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Critical flicker fusion frequency results during oxygen decompression in standard HBOT session — observational study

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ABSTRACT

Most hyperbaric medicine reports concentrate on the patient and his morbidities. In addition to the well-known indications for hyperbaric oxygen therapy (HBOT), we cannot discount possible side effects. Among medical staff regularly exposed to hyperbaric conditions the best described so far is decompression sickness. A non-invasive and easily available way to assess cognitive functioning involves the use of the critical flicker fusion frequency (CFFF) test. In the current study, the flicker test was performed several times on 21 subjects, both under normobaric and hyperbaric conditions. The test was conducted using the device that flickering was programmed according to the method of limits. While in the hyperbaric chamber, 15 of the participants breathed oxygen to reduce the risk of decompression sickness. Flicker and fusion frequencies differed from each other in both normo- and hyperbaric conditions ($p < 0.01$). CFFF results were dependent on oxygen breathing during decompression.

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Keywords: flicker test, critical flicker fusion frequency, CFFF, flicker, fusion, oxygen, hyperbaric oxygen therapy

INTRODUCTION

Critical flicker fusion frequency (CFFF) is the frequency at which flickering light appears continuous (fusion) or begins to be perceived as flashing (flicker). It reflects the upper limit of visual processing abilities, known as the critical flicker fusion threshold. CFFF is used as an index of cerebral nervous system function, indicating alertness and cortical arousal. Numerous factors influencing CFFF have been described in previous articles, including characteristics of both the individual and the light. One of its biggest advantages is that CFFF is not affected by the learning effect, so it can be repeatedly assessed even within short intervals [1, 2].

The critical flicker fusion frequency (CFFF) test (or *flicker test*) involves focusing on a light source and pressing a button when the flickering stops or starts to become visible. The most common method of performing the flicker test is the method of limits, which is based on presenting flickering light with ascending and descending frequency, resulting in fusion and flicker frequencies, accordingly [3]. Following this, the CFFF threshold is calculated as the average of the results of fusion and flicker. There are reports of differences between CFFF test results under conditions of increasing (fusion) and decreasing (flicker) frequency. However, these differences can vary over time and may depend on the subject's health status [4]. Due to its characteristics, the flicker test can be successfully used in unusual, extreme conditions, i.e., diving and hyperbaric chambers. Oxygen (or other breathable gases) levels, pressure, and depth

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impact CFFF test performance [5]. The National Centre for Hyperbaric Medicine in Gdynia (Poland) practices hyperbaric oxygen therapy (HBOT) in defined indications [6]. This study was based on attendant's exposure during each, standard hyperbaric oxygen therapy in our centre.

Standard hyperbaric oxygen therapy for conscious patients consists of three 20-minute sessions with 5-minute air intervals, performed a few times a day. During each session a patient with a particular indication breathes 100% oxygen at 2.5 ATA (15 meters' water equivalent). At the same time, the medical attendants who must obligatorily assist patient(s) in the chamber are at risk of decompression disease (DCS) [7–9]. This risk can be mitigated by pre-breathing 100% oxygen several minutes before starting decompression and during the whole process of decompression. Most staff follow this safety precaution, but it is only a recommendation rather than an obligation. Therefore, the decision whether to breathe oxygen or not during decompression is on attendant's site. In our centre, the same as during our observational study the decision making about breathing mixture during decompression remains on the attendant. There are described situations in which staff choose to breathe compressed air during the entire session [9–11].

The role of oxygen in human body is far from being obvious. Apart from its role in respiratory chain, molecular reactions and organs functioning, the oxygen can be toxic as well [10]. Effects of oxygenation are well-known in relation to respiratory and circulatory system. Neurotoxicity and pulmonary toxicity are well-described [12, 13].

The effects of oxygen on the brain are not yet well understood, and exploring this topic is important for specific occupational groups such as hyperbaric workers, divers and astronauts who are at risk for DCS [14]. The possible mechanism has been described by Bliznyuk et al. [15] basing on N-methyl-D-aspartate receptor (NMDAR) and its role in cognitive deficits especially under the pressure over 2 ATA (10 meters' water equivalent).

Also, changes in neurological functioning due to gas bubbles can significantly affect neurological functions. As HBOT may impact cognitive functioning [16], therefore we compared CFFF test results among medical staff who are regularly exposed to hyperbaric conditions during working hours, dividing them into oxygen and air groups, as depending on which breathing gas (oxygen or air) they used during decompression.

RESEARCH ASSUMPTIONS

Breathing ambient air (1 ATA, 760 mmHg) consisting of 20.9% oxygen delivers a pO_2 (oxygen partial pressure) of approx. 158 mmHg. Therefore, while breathing 100% oxygen at 2.5 ATA, the human body is exposed to pO_2 1900 mmHg. Hyperoxia leads to the production of reactive oxygen species, which increase neuronal electrical activity because of membrane lipid peroxidation and changes in enzymatic functioning [10]. Any neurologically toxic effects that are dependent on oxygen partial pressure and exposure time are reversible as soon as the oxygen supply is interrupted.

The central nervous system is primarily affected by hyperoxic conditions, and the most frequently observed symptoms of oxygen neurotoxicity are changes in alertness and consciousness, visual disturbances, perception abnormalities, and clonic seizures in some cases [13].

The main aim of this study was to compare CFFF test results during oxygen and air decompression. We also planned to compare flicker and fusion conditions, as they can generate differences in flicker test results. As hyperbaric oxygen impacts cognitive functioning [16], our aim was to compare CFFF results among medical staff who are regularly exposed to hyperbaric conditions during working hours, marking them as oxygen and air groups. The division into oxygen and air group is quasi-random as the decision whether to breathe air or oxygen during decompression remains on the attendants.

We hypothesized that:

— There are differences in flicker test results between oxygen and air groups during decompression.
— There are differences between the frequencies obtained in flicker and fusion conditions.

MATERIALS AND METHODS

The study was conducted at the Department of Hyperbaric Medicine and Maritime Rescue — National Center for Hyperbaric Medicine of the Medical University of Gdansk and involved 21 healthy individuals working there (16 women and 5 men), aged 28–62 (mean age = 48.14; standard deviation = 9.97), who had no contraindications to being in a hyperbaric chamber (neurological and respiratory diseases, previous laryngological and thoracic surgeries, cancerous diseases in advanced stages, pregnancy). The oxygen group consisted of 15 subjects (5 men, 10 women, aged 28–62, mean age = 48.27, median age = 52, standard deviation = 11.34), while the air group — 6 women (aged 42–53, mean age = 47.83, median age = 48, standard deviation = 3.39). All subjected agreed freely and signed informed consent. The study obtained a consent of Bioethics Committee for Scientific Research at Medical University of Gdansk, Poland.

Subjects performed a trial of 6 measurements in the flicker test, standardized according to the criteria described in Table 1. The diode of the flicker test device (shown in Fig. 1) was 4 mm in diameter and was flickering with a light blue color presented from about 30 cm.

Table 1. Parameters of flicker test devices

Parameter	Value
Minimum frequency	10.0 Hz
Maximum frequency	50.0 Hz
Frequency change rate	1.0 [Hz/s]
Frequency change step	0.01 [Hz]

Figure 1. The device used in the flicker test

Each series included performance of the flicker test:

- under normobaric conditions before the start of the session (1 ATA, air),
- after compression to hyperbaric conditions (2.5 ATA) using air,
- during the hyperbaric session (2.5 ATA/air 60 min),
- before decompression (2.5 ATA, 100% oxygen or air),
- after decompression (1 ATA, 100% oxygen) before exiting the hyperbaric chamber,
- 5 minutes after the end of the hyperbaric session (1 ATA, air).

Since subjects participated in each condition a few times, the flicker test provided total 92 measurements. Measurements were taken 2 to 6 times for each person, depending on the availability of participants during standard hyperbaric oxygen therapy sessions.

We used the Wilcoxon signed-rank test to compare the CFFF results between flicker and fusion paradigms. We analyzed the results without grouping, as well as for groups of oxygen and air breathers. Each pair (flicker-fusion) was considered a separate observation, as they were performed as part of a single flicker test, and the Wilcoxon signed-rank test does not assume the need to collect measurements from different individuals. The Mann-Whitney U test was used to compare air and oxygen groups across 3 parameters (flicker, fusion, average) at all 6 stages of the study, was used, as the results of the two groups are independent of each other — each participant consistently, every time, breathed either oxygen or air. These statistical tests were chosen due to their robustness in analyzing non-normally distributed data and their ability to handle small sample sizes effectively.

RESULTS

Given the observed deviations from normality in the distribution of our data, as assessed by the Shapiro-Wilk test, we employed nonparametric tests for all analyses. Consequently, median values were utilized as central measures, providing a more representative characterization of the dataset. Missing observations were excluded from the analysis. The results were considered statistically significant at a p-value < 0.05. The statistical software packages Statistica Version 13.3 and IBM SPSS Statistics 29.0 was used for calculations.

We found differences between flicker and fusion at each time point (1–6) for both the oxygen and air breathing groups and among all measurements using Wilcoxon signed-rank test. In each case, the frequencies obtained in the fusion subtest with increasing frequency were significantly lower than in the flicker subtest with decreasing frequency (1: $p = 0.006$; 2, 3, 5 and 6: $p < 0.001$; 4: $p = 0.002$). The differences in the absolute values of the medians of these results were respectively: 2.87, 2.07, 3.91, 2.75, 4.51 and 4.32 Hz. A summary of the results is presented in Table 2. For easier readout, they are also presented on Figure 2 in changes of percentage values, where 100% is taken as baseline (test at 1 ATA air).

Given the non-parametric nature of our data, Spearman rank correlation tests were employed to assess relationships between flicker, fusion and average frequencies of the CFFF test (Tab. 3). The test stages, numbered from 1 to 6, follows the stages shown above in Table 2. The values given therein represent Spearman's rho correlation coefficient. The statistically significant correlation values at the $p < 0.001$ level are marked in orange, while those at the $p < 0.05$ level are marked in green. At the different stages of the test under normo- and hyperbaric conditions, the flicker subtest scores were strongly correlated both with each other and with the averaged CFFF test scores ($p < 0.001$). This correlation was not observed for fusion and flicker results, except during the test under 2.5 ATA and 100% oxygen conditions ($r = 0.24$; $p < 0.05$). However, the fusion results were partially correlated with the averaged results of the CFFF test.

Comparison of oxygen-breathers with air-breathers during decompression showed significant differences in all results in the flicker test for the decreasing frequency

Table 2. Comparison of results of flicker and fusion frequencies in the flicker test

Test stage		Flicker [Hz]			Fusion [Hz]			N	W	Z	p
		Me	min	max	Me	min	max				
Without grouping											
1	1 ATA air	40.37	25.44	49.80	37.50	28.40	49.25	89	1325.5	2.770	0.006*
2	2.5 ATA air	40.08	20.73	49.84	38.01	23.38	49.42	89	1149	3.492	0.000*
3	2.5 ATA air 60 min	40.90	30.64	49.83	36.99	28.94	49.27	85	1003	3.613	0.000*
4	2.5 ATA O2	39.69	28.75	49.77	36.94	30.13	49.29	72	772.5	3.039	0.002*
5	1 ATA postdeco	41.28	29.19	49.78	36.77	29.17	49.22	90	924	4.521	0.000*
6	1 ATA 5 min postdeco	41.23	26.72	49.81	36.91	27.05	49.4	89	917.5	4.439	0.000*
Air group											
1	1 ATA air	46.12	28.66	49.56	37.53	28.40	45.29	24	2	–3.346	<0.001*
2	2.5 ATA air	45.90	34.75	49.34	38.15	23.38	47.14	24	9	–3.555	<0.001*
3	2.5 ATA air 60 min	44.65	36.65	49.48	36.71	29.12	45.26	18	10	–2.911	0.004*
4	2.5 ATA O2	47.43	35.88	49.34	38.95	35.12	44.55	13	2	–2.903	0.004*
5	1 ATA postdeco	46.42	30.23	49.23	37.32	29.17	43.75	24	5	–3.772	<0.001*
6	1 ATA 5 min postdeco	45.32	36.03	49.45	37.65	29.05	45.48	23	7	–3.88	<0.001*
Oxygen group											
1	1 ATA air	37.57	25.44	49.80	37.42	31.07	49.25	66	780	–0.834	0.404
2	2.5 ATA air	39.18	20.73	49.84	37.98	31.38	49.42	68	679	–1.780	0.075
3	2.5 ATA air 60 min	38.22	30.64	49.83	37.05	28.94	49.27	68	601	–2.365	0.018*
4	2.5 ATA O2	37.53	28.75	49.77	36.81	30.13	49.29	60	645	–1.958	0.050*
5	1 ATA postdeco	39.68	29.19	49.78	36.74	29.98	49.22	67	575	–2.605	0.009*
6	1 ATA 5 min postdeco	38.10	26.72	49.81	36.84	27.05	49.4	67	559	–2.467	0.014*

Me — median; min — minimum value; max — maximum value; N — sample size; W — Wilcoxon signed-rank test; Z — Z-score; p — p-value (significant values at the < 0.05 level was marked with *)

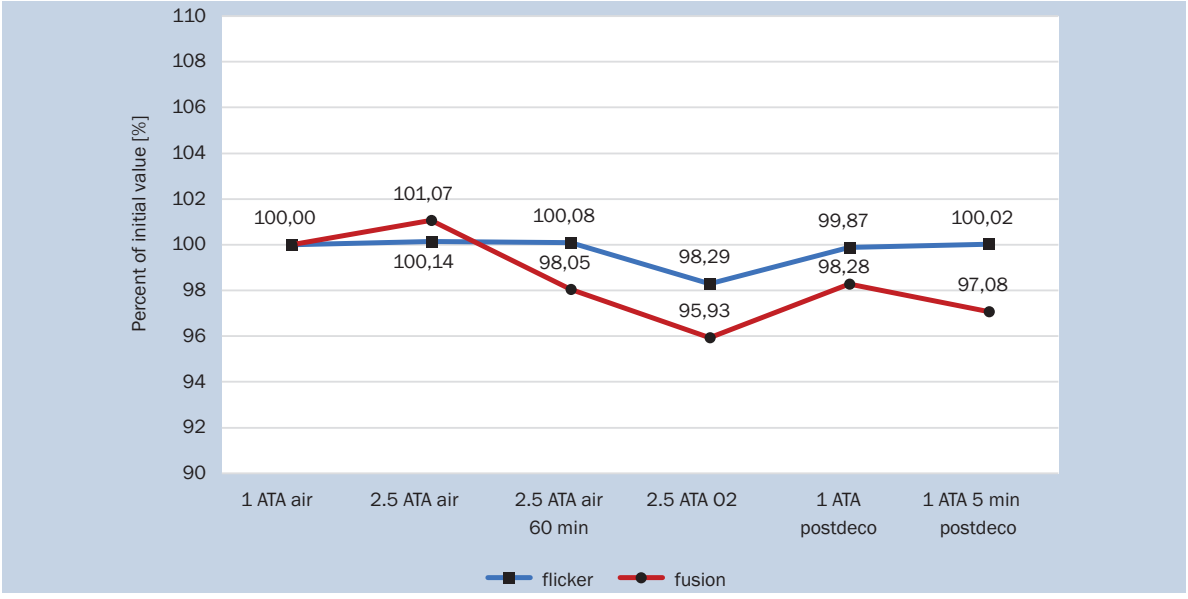


Figure 2. Relative changes of flicker and fusion frequencies for all measurements (measured as percentage of initial value — results at stage 1 ATA air) between measurements at different test stages ATA = atmosphere absolute

Table 3. Correlation matrix for results of CFFF subtests and their average

CFF subtest/ Test stage		Flicker						Fusion						Average					
		1	2	3	4	5	6	1	2	3	4	5	6	1	2	3	4	5	6
Flicker	1																		
	2	0.67																	
	3	0.67	0.84																
	4	0.72	0.86	0.93															
	5	0.68	0.85	0.92	0.93														
	6	0.68	0.85	0.93	0.92	0.91													
Fusion	1	0.06	−0.03	−0.11	−0.04	0.01	−0.08												
	2	−0.03	0.02	−0.08	0.05	0.04	−0.03	0.60											
	3	−0.06	−0.06	−0.07	−0.03	−0.05	−0.12	0.48	0.63										
	4	0.22	0.19	0.19	0.24	0.20	0.21	0.43	0.60	0.57									
	5	0.07	0.11	0.01	0.06	0.07	−0.01	0.61	0.71	0.61	0.69								
	6	0.00	0.03	−0.10	0.01	0.00	−0.01	0.51	0.58	0.52	0.59	0.65							
Average	1	0.86	0.54	0.51	0.56	0.56	0.52	0.37	0.25	0.15	0.36	0.32	0.23						
	2	0.49	0.85	0.69	0.73	0.72	0.71	0.24	0.44	0.26	0.41	0.42	0.32	0.56					
	3	0.53	0.71	0.85	0.78	0.80	0.77	0.12	0.23	0.37	0.41	0.27	0.15	0.57	0.79				
	4	0.60	0.73	0.80	0.87	0.81	0.82	0.15	0.31	0.22	0.59	0.33	0.30	0.60	0.80	0.87			
	5	0.56	0.76	0.80	0.80	0.86	0.77	0.25	0.35	0.26	0.51	0.49	0.32	0.62	0.85	0.89	0.89		
	6	0.54	0.72	0.79	0.78	0.78	0.86	0.16	0.20	0.13	0.43	0.29	0.41	0.57	0.78	0.83	0.87	0.85	

task (flicker, $p < 0.05$) and in the averaged flicker and fusion scores ($p < 0.05$), but not in the fusion subtask. The exception was the fusion subtask performed under oxygen therapy conditions (2.5 ATA, 100% O_2), where the results were statistically significant at $p = 0.01$. Detailed information is presented in Table 4. Figure 3 presents results for flicker, fusion and average frequencies separately at each test stage. The lines in the box-plots indicate median values.

DISCUSSION

The main finding of this study is that the CFFF test results were affected in the oxygen group. In particular, we observed that hyperbaric oxygen might exert either slowing down or stimulating effect, depending on the pressure. As shown in Table 4, flicker results were of higher frequency in the air-group, while in the oxygen group a slight slowing down was observed. Comparing the results of flicker and fusion frequencies there was a noticeable difference in medians. Median flicker results were the lowest at the pressure 2.5 ATA while breathing 100% oxygen. However flicker median while breathing air was still higher after the exposure at 1 ATA in relation to flicker median at 1 ATA before the hyperbaric exposure. Basing on median fusion results they were comparable under the pressure of 2.5 ATA. Whilst median fusion results were lower at the pressure 2.5 ATA, particularly while breathing 100% oxygen in comparison to air-breathing group. However, as a result the average shows statistically significant differences in the oxygen group overall, which may suggest that the averaged result of CFFF test may not be sensitive enough to changes occurring in flicker and fusion recognition.

The study has some limitations. Our sample size of the participants was relatively small, limiting the statistical power and potentially affecting the ability to detect subtle effects. In hyperbaric medicine field of research, obtaining a more representative group and repeating CFFF tests are difficult due to the limitations of access to medical workers, who are often exposed to oxygen. Notwithstanding, our observations of the effects of oxygen decompression CFFF results in medical attendants suggest that decompression on oxygen can induce a feeling of slowing down; at the same time, breathing oxygen at 2.5 ATA appears to have a stimulating effect. These results confirm that the effects of oxygen described by Kot et al. [17] are not specific only to representatives of special forces units, but also to hyperbaric unit personnel, who have about 1 exposure to oxygen per day. However, the procedure of that study differed from the current one — while that one considered exposure to 0.7, 1.4 and 2.8 ATA of oxygen, our study focuses on differentiating decompression on and without oxygen. Thus, this effect could be related to the frequent use of high partial pressures of oxygen, which is typical both for special forces divers and medical attendants. According to Kot and colleagues' results, in the range of about 1–2 ATA, oxygen slows down the specialized individual's functioning due to its narcotic effect, and above 2.5 ATA, the excitability of nerve cells increases [17]. However, that study was conducted on a different group of subjects (81 subjects, young males only) and main aim of that study was to investigate if neuronal excitability is dose dependent. The CFFF results were compared at the pressure of 0.7 ATA, 1.4 ATA and 2.8 ATA. In our study we differentiated flicker and fusion results depending on air or 100% oxygen as breathing mixtures at 1 ATA and 2.5 ATA, there was not a comparison in CFFF results depending on the oxygen dose.

The effects of hyperbaric oxygen on the organism are therefore complex, and it is difficult to determine where the line between a positive cognitive effect and a toxic effect lies as this effect seems to be dose dependent [18, 19]. However, it is possible that oxygen has a narcotic effect at doses between 1–2 ATA and decompression from 2.5 ATA on oxygen may be safer by avoiding excessive excitation of the central nervous system. Basing on our results showing a slightly slowing down in oxygen group, perhaps dose dependent effect induces excitation or inhibition of central nervous system.

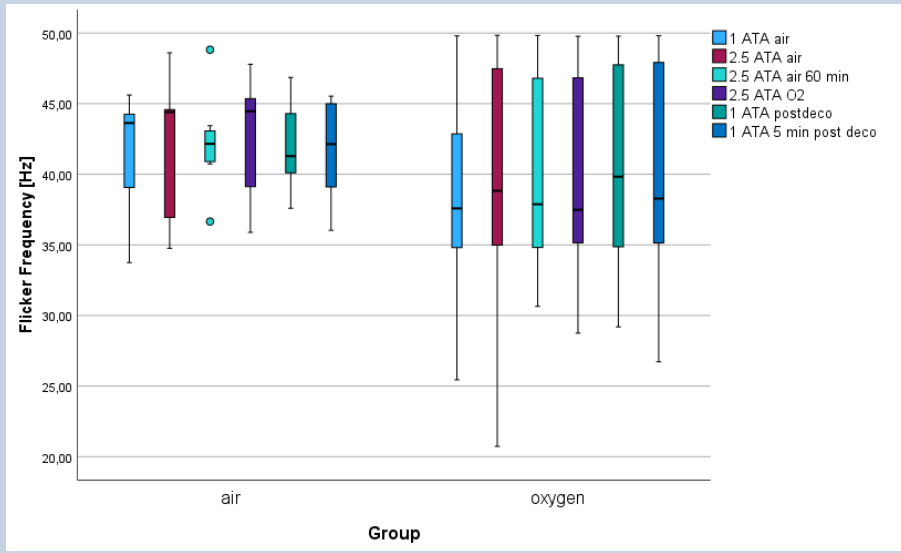
We also found the differences between flicker and fusion thresholds, that may result from individual factors or different neurophysiological and neuropsychological processes. As recently Haarlem et al. [20] indicated, the CFFF test results are relatively unchanged over time, but can depend on individual factors. Although they found no gender differences in CFFF results, it is possible that CFFF results are less stable among women, suggesting the existence of further variables that should be investigated. It is not ruled out that this can also explain outliers in the case of the data we obtained. According to Carmel et al. [21], the perception of flicker may be associated with higher activity in bilateral frontal and left parietal cortex, while perceiving of fusion is mainly related to functioning of occipital cortex. Following that, the processing of flickering may also be influenced by retinal properties [22–24]. Our finding is consistent with other studies addressing this issue [25, 26], but to date a more precise explanation for these differences is unknown. As our data show, the differences in flicker and fusion results occur regardless of the breathing mixture used by the participants. This may be related to the limitations of the limit method of testing, which, while quite popular, may not be very robust to extreme responses, as Haarlem et al. (2024) also found. Thus, this indicates a lack of validity for using averaged CFFF threshold values.

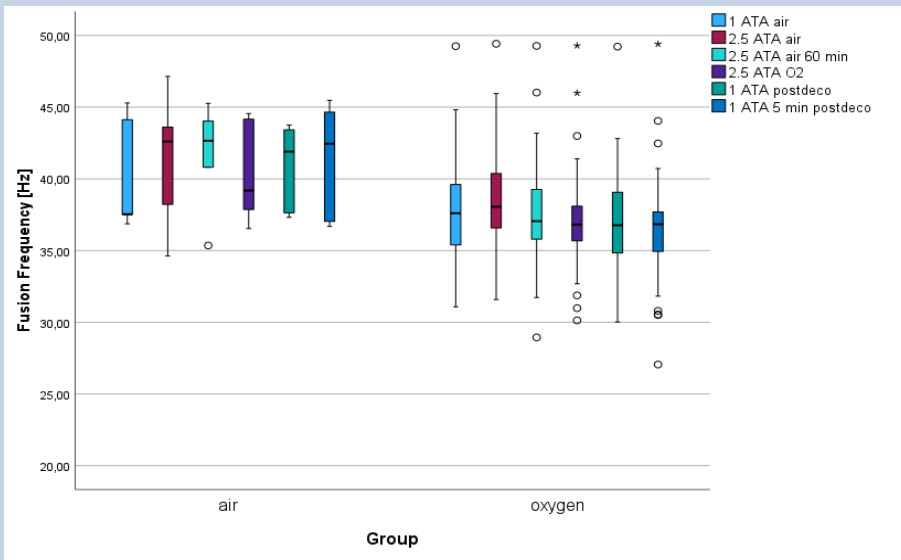
Inert gas narcosis cannot be dismissed in hyperbaric conditions. Under the pressure of 2.5 ATA (15 meters' water equivalent), not only oxygen partial pressure increases,

Table 4. Comparison of the CFFF results between oxygen and air groups

CFFF subtest/Performing time		Air					Oxygen					U	p
		Me	min	max	SD	N	Me	min	max	SD	N		
Flicker	1 ATA air	46.12	28.66	49.56	6.16	24	37.57	25.44	49.80	5.82	66	456,0	0.002*
	2.5 ATA air	45.90	34.75	49.34	4.87	24	39.18	20.73	49.84	6.37	68	512,5	0.007*
	2.5 ATA air 60 min	44.65	36.65	49.48	4.12	18	38.22	30.64	49.83	6.06	68	367,5	0.009*
	2.5 ATA O_2	47.43	35.88	49.34	4.51	13	37.53	28.75	49.77	6.26	60	216,0	0.012*
	1 ATA postdeco	46.42	30.23	49.23	4.86	24	39.68	29.19	49.78	6.31	67	491,5	0.005*
	1 ATA 5 min postdeco	45.32	36.03	49.45	3.91	23	38.10	26.72	49.81	6.61	67	474,0	0.006*
Fusion	1 ATA air	37.53	28.40	45.29	3.77	23	37.42	31.07	49.25	3.35	67	681,0	0.408
	2.5 ATA air	38.15	23.38	47.14	4.74	22	37.98	31.38	49.42	3.28	68	703,5	0.676
	2.5 ATA air 60 min	36.71	29.12	45.26	4.03	17	37.05	28.94	49.27	3.36	68	565,5	0.891
	2.5 ATA O_2	38.95	35.12	44.55	2.88	12	36.81	30.13	49.29	3.19	60	191,0	0.011*
	1 ATA postdeco	37.32	29.17	43.75	3.51	23	36.74	29.98	49.22	3.33	68	645,5	0.213
	1 ATA 5 min postdeco	37.65	29.05	45.48	3.77	22	36.84	27.05	49.40	3.35	67	605,5	0.211
Average	1 ATA air	41.60	30.85	46.59	4.50	24	38.34	22.94	45.94	3.86	67	485,0	0.004*
	2.5 ATA air	42.31	25.65	46.10	5.29	24	39.01	29.09	46.20	3.47	68	593,0	0.047*
	2.5 ATA air 60 min	41.91	30.44	45.73	3.66	18	38.31	31.59	46.21	3.37	68	418,0	0.039*
	2.5 ATA O_2	42.83	31.00	45.47	4.25	13	37.24	32.01	44.89	3.54	60	211,0	0.010*
	1 ATA postdeco	42.11	28.31	45.12	4.07	24	38.55	31.49	45.76	3.42	68	492,0	0.004*
	1 ATA 5 min postdeco	42.02	28.43	45.09	4.00	23	38.21	20.41	46.24	3.98	67	437,0	0.002*

Me — median; min — minimum value; max — maximum value; N — sample size; U — Mann-Whitney U test; p — p-value. The p-values marked with * indicate statistically significant values ($p < 0.05$)





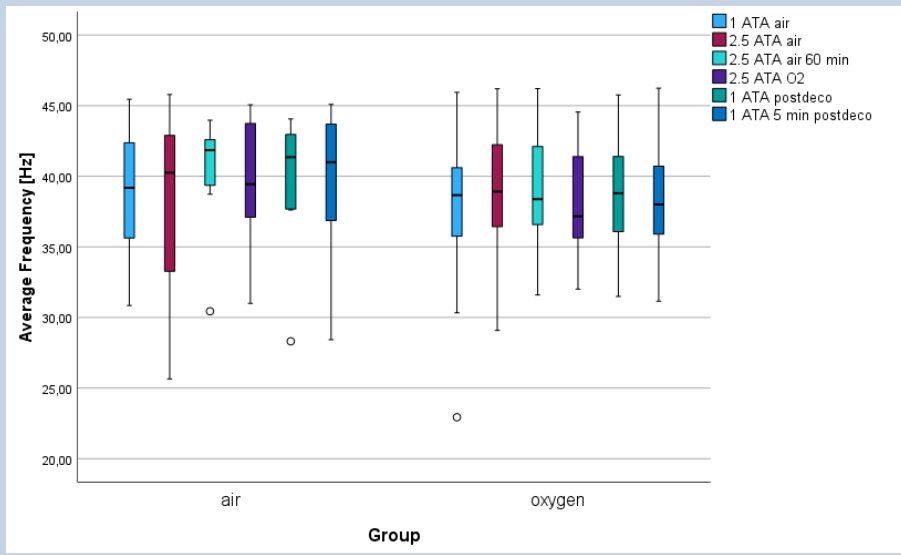


Figure 3. Comparison of the CFFF test results (flicker, fusion and average frequencies) between oxygen and air groups at each of the 6 stages of the study

but also partial pressure of each component in the breathing mixture. Undoubtedly, most publications concentrate on nitrogen narcosis [27, 28]. Its symptoms may include mood changes and cognitive functioning impairment. Therefore, there still remains a question, what is the exact impact of so-called Martini's effect on CFFF results despite our results concerning on oxygen and air groups [29]. Highlighting that hyperbaric medical staff regularly exposed to hyperbaric conditions during working time may not demonstrate mentioned symptoms due to habit, adaptation or lower sensitivity [30].

The precise oxygen exposure among medical personnel may be difficult to estimate. Depending on the mileage of HBOT session, oxygen decompression may be interrupted by patient emergency. Therefore, from medical attendant's point of view there may be a hesitancy in oxygen or air decompression in relation to decompression disease or theoretical oxygen toxicity during such a short period of time. Time of exposure and oxygen percentage plays the role. As for today, breathing 100% oxygen during decompression is a recommendation. In case of decompression sickness among medical staff, the question of not breathing 100% oxygen during decompression remains unanswered.

CONCLUSIONS

We found that breathing oxygen had a slowing down or stimulating effect, depending on the pressure (flicker vs fusion frequency). Decompression from 2.5 ATA on oxygen may be safer, not only due to decreasing the risk of decompression sickness, but also due to diminished excitation of the central nervous system. Inferring from above results, more research should be performed to define oxygen dose dependent effect on central nervous system.

ARTICLE INFORMATION AND DECLARATIONS

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