator ECMO circuit was 11.4 mmHg (range 0.6–27.3 mmHg) and in each case p_aCO_2 in the left radial artery was higher than in the postoxygenator ECMO circuit, p_aCO_2-differences according to variant 1 and 2 reached 1.8 and 14.7 mmHg (95%-CI: −2.3–14.0 and − 0.4–25.1 mmHg).

Family members of nine patients agreed for organ donation, which was eventually realized in six cases (22.2% of all patients included in this.

4. Discussion

In this study, we systematically analysed two different procedures addressing the Harlequin syndrome during AT in patients treated with VA-ECMO. As suggested by earlier reviews, the current 5th version of the German guideline advises simultaneous monitoring of p_aCO_2 from distal arterial lines in both arms (variant 1) [8,9,17]. Authors from the recently published *American Brain Death/Death by Neurological Criteria Consensus Guideline 2023* and the *World Brain Death Project* propose taking simultaneous ABG samples from a right distal arterial line and from the postoxygenator ECMO circuit (variant 2) [16,20]. In our study, we systematically compared both variants, including their ability to ensure that specified p_aCO_2-thresholds are met during AT.

Simultaneous ABG sampling proved feasible to guide AT according to both variants. As the central finding, we demonstrate that p_aCO_2 in the postoxygenator ECMO circuit does not closely reflect p_aCO_2 in the left radial artery. We found a significant overestimation of the Harlequin effect by a factor of >3 already during normocapnic baseline (p_aCO_2-

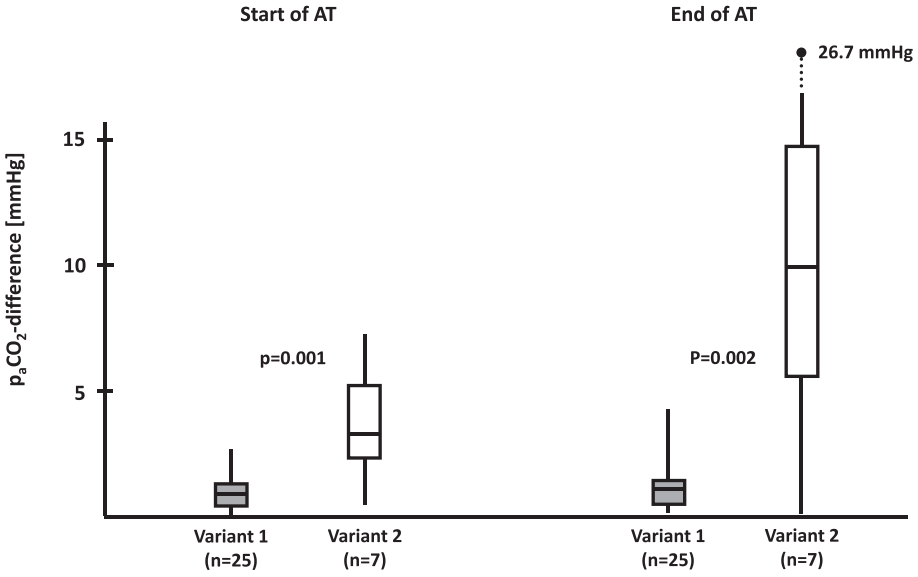

Fig. 3. Comparison of p_aCO_2-differences as illustrated by box-plots with median, interquartile ranges and ranges according to variant 1 (grey box) and variant 2 (white box) at the start of apnea testing and end of apnea testing. At both time-points, p_aCO_2-differences according to variant 2 (simultaneous ABG samples from right radial artery and postoxygenator ECMO circuit) were significantly higher as compared to variant 1 (simultaneous ABG samples from right and left radial arteries).

differences of variant 1 vs. 2 were 0.90 vs. 3.3 mmHg; $p = 0.001$). At the end of the AT, however, this factor increased to an average of 9 (p_aCO_2-differences of variant 1 vs. 2 were 1.1 vs. 9.9 mmHg; $p = 0.002$). Here individual p_aCO_2-differences reached up to 26.7 mmHg. Similar results were reported in a series of three patients, with a p_aCO_2-difference between a right distal arterial line and the postoxygenator ECMO circuit of 9, 18, and 35 mmHg at the end of AT [21]. As a practical consequence, applying a p_aCO_2-threshold of 60 mmHg in the postoxygenator ECMO circuit may prolong the apneic phase by several minutes, thereby exposing the patient to an unnecessarily severe respiratory acidosis. In the mentioned example of our study, a p_aCO_2 well above 85 mmHg was documented in the right radial artery when postoxygenator p_aCO_2 reached 60 mmHg. Respiratory acidosis endangers the through an increased risk of cardiac arrhythmia and systemic hypotension.

AT requires demonstration of apnea when the p_aCO_2 has increased above a defined threshold. The underlying assumption is that measured ABG values are representative of the p_aCO_2-level at the chemoreceptors of the medulla oblongata, which trigger spontaneous breathing. In patients on VA-ECMO, the concern is that antegrade and retrograde flow from the left ventricle and ECMO system, containing blood with different p_aCO_2-levels, may create an unpredictable mixing cloud in the aorta (Fig. 1). If located in the aortic arch, diverging p_aCO_2-levels in the right arm, medulla oblongata, left arm, and ECMO output may result. From a hypothetical point of view, the p_aCO_2 in the mixing zone cannot be lower than the p_aCO_2 minimum of the inputs. Thus, demonstrating above-threshold p_aCO_2 in the postoxygenator ECMO circuit and in the right radial artery (variant 2) is a plausible approach to guarantee above-threshold p_aCO_2 also in the medulla oblongata. However, if the mixing zone is located distal to the aortic arch, p_aCO_2 in the left subclavian, vertebral and radial arteries will be closer to that in the right radial artery than in the ECMO output, meaning that p_aCO_2 in the vertebral arteries and medulla oblongata should reach the target level earlier than p_aCO_2 in the postmembrane ECMO circuit. As reflected by the difference in p_aCO_2 between left radial artery and postoxygenator ECMO circuit, the mixing zone may often be located in the descending aorta. In our study, simultaneous ABG samples revealed a maximal p_aCO_2-difference of up to 27.3 mmHg between the left radial artery and the postoxygenator ECMO circuit at the end of AT. Based on vascular anatomy, p_aCO_2 in the left vertebral artery should closely follow p_aCO_2 in the left radial artery, regardless of the location of the mixing zone.

Independent of VA-ECMO, the safety of AT has been a matter of critical debate since the introduction of diagnostic protocols and guidelines for BD determination [23-25]. Systematic studies of cardiopulmonary parameters, cerebral hemodynamics, and invasive neuromonitoring indicate that AT is safe if performed strictly according to guideline recommendations [26,27]. In ECMO-patients, a low to moderate complication rate of AT has been reported. A recent review summarized 17 case studies and case series ($n = 67$) [6]. Here, AT was interrupted due to hemodynamic or respiratory instability in 9% of patients [21,28,29]. In our patients, results obtained with variant 1, variant 2, or both successfully guided AT without negative impact on cardiac, hemodynamic or respiratory stability. Interruption of AT was never required. To reach target p_aCO_2 while on VA-ECMO, gas flow at the oxygenator membrane is reduced to decrease CO_2 elimination. In patients with compromised alveolar diffusion, a high oxygen fraction may be required, limiting the achievable reduction in gas flow and CO_2 washout. As a workaround, adding CO_2 to the VA-ECMO gas mix has been reported in four patients [30]. Information regarding bilateral ABG sampling was not provided in the mentioned study. Based on the physiological considerations above, false-positive findings are unlikely with this method. In our study, CO_2 supplementation was not necessary.

In view of its high inaccuracy and potential risks from excessive hypercapnia, we recommend against guiding AT by applying the p_aCO_2-threshold to the posytoxygenator ECMO circuit. Instead, AT should be guided by p_aCO_2 in distal arteries of the left and right arm, whenever possible. According to previous reports by the *Extracorporeal Life Support Organization* providing data from different countries up to 7.9% of VA-ECMO patients developed BD [31]. In our study, only 1.8% of all patients treated with VA-ECMO were introduced for BD determination. This discrepancy may partly be explained by a decrease in the prevalence of acute brain injury during VA-ECMO treatment in recent years while the total number of VA-ECMO treatments has increased [32]. Another reason may be a persistent uncertainty on how to deal with challenging aspects of BD determination in patients with VA-ECMO. Our study aims to reduce this uncertainty by providing more evidence-based recommendations on how to perform the AT safely.

For future research, we recommend to continue simultaneous ABG sampling from both arms and the ECMO output to further investigate which ECMO-related parameters, echocardiography results or ABG constellations may predict the size of the p_aCO_2-differences during AT. These results could help identify situations in which a second arterial line may not be required.

4.1. Limitations of the study

Although, our study might incorporate the highest number of patients included in a study to address the Harlequin syndrome in BD determination as by now, the overall number of patients is still low. Simultaneous p_aCO_2 values from both right and left radial artery as well as postoxygenator blood gas were only available in five patients, hence majority of the data is describing interpatient comparisons. The retrospective design of our study does incorporate some methodological limitations. In fact, the retrospective design of our study, did not allow providing valid investigation of the time, which was needed to complete AT. Time between first and last ABG sampling does not necessarily reflect the actual times of the AT. Future studies should analyze *"time of AT"* as well as *"pH-changes"* prospectively.

The main objective of our study was to test the feasibility of simultaneous ABG sampling from both arms and/or the ECMO circuit to address the impact of a Harlequin syndrome. Objective visualization of the mixing cloud associated with the Harlequin syndrome itself was not part of our study. The manifestation of the Harlequin syndrome is dynamic and localization of the watershed may change across the aortic segments over time. CT-angiography may visualize the watershed across the aortic arch or the descending aorta. However, transportation of highly unstable ECMO-patients is hazardous, which was also a reason why CT-imaging was not performed in our patients for study purposes. For future studies, contrast-enhanced ultrasound might provide an easy-to-use bedside modality helping to identify patients at risk for a Harlequin syndrome.

5. Conclusions

Since further increase of ECLS utilization is expected, adequate knowledge about the specific challenges of BD determination in VA-ECMO patients is indispensable [4]. Based on our data, ABG samples taken simultaneously from right and left distal arterial lines should be the first choice in performing AT in VA-ECMO patients. If implantation of a second arterial line has failed, simultaneous ABG samples taken from the right radial artery and the postoxygenator ECMO circuit are an alternative that, however, may require more time for completion of AT.

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Authors' contributions

All authors contributed substantially to the conception and design of the study. Each author performed BD determination in patients included in this study and contributed to the acquisition of data. All authors

contributed substantially to the interpretation of the data. The first draft of the manuscript was written by the corresponding author F.S. and all authors commented on previous versions of the manuscript. All authors provided final approval of the version submitted for publication.

CRediT authorship contribution statement

Farid Salih: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Project administration, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Johann Lambeck:** Writing – review & editing, Validation, Methodology, Data curation. **Albrecht Günther:** Writing – review & editing, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Caroline Ferse:** Writing – review & editing, Methodology, Data curation, Conceptualization. **Olaf Hoffmann:** Writing – review & editing, Visualization, Methodology, Investigation, Data curation, Conceptualization. **Konstantinos Dimitriadis:** Writing – review & editing, Methodology, Investigation, Formal analysis, Data curation. **Andre Finn:** Writing – review & editing, Methodology, Investigation. **Stephan A. Brandt:** Writing – review & editing, Methodology, Investigation. **Benjamin Hotter:** Writing – review & editing, Methodology, Investigation. **Stephan Schreiber:** Writing – review & editing, Methodology, Investigation. **Florian Weissinger:** Writing – review & editing, Methodology, Investigation. **Andrea Rocco:** Writing – review & editing, Methodology, Investigation. **Hauke Schneider:** Writing – review & editing, Methodology, Investigation. **Wolf-Dirk Niesen:** Writing – review & editing, Supervision, Methodology, Investigation, Conceptualization.

Declaration of competing interest

None.

Data availability

The datasets used and analysed during the current study are available from the corresponding author on reasonable request.

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