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ORIGINAL ARTICLE

Physiological effects of mixed-gas deep sea dives using a closed-circuit rebreather: a field pilot study

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Abstract

Purpose Deep diving using mixed gas with closed-circuit rebreathers (CCRs) is increasingly common. However, data regarding the effects of these dives are still scarce. This preliminary field study aimed at evaluating the acute effects of deep (90–120 msw) mixed-gas CCR bounce dives on lung function in relation with other physiological parameters.

Methods Seven divers performed a total of sixteen open-sea CCR dives breathing gas mixture of helium, nitrogen and oxygen (trimix) within four days at 2 depths (90 and 120 msw). Spirometric parameters, SpO_2, body mass, hematocrit, short term heart rate variability (HRV) and critical flicker fusion frequency (CFFF) were measured at rest 60 min before the dive and 120 min after surfacing.

Results The median [1st–3rd quartile] of the forced vital capacity was lower (84% [76–93] vs 91% [74–107] of predicted values; $p = 0.029$), whereas FEV1/FVC was higher (98% [95–99] vs 95% [89–99]; $p = 0.019$) after than before the dives. The other spirometry values and SpO_2 were unchanged. Body mass decreased from 73.5 kg (72.0–89.6) before the dives to 70.0 kg (69.2–85.8) after surfacing ($p = 0.001$), with no change of hematocrit or CFFT. HRV was increased as indicated by the higher SDNN, RMSSD and pNN50 after than before dives.

Conclusion The present observation represents the first original data regarding the effects of deep repeated CCR dives. The body mass loss and decrease of FVC after bounce dives at depth of about 100 msw may possibly impose an important physiological stress for the divers.

Keywords Scuba · Spirometry · Closed circuit rebreather · Heart rate variability

Abbreviations

ANOVA	Analysis of variance
ApEn	Approximate entropy
BF	Body fat
BMI	Body mass index
BML	Body mass loss
CCR	Closed circuit rebreather
CFFF	Critical flicker fusion frequency
DCS	Decompression sickness
DFA	Detrended fluctuations
ECG	Electrocardiogram
EVLW	Extravascular lung water
FEF25–75	Forced expiratory flow at 25 and 75%
FEV1	Forced expiratory volume in 1 s
FVC	Forced vital capacity
He	Helium
HF	High frequency
HR	Heart rate
HRV	Heart rate variability
Ht	Hematocrit
GLI	Global lung initiative
LF	Low frequency
MET	Metabolic equivalent of the task

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msw	Meter of sea water
O2	Oxygen
pNN50	Proportion of pairs of successive NN (R-R) intervals that differ by more than 50 ms
PEF	Peak expiratory flow
Po2	Oxygen partial pressure
RMSSD	Root mean square of the successive differences
scuba	Self-contained underwater breathing apparatus
SD	Standard deviation
SDNN	Standard deviation of the NN (R-R) intervals
ShanEn	Shannon entropy
SpO2	Oxygen saturation
ULC	Ultrasonic lung comets
UTP3	Under the pole 3 expedition

Introduction

Since the development of closed-circuit rebreathers (CCRs) for the non-military market, its use by professional and recreative divers has become increasingly common. As a result, physicians are increasingly requested to perform dive medical screenings, as well as manage CCR diving related emergencies. It is therefore essential that comprehensive diving guidelines, including specific fitness to dive prerequisites and CCR diving contraindications will be made easily assessable for reference. Better knowledge of CCR diving is essential given the increased risk of fatality actually estimated to be 10 times higher than recreational open-circuit scuba diving (Fock 2013). However, data regarding the effects of bounce mixed-gas deep CCR diving are still needed.

CCRs deliver a gas mixture using different diluents (air, helium or helium-nitrogen) with a constant partial pressure of oxygen (generally 120–160 kPa), which allow longer and deeper dives than classical open-circuit scuba using less gas. Indeed, typical CCR dive correspond to a maximum depth about 90 m of sea water (msw) for 30–60 min at bottom (Mitchell and Doolette 2013), although deeper dives have also been reported. This is much deeper and longer than average recreational diving.

The physiological effects of mixed-gas dives deeper than 80 msw are known mainly from studies performed during saturation diving (Brubakk et al. 2014). However, there are important differences between these two types of diving. For instance, the partial pressure of oxygen ranges from 21 to 80 kPa during saturation dives, against 120–160 kPa in the case of CCR dive. Also, the decompression is slower (ranging from 20 to 36 msw/24 h) and performed in a dry chamber, while CCR dives decompressions are usually in water and at a speed ranging about 40 msw/h. As a result, there is a lack of data regarding deep CCR in water bounce diving at 100 msw and more.

In this regard, besides the well documented potentially deleterious action of hyperoxia on pulmonary function (Arieli 2019), immersion itself also carries a risk of pulmonary edema (Castagna et al. 2018). Indeed, during immersion, hydrostatic pressure restrains the vascular compliance which leads to a redistribution of blood volume from peripheral to central circulation. This filling of the central vascular bed leads to a relative increase in right sided heart output compared with the left, due to increased cardiac preload, which correlates with extravascular lung water (EVLW) accumulation. Additionally, the lungs are the first organ concerned by the decompression-induced vascular gas emboli that are normally filtered out by the pulmonary microcirculation and do not pass into the arterial side of the circulation (Papadopoulou et al. 2014). This situation exposes them to increased vascular damage and permeability which, in turn, can lead to inflammation and edema as already reported in animal models of decompression sickness (Bao et al. 2015). All this is consistent with the feeling of respiratory limitation that divers sometimes report after deep CCR dives.

Therefore, the purpose of this preliminary field study was to assess whether pulmonary function is impaired after deep mixed-gas CCR bounce diving.

Methods

The study was conducted during working dives performed by the Under The Pole (UTP) 3 team in the frame of the Deep Hope scientific program for the study of deep corals, known as "mesophotics". Dives were performed during September 2019 in Tahiti (water temperature 27 °C).

Study population

Characteristics of the divers are shown in Table 1. At the time of the study, all divers were healthy and had a valid medical certificate for diving. They all volunteered for this study. All the subjects were experienced CCRs mixed-gases deep divers, familiar with diving at depth greater than

Table 1 Subjects characteristics ($N=7$) before the first dive

	Median [1st–3rd quartile]
Age (years)	35 [29–39]
Height (cm)	184 [175–187]
Weight (kg)	73.0 [72.0–90.5]
BMI ($kg.m^{-2}$)	21.8 [20.6–25.9]
BF (%)	11.7 [7.1–17.2]

BMI body mass index, *BF* percentage of body fat

100 msw. They were not under medication. All participants were informed about the potential risks and discomforts associated with the study and gave their informed consent prior the study. All procedures were performed in accordance with the 1964 Helsinki Declaration.

Study design

We took advantage of a series of repeated dives planned by UTP divers to harvest samples at two depths on a coral site. Seven divers performed a total of sixteen open-sea dives within four days: 4 dove three times, 1 dove twice and 2 dove once. The diving sites were located in the relative vicinity of the field laboratory, and divers were transported within 10 min to the sites by a power boat. The dive profiles were planned as part of the UTP3—Deep Hope scientific program which aimed at harvesting biological samples. Two dive profiles were used, and a representative dive profile is shown in Fig. 1. One was at the maximum depth of 120 msw with a descent rate of 24 $m.min^{-1}$, a bottom time of 10 min and a total duration of 180 min in average; the other profile consisted on a dive to 90 msw deep with a descent rate of 16 $m.min^{-1}$, a bottom time of 10 min and a total duration of 160 min in average. Divers breathed trimix with either 6% O_2-70% He for 120 msw dives, or 8% O_2-62% He for 90 msw dives. The oxygen partial pressure was set to 120–130 kPa during the descent and at bottom, and to 150–160 kPa during the decompression. The decompression was conducted according to the Shearwater integrated multi-gas decompression dive computer based on the Petrel version 4.4 (model Buhlmann GF ZHL-16C) algorithm. Gradient Factors were set to 40–50 (low) and 60–99 (high). Subjects wore 5 mm neoprene wet suit with hood, used a JJ-CCR Rebreather DiveCAN®—CE Edition (JJ-CCR ApS, Presto, Denmark). Depth and dive time were monitored by each diver's personal dive computer.

Measurements

Spirometric parameters (including FVC, FEV1, FEV1%, PEF, and FEF25-75) and SpO_2 were recorded outside the water using a portable spirometer (Spirobank II Smart; MIR Medical International Research Srl, Rome, Italy) following GLI (Global Lung Initiative) 2017 for Caucasian adults. This model was previously validated (Degryse et al. 2012) and its technical characteristics meet the ISO26782:2009 international standards (Graham et al. 2019). All measurements are resting measurements, they were obtained by a trained operator with subjects being seated erect and not wearing diving suit. The reported values are the best of three eligible efforts.

It was reported that diving leads to the redistribution of blood centrally which, in turn, could promote the accumulation of extravascular lung water (Castagna et al. 2017). This

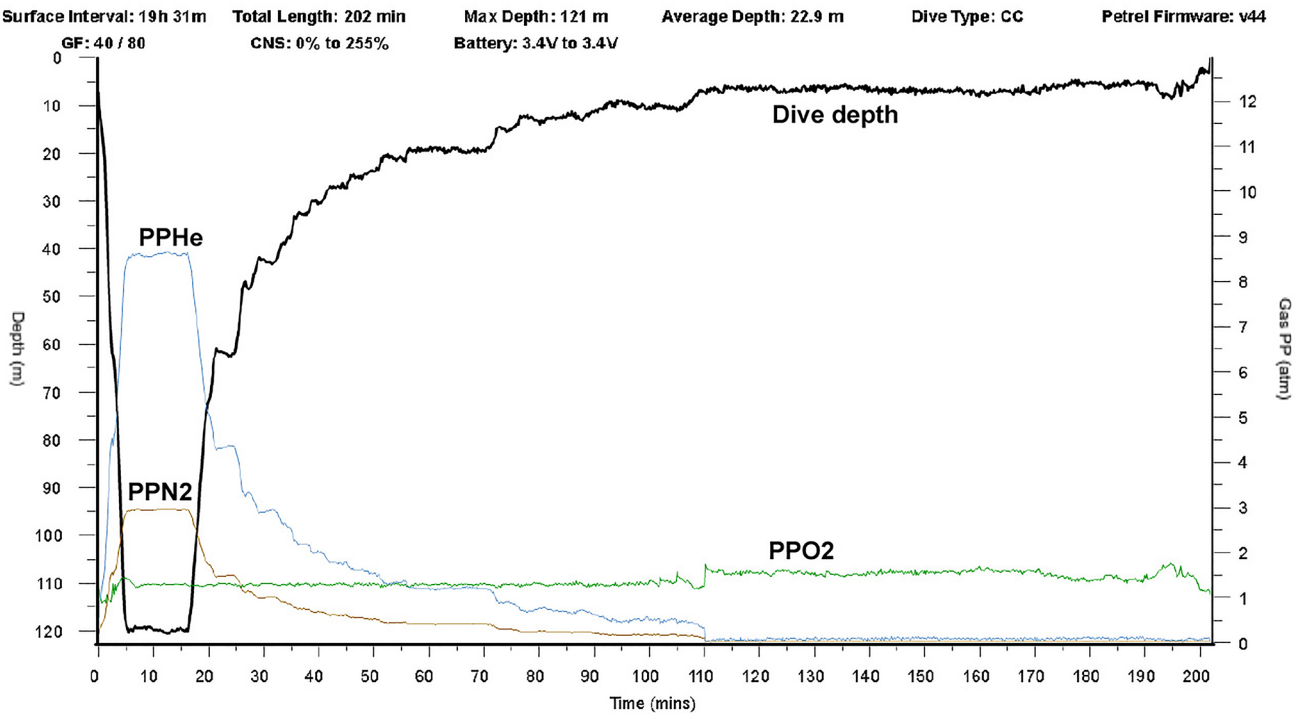

Fig. 1 typical record of diving profile at a maximum depth of 120 msw and 11 min bottom time. *PPO2* partial pressure of oxygen, *PPHe* Partial pressure of helium, *PPN2* partial pressure of nitrogen

increase in thoracic blood pressure stimulate diuresis (Castagna et al. 2013, 2015) and modifies the cardiac autonomic nervous system (Lundell et al. 2021). Therefore, subjects were weighed on an electronic device (Tanita®, Hoofddorp, The Netherlands; precision ± 20 g), and the hematocrit (Ht) was determined from blood sample taken at the fingertip.

ECG was recorded during 15 min with a Zephyr BioHarness 3 system (ZephyrTechnology 2013; Annapolis, MD, USA), subject being in supine position, and transferred to a personal computer for HRV analysis using Kubios HRV Premium Analysis Software 3.3.1. (Biosignal Analysis and Medical Image Group, University of Eastern Finland, Kuopio, Finland).

Finally, since previous studies suggest a competitive effect between oxygen and nitrogen during diving on cerebral arousal (Rocco et al. 2019), alertness was assessed using a critical flicker fusion frequency (CFFF) test with a specially designed device (Lafère et al. 2019).

All measurements were taken at rest 60 min before the dive and 120 min after surfacing, except for Ht which was measured within 30 min post-dive for safety reasons to enable subjects to drink. Post-dive measurements were taken 120 min instead of 60 min after surfacing to allow time for divers to return to a resting state after undressing and storing their rebreathers.

Statistical analysis

Statistical analysis was performed with the Statistica 13 software program (Tulsa, Oklahoma, USA). All data are presented as median (1st and 3rd quartile). A Wilcoxon test for paired data was used to compare values obtained for each subject before and after the dive. When more than two groups were analyzed, we ran a non-parametric ANOVA followed by Wilcoxon test for paired data in case of significant differences. Statistical significance was set a priori at $p < 0.05$.

Results

No signs of DCS or neural oxygen toxicity were detected.

Data from spirometry are presented in Table 2. Wilcoxon test for paired values showed statistically significantly lower FVC and higher FEV1/FVC after the dives compared to basal values. No differences were detected between pre- and post-dive values for the other pulmonary parameters.

To further analyze the modifications of FVC in relation with the repetition of dives we performed a non-parametric ANOVA test for repeated measures which showed a statistically significant difference between measurements both when expressed as absolute values ($N = 4$, $df = 5$, $F = 15.857$, $p = 0.007$) and as percentage of theoretical values ($N = 4$, $df = 5$, $F = 15{,}857$, $p = 0{,}007$). As shown in Fig. 2, FVC decreased throughout the first and second dives but the difference reached statistically significant threshold after dive 2, so that post-D2 values were statistically significantly lower than those from previous measurements (i.e. pre-dive 1, post-dive 1, and pre-dive 2). The median FVC values decreased from 5.59 L [5.33–6.09] and 109% [97–110] of individual predicted values before the first dive to 4.23 L [3.83–4.89] ($p = 0.043$) and 73% [72–83] of predicted values ($p = 0.043$) after dive 2.

Table 2 Spirometry parameters

	Pre	Post	*p* value
FVC			
L	5.13 [4.00–6.09]	4.41 [4.08–5.61]	**0.024**
% Pred	91 [74–107]	84 [76–93]	**0.029**
FEV_1			
$L.s^{-1}$	4.74 [4.00–5.40]	4.26 [4.02–5.22]	0.109
% Pred	107 [93–119]	103 [95–108]	0.133
FEV1/FVC			
%	95 [89–99]	98 [95–99]	**0.019**
% Pred	117 [111–121]	120 [119–123]	**0.019**
PEF			
$L.s^{-1}$	10.77 [8.65–11.32]	10.19 [8.83–11.50]	0.501
% Pred	109 [89–116]	101 [90–116]	0.501
FEF 25–75			
$L.s^{-1}$	5.78 [5.19–6.37]	5.73 [4.93–6.66]	0.717
% Pred	132 [121–148]	135 [120–149]	0.756

Values are expressed as median [1st–3rd quartile] before (pre) and after (post) dives. The percentage of expected values according to the GLI (% pred), and the *p* values of the pre vs post differences are also reported

Statistically significant differences ($p < 0.05$) are indicated in bold

SpO_2 values were not statistically significantly different after the dives compared to basal values (96.5% [95.7–97.9] vs 97% [95.9–97.9], pre and post dive, respectively, $p = 0.717$).

Median body weight before the dives was 73.5 kg [72.0–89.6] and was statistically significantly decreased to 70.0 kg [69.2–85.8] after surfacing ($p = 0.001$). Nevertheless, dives did not change Ht (51.5% [46.0–56.5] vs 45.0% [44.0–56.0], pre and post dive, respectively, $p = 0.489$).

CFFF did not change throughout the dives (42.12 Hz [39.50–44.12] vs 42.37 Hz [39.87–44.50], pre and post dive, respectively, $p = 0.576$).

HRV parameters are presented in Table 3. The standard deviation of the R-R intervals (SDNN), the root mean square of the successive differences (RMSSD) and the proportion of pairs of successive R-R intervals that differ by more than 50 ms (pNN50), as well as the non-linear parameter SD1 (standard deviation 1 of the instantaneous beat-to-beat

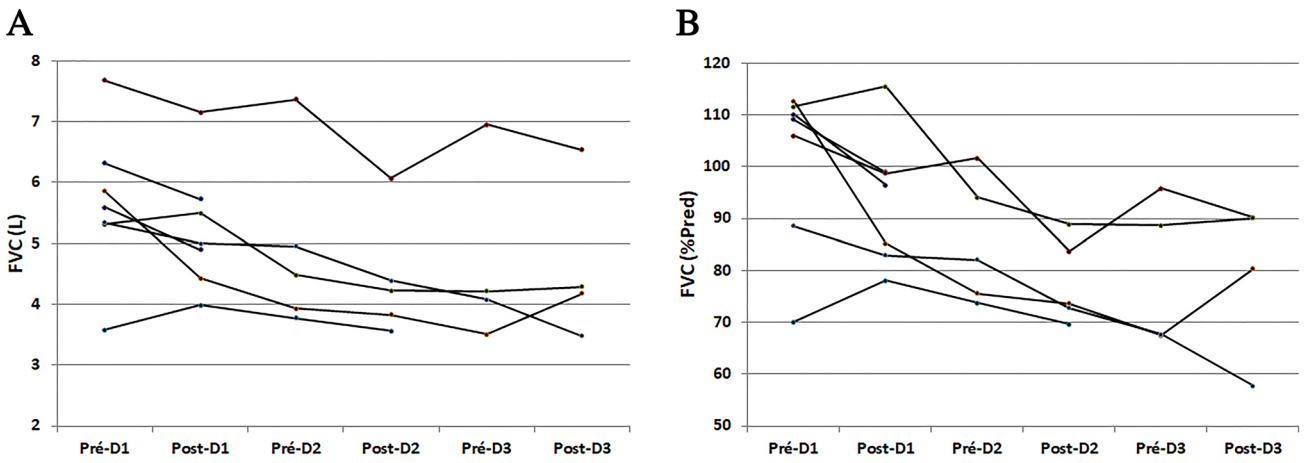

Fig. 2 Evolution of FVC throughout repetition of dives when FVC is expressed in L (**A**) and as percentage of individual expected values according to the GLI (**B**)

Table 3 Short term heart rate variability indicators obtained before (pre) and after (post) dives

	Pre	Post	*p* value
Time domain			
HR (BPM)	145 [138–155]	148 [146–161]	0.074
Mean RR (ms)	417 [389–436]	409 [374–415]	0.074
SDNN (ms)	27.4[23.9–34.0]	34.7 [28.3–44.3]	**0.012**
RMSSD (ms)	15.2 [13.2–47.7]	18.8 [17.6–28.9]	**0.012**
pNN50 (%)	0.6 [0.2–8.2]	1.7 [0.7–6.9]	**0.014**
RR triangular index	6.98 [5.30–7.86]	8.01 [5.86–9.41]	0.332
Frequency domain			
LF (ms^2)	189 [159–248]	223 [102–276]	0.386
HF (ms^2)	211 [132–609]	320 [191–590]	0.332
LF/HF	1.205 [0.506–1.362]	0.579 [0.312–0.890]	0.168
Total power (ms^2)	622 [497–902]	822 [629–1410]	0.202
Non linear			
Poincarre plot SD1	10.7 [9.3–33.8]	13.3 [12.5–20.5]	**0.012**
Poincarre plot SD2	37.0 [32.5–42.9]	42.5 [38.1–57.2]	0.102
DFA: α1	1.388 [0.741–1.460]	1.304 [0.642–1.381]	0.059
DFA: α2	0.805 [0.688–0.901]	0.852 [0.666–0.983]	0.332
ApEn	1.090 [0.938–1.273]	1.095 [0.846–1.243]	0.444
ShanEn	3.378 [3.200–3.715]	3.445 [3.036–3.827]	0.798

Statistically significant differences ($p < 0.05$) are indicated in bold

variability) were statistically significantly higher after than before dives, indicating an increased variability.

Discussion

The purpose of this preliminary study was to assess the effects of mixed-gases CCR sea diving at depth up to 120 msw on pulmonary function in relation with other physiological parameters. The main outcome is a post-dive decrease of FVC, together with increased FEV1/FVC ratio. It was associated with up to 3.5 kg body mass loss and increased heart rate variability.

We are not aware of previous studies which have assessed the effects of scuba on pulmonary function after open-sea dives deeper than 80 msw and, therefore, the present data represent the first ones obtained after dives at 90–120 msw depth. In these previous studies, FVC was either decreased (Cirillo et al. 2003; Wilson 2011) or unchanged (Tetzlaff et al. 2001; Ljubkovic et al. 2010) after bounce diving up to 65 msw with open circuits whereas FEV1 was unchanged. When CCRs were used for diving, neither FVC nor FEV1 were altered after dives at depths ranging from 15 msw to an average of 69 msw (Fock et al. 2013; Bosco et al. 2018; Castagna et al. 2019). The present data indicate that for deeper dives FVC is decreased after diving. Along with this, we found that not only FEV1 was not different but FEV1/FVC was even statistically significantly higher after than before diving, indicative of unchanged pulmonary resistance. Strikingly, when analyzing the influence of the repetition of dives, we found that FVC gradually decreased so that after the second dive the median was reduced from 109 to 73% of the predicted value only. Although lung volume measurements are needed for confirmation (Krol et al. 2019) the association of FVC < 80% predicted and the absence of airflow obstruction (denoted by an FEV1/FVC > 70%) is classically interpreted as a spirometry restrictive pattern (Godfrey and Jankowich 2016).

When trying to understand the reasons of these changes, the first hypothesis which comes to mind is hyperoxia. Indeed, the partial pressure of oxygen was set at 130–140 kPa during both the descent and the stay at bottom and was further increased to 150–160 kPa during decompression. This marked continuous hyperoxia lasted for 120–180 min which might have potentially carry a risk of pulmonary toxicity as suggested from previous studies (Shykoff and Florian 2018; Arieli 2019). However, according to Arieli's approach (Arieli 2019), breathing 150 kPa Po_2 during 120 or 180 min would decrease FVC by only 0.21 or 0.47%, respectively. Moreover, assessment of pulmonary function after shallow CCR dives showed that neither pulmonary forced vital capacity nor pulmonary flows are changed after breathing 140 kPa of oxygen partial pressure during a 20 min CCR dive at 15 msw depth (Bosco et al. 2018), or 150 kPa during a 12 h CCR dive at 20 msw (Castagna et al. 2019). They also remained unchanged after repeated dives (20 dives within 11 days) at an average depth and duration of 69 msw during 112 min with a 130–140 kPa Po_2, and even 150–160 kPa during decompression (Fock et al. 2013). It is therefore unlikely that the decreased FVC that we found results from hyperoxia.

On the other hand, the lungs are the primary organ reached by the intravascular gas emboli formed during decompression (Papadopoulou et al. 2014). Animal models clearly show a relationship between decompression stress and pulmonary inflammation and edema (Bao et al. 2015). In humans, accumulation of extravascular lung water (EVLW) has been evidenced after scuba dives by an increase of the number of ultrasonic lung comets (ULC) detected. It is noteworthy that when assessing the sum of ULC after diving, the same group reported a higher post-dive increase of the number of ULC after trimix open-circuit dives up to 80 msw depth (Marinovic et al. 2010) than after dives up to 65 msw (Ljubkovic et al. 2010), and no change after a 20 min air open-circuit dive to 33 msw (Dujic et al. 2011). This suggests that EVLW accumulation could increase in relation with either the depth of the dive or the stress of decompression. In this regards, data from another deep dive expedition that have been shared (the Gombessa 5 expedition) are of great interest. Indeed, during this expedition, four divers performed 20 trimix CCRs dives at depths ranging from 70 to 130 msw within a total time laps of 23 days. Interestingly, during Gombessa 5, the divers were maintained in hyperbaric saturation conditions during the whole expedition, so that no decompression was needed at the end of the CCR dives. Nevertheless, spirometry measurements performed before and after each dives on two divers showed a statistically significant decrease of FVC from 5.39 [4.86–6.96] L before diving to 4.98 [4.61–6.75] L after the dives ($p = 0.005$). This was associated with a slight and not statistically significant increase of FEV1/FVC (75 (60–81] % vs 77 [60–83] % before and after diving, respectively; $p = 0.061$). It appears therefore that the post-dive decrease of FVC could be present even in the absence of decompression. Furthermore, the post-dive decrease of FVC during UTP3 was not statistically significantly different compared to Gombessa 5 data (93 [87–99] vs 95[92–101] percent of pre-dive values for UTP3 and Gombessa 5 dives, respectively; $p = 0.147$). Taken together, these data strongly suggest that the post-dive decrease of FVC is not a consequence of decompression stress, but, more likely results from the effects of immersion at depth itself.

In this regard, together with the compression effect exerted by the neoprene wet suit, immersion is considered as the main mechanism responsible for the redistribution of blood centrally into the heart and lungs (Castagna et al. 2013). The increase in thoracic blood pressure stimulates diuresis to correct central blood volume. The body mass loss (BML) subsequent to this water elimination was reported to average 0.90 to 0.97% after static immersion (Castagna et al. 2013, 2015) but can reach 1.34% after underwater fin swimming during 2 h at 3 msw depth (Castagna et al. 2015). This is lower than the median 3% BML we found after the dives although water temperature was lower (27 °C in our study vs 29 °C) and the durations of immersion were similar. Moreover, the loss in body weight is therefore probably underestimated in our study since, for safety reasons, divers were allowed to freely rehydrate after surfacing (which probably explains the lack of change of the hematocrit after dives since divers drank even before blood sampling). Alternatively, most of the studies regarding fluid balance during scuba diving were conducted at shallow depth. It is of note that total peripheral resistances are increased (Gaustad et al. 2020) and urine flow is augmented during deep dry hyperbaric exposure, i.e. in the absence of immersion (Hong and Claybaugh 1989; Brubakk et al. 2014), which could have added to further increase the water loss in our divers. Therefore, the restrictive spirometry pattern present in this study but not in previous measurements after shallow dives, together with the greater BML is coherent with the augmentation of the number of ULC previously reported after deep dives. This suggests that the increase in central blood pressure, due to the immersion-induced blood shift, would be further amplified by the high environmental pressure. Besides a higher diuresis, as suggested by the BML, this would lead to increased EVLW and decreased FVC with depth. This hypothesis should be addressed in future studies. Moreover, although this might have led to altered pulmonary gas exchanges, it is noteworthy that, in our study, SpO_2 remained unchanged after the dives. Whether, it is because pulmonary gas exchanges were not impaired post-dive, as previously reported after the dives up to 50 msw (Ljubkovic et al. 2010), or because their alteration were compensated by hyperoxia is still to be clarified.

During diving, cardiac autonomous nervous system (ANS) is challenged by various factors. Previous studies reported that heart rate variability is increased by immersion (Schipke and Pelzer 2001), elevated environmental pressure (Barbosa et al. 2010; Noh et al. 2018), and hyperoxia (Lund et al. 1999; Schirato et al. 2018), whereas it is decreased by cold temperature (Lundell et al. 2020) and decompression stress (Schirato et al. 2020). Overall, the combination of these factors usually results in increased HRV during diving in neutral or medium temperature water (Chouchou et al. 2009; Noh et al. 2018) and decreased HRV in cold water (Lundell et al. 2020, 2021), as assessed by the modifications in SDNN and RMSSD. It is stated that the increase in HRV during diving results from both the increase in vagal tone and the decrease in sympathetic activity which are elicited mainly by the immersion-induced increase in central blood volume (Schipke and Pelzer 2001). When measured post-dive, heart rate variability was either unchanged (Chouchou et al. 2009; Schirato et al. 2018; Lundell et al. 2021) or increased (Schirato et al. 2020) post-dive, as indicated by increased RMSSD and SDNN. Interestingly, Schirato et al. (2018) reported that both SDNN and RMSSD are increased by breathing of hyperoxic mixtures even in the absence of hyperbaric exposure whereas SDNN is decreased correspondingly to decompression stress (Schirato et al. 2020). In line with these previous reports, besides they are indicative of an increased HRV post-dive, the statistically significantly higher SDNN and RMSSD we found after than before the dives suggest a moderate level of decompression stress, which would further support the hypothesis that the decreased FVC is not a consequence of decompression stress. They are also indicative of a remaining vagal activation which might result from either the exposition to hyperoxia during the dive and/or persistance of central blood shift. Although hypothetical, it would be coherent with the decreased FVC post-dive.

Besides its influence on physical and cognitive performance which may be important for safety during such extreme dives, dehydration is also an acknowledge risk factor for decompression sickness. Indeed, animal experiment showed that 4% BML increased the risk of DCS by 40% (Fahlman and Dromsky 2006). However, no signs of DCS were reported in our study, and the lack of post-dive modification of the critical flicker fusion frequency in our study does not support the hypothesis of impaired cognitive performance. Critical flicker fusion frequency test has proven to be a reliable tool for the objective evaluation of cognitive function and alertness (Lafère et al. 2019). In the context of scuba diving, it has been used for the evaluation of inert gas narcosis and/or hyperoxia. Only two studies assessed CFFF in relation with helium dives (Rocco et al. 2019; Vrijdag et al. 2020). Of these two studies, Vrijdag et al. (2020) did not find any change of the CFFF during simulated dives at 608 kPa with heliox, whereas Rocco et al. (2019) reported an increased fusion frequency which persisted at the end of 50 msw depth dives breathing either heliox or trimix. They attributed these results to the excitatory effect of hyperoxia which was not counterbalanced by the narcotic inhibitory effect of nitrogen, which was either low (for trimix) or absent (with heliox). The lack of post-dive modification of the critical flicker fusion frequency in our study does not support the hypothesis of impaired cognitive performance, coherent with these previous studies.

The main limitation of this preliminary study is clearly the small number of subjects. Spirometry is an accessible, non-invasive and relatively simple mean for testing pulmonary function and, as such, it is invaluable for in-field studies. However, although FVC was significantly decreased post-dive in our study, lung volume measurement is needed in the case of restrictive process before being able to formally conclude with sufficient confidence.

Conclusion

In conclusion, the present observation represents the first original data regarding the effects of deep trimix repetitive sea dives. It represents the first, although preliminary, evidence for building our knowledge allowing developing comprehensive diving guidelines regarding deep CCRs diving. Indeed, we found a 3% body mass loss and almost 30% decrease of FVC after bounce dives at depth of about 100 msw which may possibly impose an important physiological stress for the divers. However, as stated above, more studies are needed to be able to precisely determine prerequisites for fitness to dive and diving contraindications.

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Author contributions ED, EG, FG conceived and designed the UTP3 research. ED, EG conducted UTP3 measurements. CB designed and conducted the Gombessa 5 research. ED, FG, EL analyzed data. ED, CB, EG, EL and FG wrote the manuscript. All authors read and approved the manuscript.

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Availability of data and material The datasets generated during the current study are available from the corresponding author on reasonable request.

Declarations

Conflict of interest ED is the president of Tek Diving s.a.s., a R&D company dedicated to the development of safety procedures for diving. At the time of the study, EG was in charge of the medical survey of Under The Pole divers.

Consent to participate All participants were informed of the measurements objectives, procedures, potential risks, discomforts and benefits associated with their involvement. Informed consent was obtained from all individual participants included in the study.

Ethical approval The study was conducted during working dives performed by the Under The Pole (UTP) 3 team in the frame of the medical survey of divers participating to the Deep Hope scientific program. It was performed in accordance with the ethical standards as laid down in the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards.

Code availability Not applicable.

References

Arieli R (2019) Calculated risk of pulmonary and central nervous system oxygen toxicity: a toxicity index derived from the power equation. Diving Hyperb Med 49:154–160. https://doi.org/10.28920/dhm49.3.154-160

Bao X-C, Chen H, Fang Y-Q et al (2015) Clopidogrel reduces the inflammatory response of lung in a rat model of decompression sickness. Respir Physiol Neurobiol 211:9–16. https://doi.org/10.1016/j.resp.2015.02.003

Barbosa E, García-Manso JM, Martín-González JM et al (2010) Effect of hyperbaric pressure during scuba diving on autonomic modulation of the cardiac response: application of the continuous wavelet transform to the analysis of heart rate variability. Mil Med 175:61–64. https://doi.org/10.7205/MILMED-D-02-00808

Bosco G, Rizzato A, Quartesan S et al (2018) Spirometry and oxidative stress after rebreather diving in warm water. Undersea Hyperb Med 45:191–198

Brubakk AO, Ross JAS, Thom SR (2014) Saturation diving; physiology and pathophysiology. In: Terjung R (ed) Comprehensive physiology. John Wiley & Sons Inc, Hoboken, NJ, USA, pp 1229–1272

Castagna O, Blatteau J-E, Vallee N et al (2013) The underestimated compression effect of neoprene wetsuit on divers hydromineral homeostasis. Int J Sports Med 34:1043–1050. https://doi.org/10.1055/s-0033-1345136

Castagna O, Desruelle A, Blatteau J-E et al (2015) Alterations in body fluid balance during fin swimming in 29 °C water in a population of special forces divers. Int J Sports Med 36:1125–1133. https://doi.org/10.1055/s-0035-1555854

Castagna O, Gempp E, Poyet R et al (2017) Cardiovascular mechanisms of extravascular lung water accumulation in divers. Am J Cardiol 119:929–932. https://doi.org/10.1016/j.amjcard.2016.11.050

Castagna O, Regnard J, Gempp E et al (2018) The key roles of negative pressure breathing and exercise in the development of interstitial pulmonary edema in professional male SCUBA divers. Sports Med-Open. https://doi.org/10.1186/s40798-017-0116-x

Castagna O, Bergmann C, Blatteau JE (2019) Is a 12 h nitrox dive hazardous for pulmonary function? Eur J Appl Physiol 119:2723–2731. https://doi.org/10.1007/s00421-019-04248-w

Chouchou F, Pichot V, Garet M et al (2009) Dominance in cardiac parasympathetic activity during real recreational SCUBA diving. Eur J Appl Physiol 106:345–352. https://doi.org/10.1007/s00421-009-1010-0

Cirillo I, Vizzaccaro A, Crimi E (2003) Airway reactivity and diving in healthy and atopic subjects. Med Sci Sports Exerc 35:1493–1498. https://doi.org/10.1249/01.MSS.0000084424.67486.5B

Degryse J, Buffels J, Van Dijck Y et al (2012) Accuracy of office spirometry performed by trained primary-care physicians using the MIR spirobank hand-held spirometer. Respiration 83:543–552. https://doi.org/10.1159/000334907

Dujic Z, Marinovic J, Obad A et al (2011) A no-decompression air dive and ultrasound lung comets. Aviat Space Environ Med 82:40–43

Fahlman A, Dromsky DM (2006) Dehydration effects on the risk of severe decompression sickness in a swine model. Aviat Space Environ Med 77:102–106

Fock AW (2013) Analysis of recreational closed-circuit rebreather deaths 1998–2010. Diving Hyperb Med: J S Pac Underw Med Soc 43:78–85

Fock A, Harris R, Slade M (2013) Oxygen exposure and toxicity in recreational technical divers. Diving Hyperb Med 43:67–71

Gaustad SE, Kondratiev TV, Eftedal I, Tveita T (2020) Continuous hemodynamic monitoring in an intact rat model of simulated diving. Front Physiol 10:1597. https://doi.org/10.3389/fphys.2019.01597

Godfrey MS, Jankowich MD (2016) The vital capacity is vital. Chest 149:238–251. https://doi.org/10.1378/chest.15-1045

Graham BL, Steenbruggen I, Miller MR et al (2019) Standardization of spirometry 2019 update. An official American thoracic society and European respiratory society technical statement. Am J Respir Crit Care Med 200:e70–e88. https://doi.org/10.1164/rccm.201908-1590ST

Hong SK, Claybaugh JR (1989) Hormonal and renal responses to hyperbaria. In: Claybaugh JR, Wade CE (eds) Hormonal regulation of fluid and electrolytes. Springer US, Boston, MA, pp 117–146

Krol K, Morgan MA, Khurana S (2019) Pulmonary function testing and cardiopulmonary exercise testing. Med Clin North Am 103:565–576. https://doi.org/10.1016/j.mcna.2018.12.014

Lafère P, Hemelryck W, Germonpré P et al (2019) Early detection of diving-related cognitive impairment of different nitrogen-oxygen gas mixtures using critical flicker fusion frequency. Diving Hyperb Med 49:119–126. https://doi.org/10.28920/dhm49.2.119-126

Ljubkovic M, Gaustad SE, Marinovic J et al (2010) Ultrasonic evidence of acute interstitial lung edema after SCUBA diving is resolved within 2–3 h. Respir Physiol Neurobiol 171:165–170. https://doi.org/10.1016/j.resp.2010.02.008

Lund, Scheinin K et al (1999) Heart rate variability in healthy volunteers during normobaric and hyperbaric hyperoxia: heart rate variability during hyperoxia. Acta Physiol Scand 167:29–35. https://doi.org/10.1046/j.1365-201x.1999.00581.x

Lundell RV, Räisänen-Sokolowski AK, Wuorimaa TK et al (2020) Diving in the arctic: cold water immersion's effects on heart rate variability in navy divers. Front Physiol 10:1600. https://doi.org/10.3389/fphys.2019.01600

Lundell RV, Tuominen L, Ojanen T et al (2021) Diving responses in experienced rebreather divers: short-term heart rate variability in cold water diving. Front Physiol 12:649319. https://doi.org/10.3389/fphys.2021.649319

Marinovic J, Ljubkovic M, Obad A et al (2010) Assessment of extravascular lung water and cardiac function in trimix SCUBA diving. Med Sci Sports Exerc 42:1054–1061. https://doi.org/10.1249/MSS.0b013e3181c5b8a8

Mitchell SJ, Doolette DJ (2013) Recreational technical diving part 1: an introduction to technical diving methods and activities. Diving Hyperb Med 43:86–93

Noh Y, Posada-Quintero HF, Bai Y et al (2018) Effect of shallow and deep SCUBA dives on heart rate variability. Front Physiol. https://doi.org/10.3389/fphys.2018.00110

Papadopoulou V, Tang M-X, Balestra C et al (2014) Circulatory bubble dynamics: from physical to biological aspects. Adv Coll Interface Sci 206:239–249. https://doi.org/10.1016/j.cis.2014.01.017

Rocco M, Pelaia P, Di Benedetto P et al (2019) Inert gas narcosis in scuba diving, different gases different reactions. Eur J Appl Physiol 119:247–255. https://doi.org/10.1007/s00421-018-4020-y

Schipke JD, Pelzer M (2001) Effect of immersion, submersion, and scuba diving on heart rate variability. Br J Sports Med 35:174–180. https://doi.org/10.1136/bjsm.35.3.174

Schirato SR, El-Dash I, El-Dash V et al (2018) Heart rate variability changes as an indicator of decompression-related physiological stress. Undersea Hyperb Med 45:173–182

Schirato SR, El-Dash I, El-Dash V et al (2020) Association between heart rate variability and decompression-induced physiological stress. Front Physiol 11:743. https://doi.org/10.3389/fphys.2020.00743

Shykoff BE, Florian JP (2018) Pulmonary effects of repeated six-hour normoxic and hyperoxic dives. PLoS ONE 13:e0202892. https://doi.org/10.1371/journal.pone.0202892

Tetzlaff K, Friege L, Koch A et al (2001) Effects of ambient cold and depth on lung function in humans after a single scuba dive. Eur J Appl Physiol 85:125–129. https://doi.org/10.1007/s004210100421

Vrijdag XC, van Waart H, Sleigh JW et al (2020) Investigating critical flicker fusion frequency for monitoring gas narcosis in divers. Diving Hyperb Med 50:377–385. https://doi.org/10.28920/dhm50.4.377-385

Wilson A (2011) Prevalence and characteristics of lung function changes in recreational scuba divers. Prim Care Respir J 20:59–63. https://doi.org/10.4104/pcrj.2010.00063