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#### **ORIGINAL ARTICLE**



# Physiology of repeated mixed gas 100-m wreck dives using a closed-circuit rebreather: a field bubble study

Costantino Balestra 1,2,5 • François Guerrero • Sigrid Theunissen • Peter Germonpré 1,3,5 • Pierre Lafère 1,3,4,5 •

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#### Abstract

**Purpose** Data regarding decompression stress after deep closed-circuit rebreather (CCR) dives are scarce. This study aimed to monitor technical divers during a wreck diving expedition and provide an insight in venous gas emboli (VGE) dynamics. **Methods** Diving practices of ten technical divers were observed. They performed a series of three consecutive daily dives around 100 m. VGE counts were measured 30 and 60 min after surfacing by both cardiac echography and subclavian Doppler graded according to categorical scores (Eftedal–Brubakk and Spencer scale, respectively) that were converted to simplified bubble grading system (BGS) for the purpose of analysis. Total body weight and fluids shift using bioimpedancemetry were also collected pre- and post-dive.

**Results** Depth-time profiles of the 30 recorded man-dives were  $97.3 \pm 26.4$  msw [range: 54-136] with a runtime of  $160 \pm 65$  min [range: 59-270]. No clinical decompression sickness (DCS) was detected. The echographic frame-based bubble count par cardiac cycle was  $14 \pm 13$  at 30 min and  $13 \pm 13$  at 60 min. There is no statistical difference neither between dives, nor between time of measurements (P=0.07). However, regardless of the level of conservatism used, a high incidence of high-grade VGE was detected. Doppler recordings with the O'dive were highly correlated with echographic recordings (Spearman r of 0.81, P=0.008).

**Conclusion** Although preliminary, the present observation related to real CCR deep dives questions the precedence of decompression algorithm over individual risk factors and pleads for an individual approach of decompression.

 $\textbf{Keywords} \ \ \text{Bubble detection} \cdot \text{Bubbles} \cdot \text{Decompression} \cdot \text{Technical diving} \cdot \text{Risk assessment} \cdot \text{Echocardiography} \cdot \text{Diving} \cdot \text{Doppler} \cdot \text{CCR}$ 

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#### **Abbreviations**

BGS Simplified bubble grading system

CCR Closed-circuit rebreather

DAN Divers alert network

DCS Decompression sickness

ECW Extracellular water

ICW Intracellular water

OC Open circuit

PPO<sub>2</sub> Partial pressure of oxygen

TBW Total body water

VGE Vascular gas emboli

#### Introduction

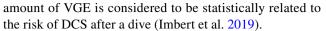
Although, there is no universally accepted definition, the popularity of "technical diving" has increased in the diving community over the last 20 years (Mitchell and Doolette 2013). This term encompasses all kind of diving beyond



the generally accepted limits of 40 m for recreational diving. According to the NOAA, it includes deep (in the range of 50–140 m, sometimes even deeper), decompression, mixed-gas, and multi-gas diving for non-professional purpose (Mixed-gas diving 2017).

These deeper and longer dives translate into an increased risk of decompression sickness (DCS) (Lundell et al. 2019; Peddie and Watson 2020). Indeed, technical divers use decompression tools, whose functioning is based on predictive models that are made operational through algorithms whose aim is to deal with the gas and vascular gas emboli (VGE) load. However, most of these models, simple or complex, owe their functioning not so much to an underlying theory, but to data fitting (Angelini 2018). Indeed, validated decompression procedures are the results of human dive trial in which both risks factors for DCS, and dive outcomes are well-documented. Then, if the dive profile falls within the range of operational validity of a specific model, the resulting decompression schedule may be used with a fair degree of safety. This is, however, not the case for technical divers as depths and decompression schedules are well outside of the range of any testing. Even more, many modifications to the decompression schedule, implemented by technical divers, such as gradient factors or deep stops, have not been validated by any human dive trials (Doolette and Mitchell 2013). Although several thousand dives are reported to have been conducted safely in the field, one cannot assume a similar degree of confidence without proper data gathering (Imbert et al. 2019; Buzzacott et al. 2014), It has indeed already been demonstrated that technical diving in open circuit up to 70 m is associated with a high decompression stress (Ljubkovic et al. 2010).

However, within technical diving, one of the most important emerging trends is the shift from open circuit diving to close-circuit rebreather (CCR) diving, which is unfortunately associated with a tenfold increased fatality rate compared to open circuit (OC) scuba diving (Fock 2013). Besides the complexity of CCRs, which means that they are more prone to failure than OC equipment, the lack of formal detailed dive planning facilitated by the availability of continuous decompression solutions produced by decompression computers might also explain these figures (Fock 2006). These are only hypothesis as available medical literature about CCR diving is scarce. Recently, one study was published on the oxidative stress caused by hyperoxic CCR diving (Bosco et al. 2018), but not much is known about VGE dynamics, which is a universally accepted predisposing condition for DCS (Wienke 2009). Indeed, regardless of the decompression algorithm used, detectable VGE may be measured after recreational, technical and (to a lesser degree) professional diving (Dunford et al. 2002). The



Therefore, contributions of organizations like divers alert network (DAN) may help to better understand the consequences on the decompression stress of these extrapolations outside of the tested range. Recently, a team of researchers from DAN Europe participated in a technical diving expedition to the Red Sea whose goal was to dive unusual wrecks in the Gulf of Suez (SS Zealote—110 m of sea water (msw) or SS Dacca—125 msw for instance). Although most of the planned dives would have been placed in the "extreme exposure" category in the DCIEM decompression tables, it was not the purpose of the research team to recommend any specific diving procedure. This study only aimed to monitor divers and provide an insight of their post-dive physiological status and VGE dynamics.

#### **Methods**

The diving expedition took place from 13 to 20 March 2020, aboard the Red Sea Explorers liveaboard MV Nouran. This observational study was conducted in accordance with the Declaration of Helsinki. Ethics approval was obtained from the Bio-Ethical Committee for Research and Higher Education, Brussels (No B200-2020-088). Prior to departure, informed written consent was sought from the ten divers participating in the expedition. All divers were assessed fit to dive by their own diving medicine specialist prior to the expedition.

#### **Diving procedures**

Dives were conducted from a 36-m dive boat based out of Hurghada—Egypt. On-board facilities included continual blending of nitrox and trimix gas mixes and pure oxygen (O<sub>2</sub>). Due to the outbreak of the COVID-19 pandemic, all planned dives could not be achieved. Since no adequate medical support could have been provided in case of any diving related problem, it was decided to stop diving according to DAN Europe directions and European Committee for Hyperbaric Medicine (ECHM) guidelines. Therefore, the dives were conducted over a 5-day period only, with two dives a day (30-65 msw) with a surface interval of 3-4 h between dives on day 1 and 2, followed by 3 consecutive days with a single dive around 100 msw deep or beyond (Day 3: 64 msw; Day 4: 98 msw and Day 5: 123 msw). The first 2 days were used to accustom the whole diving team and boat crew to scientific procedures and not full data collection allowing us to perfect the installation work, but also to allow divers to fine tune their set-ups and procedures.

Typically, a dive briefing was conducted before the dive on each of six wreck sites. Then, divers were dropped



directly over the wreck. Weather conditions were good with daily air temperature between 25 and 30 °C, and surface water temperature between 24 and 28 °C. All dives were conducted with the help of underwater scooters.

During the dive, divers relied on their dive computers (Shearwater Petrel dive computers) with build-in ZHL-16C algorithm, to incorporate the readings from the CCR O<sub>2</sub> cells to calculate decompression requirements. However, based on experience and utilization of dive-planning software, most divers had a fair idea of the required total dive time and final stop time for a given bottom time. Each diver had the liberty to personalize their own decompression schedule as desired, by selecting a different 'conservatism' by means of gradient factors (GF), determining maximum allowed supersaturation limits for the deep and shallow parts of the dive. Therefore, little formal dive planning was performed, and participants, although in team, essentially dived 'solo'. In general, bottom time was planned on estimated decompression obligation rather than gas requirements.

All divers used JJ-CCRs rebreather (JJ-CCR ApS, Presto, Denmark) equipped with an integrated multi-gas decompression computer Petrel 2 (Shearwater Research Inc., Richmond, BC, Canada). A detailed description of the functioning of these units is beyond the scope of this article but interested readers may referred to the review by Walker and Murphy-Lavoie (2021). Basically, with a CCR, the exhaled gas is scavenged, scrubbed of carbon dioxide, enriched with oxygen, and then sent back to the diver in the breathing loop. The oxygen consumed by the diver via metabolic consumption is replaced to maintain a selected partial pressure of O<sub>2</sub> (PPO<sub>2</sub>). All divers used a PPO<sub>2</sub> set point of 1.3 ATA at depth and for ascent, then, for most of the divers, this was manually increased when arriving at a depth of 6 msw to be between 1.5 and 1.6 ATA (i.e., breathing 100% O<sub>2</sub>). Diluent gas for all 100 msw dives and beyond was trimix 10/70 (O<sub>2</sub>) 10%, helium 70%, nitrogen 20%). All gas compositions were verified using a helium/O2 analyzer (ATA PRO, Analox, UK). All divers also carried 3-4 off-board ('bail-out') dive tanks to allow an independent return to the surface in the event of CCR failure. These 'stage cylinders' (or 'stages') contained either trimix, nitrox or pure oxygen depending on the decompression planning. One diver carried a second rebreather as bail-out.

#### Measurements

Evaluation of decompression stress and of the potential benefit of preventive measures has been done historically based on the presence or absence of clinical symptoms of DCS. However, for obvious ethical reasons, this is not acceptable in the field of recreational diving (Ozyigit et al. 2019). Although imperfect, it is now accepted that research projects can use VGE data as a surrogate endpoint (Doolette 2016;

Balestra et al. 2019). Different methods of detection of VGE are possible, such as Doppler ultrasonic bubble detectors or 2D cardiac echography (Mollerlokken et al. 2016). During field studies, bubbles are usually detected in the right atrium, ventricle, and pulmonary artery. Then, the amount of detected VGE is graded according to different systems, either, categorical (Eftedal and Brubakk 1997), semi-quantitatively (Germonpre et al. 2014) or continuous (Imbert et al. 2019; Papadopoulou et al. 2018; Balestra et al. 2016).

In the present setting, two measurements were taken at 30- and 60-min post-dive. This number of measurements were limited by practical reasons. VGE signals were measured using both a portable echocardiography device (Sonosite M-Turbo, FUJIFILM Sonosite Inc, Amsterdam, The Netherlands) with a sectorial array ultrasound probe on a four-chamber view by an experienced researcher (CB), and by divers' self-assessment using the O'Dive sensor (Doppler ultrasound signals) positioned under the clavicle and then transmitted to a smartphone equipped with the O'Dive technical application (Azoth Systems, Ollioules, France). This version of the application includes the possibility to enter various gas mixes (bottom gas, deco gas) into the data, select open or closed-circuit diving, provide gradient factors, as well as a link to upload the actual dive profile directly from the dive computer. Once synchronized, Doppler data are analyzed directly by the Azoth Systems server, according to an undisclosed protocol (Hugon et al. 2018).

The echocardiographic VGE signals over the 1 min recording were evaluated by frame-based bubble counting as described by Germonpré et al. (2014), but also scored according to the Eftedal–Brubakk categorical score (Eftedal and Brubakk 1997), while Doppler signals recording retrieved from the server were blindly reviewed to obtain a posteriori bubble grade according to the expanded Spencer scale. For the purpose of comparison and correlation testing both categorical score were converted within the simplified bubble grading system (BGS) (Marroni et al. 2004).

Since several reports suggest that post-dive dehydration could promote the development of DCS or at least modified VGE production (Blatteau et al. 2008; Fahlman and Dromsky 2006; Sengun et al. 2012; Skogland et al. 2008), subjects were weighed on an electronic device (Mini Crane Scale OCS-300, Vetek Weighing AB, Väddö, Sweden; precision ± 50 g) before and 15 min after surfacing. Simultaneously, intracellular, extracellular, and total body water were also estimated by means of multifrequency tetrapolar impedancemetry (Biody Expert system, Cagnes sur Mer, France).

#### Statistical analysis

After evaluation of the normality distribution with the Kolmogorov-Smirnov test, differences between preand post-dive measurements were assessed using either



Friedman test with Dunn's post-hoc or one-way ANOVA with Bonferroni post-hoc as appropriate for multiple comparisons and by paired t-test or Mann–Whitney for simple comparison. Correlation between echographic and O'Dive bubble grades was evaluated through a Pearson r calculation. A P value of < 0.05 was considered significant for all statistical tests. Statistical analysis was performed using GraphPad Prism version 9.1.0 for MacOS (GraphPad Software, San Diego, CA, USA).

#### Results

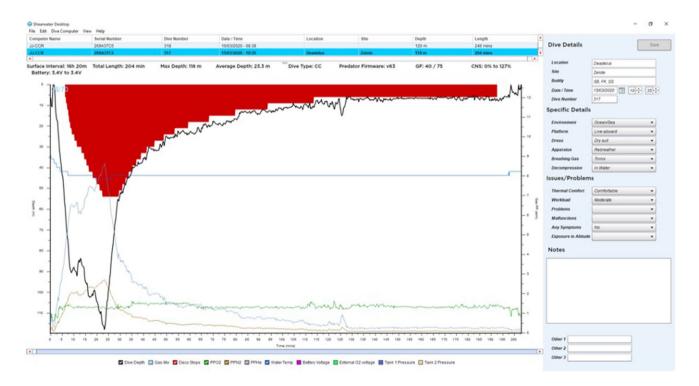
All divers were males, were declared medically fit to dive prior to the first dive and in possession of a valid diving certification (at least GUE CCR diver level 2) and insurance. They were all familiar with diving at depth of 100 msw or beyond. Characteristics of the divers are shown in Table 1.

A typical recorded profile is shown in Fig. 1. Depth-time profiles of the 30 man-dives are given in Fig. 2. The average depth was  $97.3 \pm 26.4$  msw [100 msw; 70–120] with a runtime of  $160 \pm 65$  min [130 min; 105-207]. Run time was

**Table 1** Subjects' characteristics (n = 10) before the first dive

	Diver 1	Diver 2	Diver 3	Diver 4	Diver 5	Diver 6	Diver 7	Diver 8	Diver 9	Diver 10	Mean [range]
Age	58	27	44	54	33	50	43	54	46	35	45 [27–58]
Weight (kg)	66	76.5	81	72.9	76	83.2	79	75.3	91	68	71.7 [66–91]
Height (cm)	180	183	173	181	193	181	172	178	175	178	174.5 [172–193]
BMI (kg/m <sup>2</sup> )	20.4	22.8	27.1	22.2	20.4	25.4	26.7	23.8	29.7	21.5	23 [20.4–29.7]
BF (%)	15.0	18.9	25.5	16.2	18.3	24.9	24.0	17.8	23.3	14.6	18.6 [14.6–25.5]
Experience (years)	13	13	29	40	10	36	21	19	26	16	20 [10-36]
Experience (dives)	800	1000	10,000	1500	600	5480	1800	700	700	7000	1250 [600–10,000]
Max depth (msw)	100	116	124	120	131	136	125	100	100	116	118 [100-136]

BMI body mass index, BF body fat



**Fig. 1** Typical record of diving profile at a maximum depth of 120 msw and 11 min bottom time.  $PPO_2$  partial pressure of oxygen (Green line), PPHe partial pressure of helium (Brown line),  $PPN_2$ 

partial pressure of nitrogen (Grey line). The Black line correspond to the depth profile while the red zone illustrates the decompression ceiling



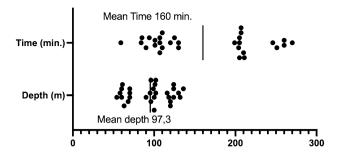


Fig. 2 Mean and standard deviation of depth and dive time (n=30)

primarily affected by the maximal depth and bottom time but also by the level of conservatism which was altered by selecting gradient factors to modify the allowed maximum super-saturation limits) at each diver's discretion as illustrated in Table 2.

There were no incidents of divers breaching their decompression profile as calculated by their dive computers. One diver developed unusual fatigue, suggestive of potential DCS, after the 6th dive; the diver chose not to be treated and continued diving for the remainder of the expedition. His symptoms resolved spontaneously. All divers flew home more than 48 h after the last dive, without developing any symptoms of DCS.

Because of technical and organizational mishaps, only seven full sets out of ten datasets were available for analysis.

Results of the echocardiographic frame-based bubble counting are shown in Fig. 3. The mean bubble count per cardiac cycle was  $14 \pm 13$  at 30 min and  $13 \pm 13$  at 60 min. Because of data variability, no statistical significance was identified neither between dives, nor between times of measurement (P = 0.07, Friedman test).

When converted into BGS, echographic bubble grades were highly correlated with the BGS given by Azoth

Table 2 Example of decompression procedures, breathed gases, and bubble grade recorded after the Zealot wreck-dive

	Diver 1	Diver 3	Diver 4	Diver 5	Diver 6	Diver 7
GF L/H	50/80	50/90	45/75	55/90	50/99	50/90
Bailout stages (O <sub>2</sub> /He) (%)	12/60 18/54 50	10/70 28/34 50	12/60 28/34 52	10/70 30/30 50	10/70 18/45 50/15	10/70 Second CCR Bailout
	100	100	100	100	100	
Depth max/avg (msw)	116/24	124/27	120/23	120/27	132/30	118/26
Deco stops (depth/duration) (msw/min)	51/1 48/1 45/1 42/2 39/2 36/2 33/3 30/4 27/4 24/5 21/6 18/9 15/9 12/14 9/16 6/22 3/42	54/1 51/1 48/2 45/2 42/3 39/2 36/2 33/3 30/3 27/4 24/4 21/6 18/7 15/9 12/11 9/17 6/27	54/1 51/1 48/1 45/2 42/2 39/2 36/3 33/3 30/3 27/5 24/5 21/8 18/9 15/13 12/17 9/23 6/95	48/1 45/1 42/3 39/3 36/3 33/3 30/3 27/4 24/4 21/6 18/8 15/12 12/23 9/20 6/31 3/46	48/2 45/1 42/1 39/3 36/3 33/3 30/3 27/4 24/4 21/6 18/8 15/12 12/23 9/30 6/70	51/1 48/1 45/1 42/2 39/2 36/2 33/3 30/3 27/4 24/5 21/6 18/8 15/9 12/12 9/18 6/26 3/42
Runtime (min)	204	3/47 221	246	207	270	211
Eftedal–Brubakk/BSG	204 3/HBG	4/HBG	240 2/LBG	207 3/HBG	270 4/HBG+	1/LBG
O'DIVE/BSG	3/HBG	4/HBG	2/LBG	2/LBG	4/HBG	0/LBG
Diluent (O <sub>2</sub> /He) (%)	10/70	10/70	9/68	10/70	10/70	11/60
PPO <sub>2</sub> set point (ATA)	1.3/1.5/1.3	1.2/1.5	1.3/1.5/1.3	1.3/1.4	1.3/1.5	1.3/1.4
CNS exposure to oxygen (min)	142	120	127	131	154	129

Bailout stages are carried for use as an emergency supply of breathing gas in the event of CCR loop failure

BSG simplified bubble grading system, PPO<sub>2</sub> partial pressure of oxygen, CNS central nervous system oxygen exposure according to dive computer calculations, GF L/H gradient factor low/high



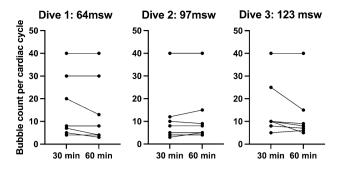


Fig. 3 Individual bubble count changes after 100 m depth dives

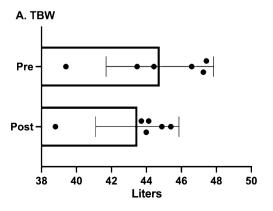
System's analysis based on the Doppler recording made (Spearman r of 0.81, P = 0.008).

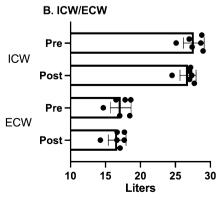
Total body water was reduced by  $1.0\pm1.21$  between preand post-dive measurements. However, this was not statistically significant (P=0.4, Mann–Whitney). No variation is seen in intracellular (pre:  $61.8\pm1.1$  vs. post:  $61.9\pm0.9\%$ , P=0.4, paired t-test) and extracellular water (pre:  $38.4\pm0.7$  vs. post:  $38.3\pm0.9\%$ , P=0.7, paired t-test) either (Fig. 4). No specific correlation was identified between BMI or hydration status and bubble grade.

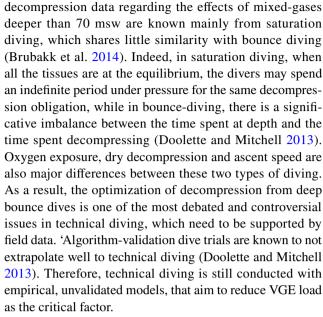
#### **Discussion**

Since the reconfiguration of Biomarine CCR155, one of the first mixed gas rebreathers to be adopted by sport divers in the early 1990s, the use of CCRs by recreational or technical divers has become increasingly common. This, however, comes with a cost, as the fatality rates for rebreather diving are 8–10 times higher than for open circuit diving (Fock 2013). Although the primary trigger for most of these accidents can be identified as a failure of the 'human–machine interface', two-thirds of fatal dives were associated with a high-risk dive or high-risk behavior such as solo diving, exceeding depth limits, or carrying insufficient gases. The latter two are directly related to decompression management, which is a major factor in diver's safety. Unfortunately,

Fig. 4 Total body water (TBW), extracellular (ECW) and intracellular water (ICW) difference after one 100 msw bounce-dive to 123 msw







The results of this observational study suggest they may not be capable of doing so. Indeed, although the diving profile were allegedly conservative according to the decompression algorithm used, a high incidence of high-grade VGE was detected. Moreover, it seems that the deepest divers, while using similar decompression scheme, exhibit different bubbling patterns, a feature already demonstrated in an open circuit single exposure (Papadopoulou et al. 2018). More, regardless of the level of conservatism used, the amount of detectable VGE is rather constant from dive to dive. Indeed, it can be observed that VGE counts do not significantly increase with succeeding dives, despite the facts that these dives may be considered 'provocative' with depths repeatedly > 39 msw daily. This highlights the importance of individual factors to manage decompression safety. It must also be noted that the lone diver who developed unusual fatigue suggestive of potential DCS, owned the highest bubble grade, highest BMI and lowest fitness level among the divers who volunteered for this observational study. Although anecdotal, this might also be seen as an argument for a more diver-centric approach.



During this observational study, two individual parameters were measured, dehydration and physical status. Although animal studies have produced conflicting results, divers' hydration state has long been considered as a critical point for decompression safety. This is partially supported among humans by a field study demonstrating that pre-dive hydration reduces VGE (Gempp et al. 2009). In this study, using a shallower profile (30 min/30 msw with a 9 min safety stop at 3 msw), in the absence of prehydration, the plasma volume was significantly reduced by 2.2% as well as the total body water with a mean 1.2 l, ranging from 800 to 1500 ml. In our setting, the dives were deeper and longer (160 min/100 msw) and seem to dehydrate the diver in a similar fashion (mean of 1.0 l, ranging from 500 to 2400 ml); however, there was no significant difference between pre- and post-dive measurements. Also, fluids shift between the extracellular and intracellular did not seem to occur. This might be explained by CCR technology, which uses a closed loop. It is indeed a known fact in anesthesiology that a closed-circuit conserve respiratory heat and humidity. It is also known that relationships between VGE grades and age, maximal oxygen uptake, and percent body fat might also exist. Younger, slimmer, or aerobically fitter divers produced fewer bubbles compared with older, fatter, or poorly physically fit divers (Carturan et al. 2002). Coincidentally, although declared fit to dive, the heaviest bubbler was closer to the second diver profile with some minor comorbidities than to the first description. Nonetheless, we couldn't demonstrate such correlation among our results. More research is needed to extrapolate on this specific hypothesis.

It must be acknowledged that the number of subjects is low, so the reproducibility and power of the study cannot be ascertained. However, given the technical difficulty of retrieving exploitable data (the ideal tool being cardiac echography recordings) in the field, an easier way to gather reliable data needs to be considered. Recently, the detection of VGE in subclavian veins has been proposed and validated (Hugon et al. 2018), and a simple, do-it-yourself tool has been developed and is currently marketed. The feasibility of the O'Dive system for collection of diver's VGE data has been recently reported (Germonpre et al. 2020). In this study, divers performed the O'Dive measurements on themselves without any problems. There appears to be an excellent correlation between the BGS reported by the O'Dive (Azoth Systems server data) and the Eftedal-Brubakk scores obtained from the echocardiography images. This would indicate that in the future, O'Dive measurements could well become a major asset in large-scale diving decompression research. For the present time however, we recommend that O'Dive data still be 'backed' by a validated VGE measurement.

#### Conclusion

In conclusion, the present observation represents an original dataset regarding VGE count related to CCR deep trimix dives performed in open sea, by technical divers. Although preliminary, it questions the precedence of decompression algorithm over individual risk factors and pleads for an individual approach of decompression. It also highlights the gap of knowledge to develop comprehensive diving guidelines regarding deep CCRs diving.

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Author contributions CB, FG, and PL contributed to the study conception and design. Material preparation, data collection and analysis were performed by CB, FG, PG, PL. The first draft of the manuscript was written by CB and PL. PG and FG revised the manuscript critically for important intellectual content and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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**Data availability** The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

#### **Declarations**

Conflict of interest The authors declare that they have no conflict of interest.

**Ethical approval** Study approved by the he Bio-Ethical Committee for Research and Higher Education, Brussels (No B200-2020-088).

Consent for publication Not applicable.

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