Functional comparison between critical flicker fusion frequency and simple cognitive tests in subjects breathing air or oxygen in normobaria

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Abstract

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Introduction: Measurement of inert gas narcosis and its degree is difficult during operational circumstances, hence the need for a reliable, reproducible and adaptable tool. Although being an indirect measure of brain function, if reliable, critical flicker fusion frequency (CFFF) could address this need and be used for longitudinal studies on cortical arousal in humans.

Methods: To test the reliability of this method, the comparison between CFFF and three tests (Math-Processing Task, Trail-Making Task, and Perceptual Vigilance Task) from the Psychology Experiment Building Language battery (PEBL) were used to evaluate the effect of 10 minutes of 100% normobaric oxygen breathing on mental performance in 20 healthy male volunteers.

Results: Breathing normobaric oxygen significantly improved all but one of the measured parameters, with an increase of CFFF (117.3 ± 10.04% of baseline, $P < 0.0001$) and a significant reduction of time to complete in both the math-processing (2,103 ± 432.1 ms to 1,879 ± 417.5 ms, $P = 0.0091$) and trail-making tasks (1,992 ± 715.3 to 1,524 ± 527.8 ms, $P = 0.0241$). The magnitude of CFFF change and time to completion of both tests were inversely correlated (Pearson r = -0.9695 and -0.8731 respectively, $P < 0.0001$). The perceptual vigilance task did not show a difference between air and O_2 ($P > 0.4$).

Conclusions: The CFFF test provides an assessment of cognitive function that is similar to some tests from PEBL, but requires a less complicated set up and could be used under various environmental conditions including diving. Further research is needed to assess the combined effects of increased pressure and variations in inspired gas mixtures during diving.

Key words

Air, oxygen, narcosis, performance, psychology, research

Introduction

For the diver, the probability of being victim of a narcotic event is higher than that of having a decompression accident. Yet fundamental studies about the mechanisms of inert gas narcosis (IGN), conducted mainly in the context of air diving, saw a rapid decline linked to the introduction of helium diving.[1] However, the popularity of recreational diving and, in particular, technical diving, calls for further study.

Nitrogen narcosis occurs in man at around 0.4 MPa and includes spatial and temporal disorientation, euphoria, hallucinations, disruption in motor and locomotor coordination, mood changes and cognitive impairment.[2] The behavioural approach to its study claims that the majority of the cognitive deficits are caused by a single fundamental deficit: the slowing of information processing due to decreased arousal, which is controlled within the reticular formation.[3,4] This hypothesis seems to be confirmed by neurochemical studies on rats.[5]

One of the most compelling questions within the field of neuroscience is how the complexity of human behaviour emerges from activities at the synaptic and cellular level and, in spite of the continuing advancements in technology, the current and dominant emphasis within this field largely fails to address questions and applications outside the laboratory.[6] This is especially true in the case of IGN as neurophysiologic or neurochemical measurements could be difficult to assess directly underwater, undoubtedly because of the lack of available technical and methodological means.

The re-emergence of the critical flicker fusion frequency (CFFF) test in experimental diving medicine may address this need. Historically a correlation between change in the mental state of divers, CFFF and electro-encephalogram (EEG) has been reported.[7] Also, changes in CFFF during a helium-oxygen dive to 62 ATA (6.28 MPa) showed systematic variations and a relationship between compression and pressure.[8] These variations were grossly parallel to EEG modifications. Since EEG reflects a range of complex brain activities, CFFF, which appears to correlate with EEG, might usefully reflect changes in brain function.[9] Unfortunately, subsequent investigators could not replicate these earlier results, and the use of CFFF was abandoned, it being considered unreliable and non-specific.[10,11]

Outside of the field of diving, despite some limitations, several authors have emphasized the advantages of CFFF assessment as a simple, objective, quantitative method for measuring alertness and arousal in humans.[12–17] In studies during anaesthesia, CFFF revealed brain impairment earlier than some behavioural tests or subjective symptom appearance.[18, 19] Under standard conditions, the CFFF test may be used for longitudinal studies on cortical arousal in humans.[13]

The purpose of the present study was to assess brain performance using CFFF and behavioural (psychometric) tests to validate the possibility of detecting changes while breathing 100% oxygen (O_2) or air under normobaric conditions. The choice of O_2 was made because previous studies of IGN showed that the changes observed were also related to the oxygen partial pressure.[20–22] Should the results be favourable, the method could then be extended to the evaluation of performance under various environmental diving conditions.

Materials and methods

Experimental procedures were conducted in accordance with the Declaration of Helsinki and were approved by the Academic Ethical Committee of Brussels (CE-B200-2011-5). All methods and potential risks were explained to 20 male volunteers who gave their written, informed consent prior to the experiment. All subjects were healthy non-smokers, undertook regular physical activity (aerobic exercise one to three times a week), were on no medications and had no history of migraine. The participants were instructed not to take any alcoholic beverages for 72 hours and no caffeine-containing beverages for 4 hours before the experiments.

PROCEDURES

All tests were carried out in a quiet room at a constant temperature of 22°C to avoid any disturbance of concentration. All subjects were asked to wear a Tru-fit Mask with a demand valve (Life Support Products, USA) and breathe either air or O_2 for 10 minutes in a randomized order. After 10 minutes for each breathing condition, three tests from the Psychology Experiment Building Language (PEBL) battery were performed and immediately after this the CFFF test was undertaken. The total procedure time was 16.5 ± 1.55 min.

CRITICAL FLICKER FUSION FREQUENCY TEST (CFFF)

In the present study, CFFF was assessed with a specific watertight device (Human Breathing Technology, Trieste, Italy).[10,11] The device consists of a rotating ring, surrounding a short cylindrical waterproof plastic housing of 8 cm diameter containing the numeric (digital) frequency indicator covered by an acrylic transparent window. Attached to this housing a flexible cable is connected to a single blue light emitting diode (LED, colour temperature 8000 Kelvin), inserted in a small, waterproof, cylindrical container (to shield it from stray light and reflections). During the test the subject is looking straight at the LED at a distance individually adapted to their personal vision, generally around 50 cm. The investigator increases or decreases the flickering frequency of the LED by operating the rotating ring in the appropriate direction.

Thanks to the design of the device, subjects are not aware of the starting and actual flicker frequency before, during and after the test. When the subject sees the light change from fusion to flicker (or flicker to fusion), the subject signals the investigator and the test is stopped. The frequency reached is recorded. Each subject performed the test three times and the average was calculated for statistical analysis.

THE PSYCHOLOGY EXPERIMENT BUILDING LANGUAGE BATTERY TESTS (PEBL)

PEBL tests were specifically chosen to track deterioration in visual-perceptual organization, visual-motor coordination as well as integration, and visual memory.[23] Three PEBL tests were used: math processing, trail making and perceptual vigilance.

Math-processing task (MathProc)
The subject is asked to subtract and/or add one- or two-digit numbers presented on a screen and to assess whether the result is more or less than five in a maximum 4-second time frame; this procedure is limited to 2 minutes. Time to complete the task within the 4-sec time frame, and correct, incorrect and timed-out answers are used for analysis.

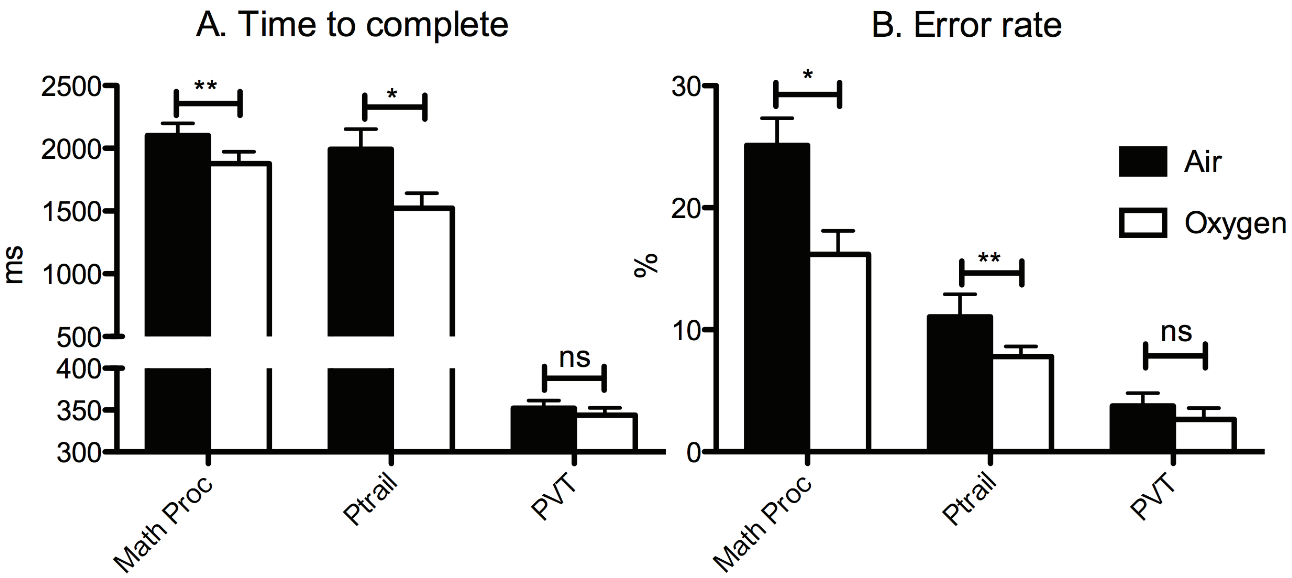
Trail-making task (Ptrail)
This test is used to assess brain injury, hand-eye coordination and general intelligence. The test is divided into 2 parts. In the first part, the circles are numbered (1, 2, 3, etc.) and the subject has to connect them in numerical order (1, 2, 3, etc.). In the second part, the circles have both numbers and letters and the subject has to click on them in alternating order (1-A, 2-B, 3-C, etc.). The trial continues until the test person has connected all the circles in the correct order. The number of clicks to finish the test and erroneous clicks are stored for analysis.

Perceptual vigilance task (PVT)
This test is commonly used to measure simple response time. Using a computer screen and a keyboard, the participant has to press the spacebar as quickly as possible when a red circle stimulus appears randomly at delays between 2 and 12 seconds for 16 times for a maximum of 2 minutes. The reaction times are captured for analysis.

STATISTICS

As they are time-limited tasks, each subject performed a different number of calculations for MathProc or simple response times for PVT in the defined time periods. Before analysis, we calculated the mean for each test and participant in order to obtain a unique set of 20 measures for each breathing condition. Since all data passed the Kolmogorov-

Figure 1
A. Time to complete MathProc and Ptrail tests and reaction time (PVT)
B. Error rate (%) in the three PEBL tests used during air or oxygen breathing (ns = not significant; * $P < 0.05$; ** $P < 0.01$)



Smirnov and Shapiro-Wilk tests, allowing us to assume a Gaussian distribution, they were analysed with a Student's paired test and two-way ANOVA.

Taking the initial (pre-breathing) values as 100%, percentage changes were calculated for each parameter, allowing an appreciation of the magnitude of change rather than the absolute values. Then, a possible correlation was assessed through a Pearson test and linear regression. All tests were performed using a standard computer statistical package, GraphPad Prism version 5.00 for Windows (GraphPad Software, San Diego, California, USA). A threshold of $P < 0.05$ was considered statistically significant. All data are presented as mean ± standard deviation (SD).

Results

Mean age of the 20 subjects was 25 ± 6.6 years; weight 71.4 ± 9.5 kg; height 1.77 ± 0.06 m; BMI 22.8 ± 2.0 kg m^{-2}.

CRITICAL FLICKER FUSION FREQUENCY TEST

The mean CFFF while breathing 100% normobaric O_2 was significantly higher (117.3 ± 10.04 %, $P < 0.0001$) than when the subjects breathed air (baseline, 100%).

PEBL TESTS

In general, breathing normobaric O_2 significantly improved all measured parameters. The mean time to complete the math-processing task while breathing air was 2,103 ± 432 ms and, for trail-making, 1,992 ± 715 ms. After O_2 breathing, these times significantly decreased to 1,879 ± 418 ms (P = 0.0091) and 1,524 ± 528 ms (P = 0.0241) respectively (Figure 1A). A comparable effect was observed in the math-processing error rate from 25.1 ± 10% to 16.2 ± 8.6% (P = 0.0263) and, for trail-making, from 11.06 ± 8.29% to 7.8 ± 3.7% (P = 0.0066) (Figure 1B). The perceptual vigilance task did not show a significant difference between air and O_2 ($P > 0.4$). All values have been rounded up or down.

Two-way ANOVA showed a 6.8% chance of randomly observing as much interaction between the PEBL tests and O_2 breathing. Therefore, results of each test are not influenced by other tests (F = 2.75, DFn = 2, DFd = 114, P = 0.068).

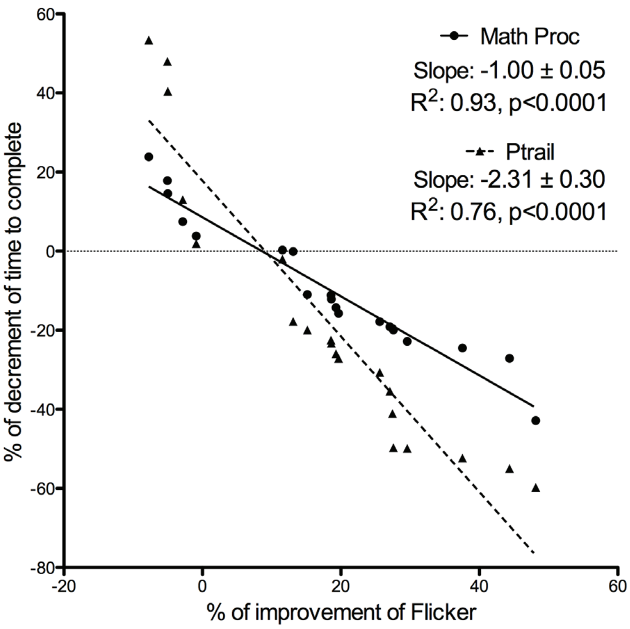
Oxygen breathing affects both time to complete (1.86% of the total variance) and error rate of MathPoc and PTrail (4.58% of the total variance) as there is respectively a 0.43% (F = 8.51, DFn = 1, DFd = 114, P = 0.0043) and a 0.008% (F = 11.87, DFn = 1, DFd = 114, P = 0.0008) chance of randomly observing an effect this big in an experiment of this size.

The kind of test accounted for 72% of the variance. This suggests that all tests are not equal in detecting a modification of brain performance (F = 164.29, DFn = 2, DFd = 114, $P < 0.0001$). This assumption is logical since we did not observe a significant change in the perceptual vigilance task.

COMPARISON BETWEEN CFFF AND PEBL TEST

The magnitude of CFFF change and the time to complete both the math-processing and trail-making tasks are inversely correlated (Pearson r = -0.9695 and -0.8731 respectively; Figure 2). Since $P < 0.0001$, we can reject the idea that the correlation is due to random sampling. This relation was further confirmed by linear regression.

Figure 2
Correlation calculation and linear regression of the magnitude of CFFF change and time to complete the MathProc and Ptrail tests



Discussion

The environmental characteristics of diving include pressure and breathing various gas mixtures adapted to the planned depth. According to Dalton's and Boyle's Laws, inert gas partial pressure increases with depth, and may generate significant nervous system dysfunction with disturbances of all memory, intellectual operation and locomotor activity.[2] As a consequence, the diver's safety may be impaired. Therefore, there is a need for an objective, valid and reliable measurement tool to evaluate brain performance in divers. Ideally, these indices should be reproducible, less subject- or investigator-dependent than a psychometric behavioural approach, based on observing a change in neurological parameters like EEG measurement, but also easy to implement underwater. Unfortunately this tool does not seem to exist unless we consider the use of CFFF, as has been done in several studies.[7,8,10,11] However, CFFF has never been validated or correlated to an independent, reliable set of brain-performance evaluations. Since there are two effects to consider (pressure and the nature of the gas breathed), the present study focuses on the easiest condition to test first – the effect of the mixture breathed under normobaric conditions – in order to later differentiate this effect from those of pressure or environment.

Normobaric hyperoxia has been shown to accelerate nerve conduction and although the exact mechanism is still debated, we would expect to see an improvement in brain performance. The results of the MathProc and Ptrail tests and the changes in the CFFF support this. Also, the regression graph shows a significant inverse correlation between CFFF and the time to completion of these tasks, suggesting that these tests might be considered comparable in providing assessment of cortical functions.

This may be because of the neural pathways involved in these processes. When experiments combine neural and mental chronometry, the contribution of perceptual and motor processes to the duration and variability of behavioural reaction time must be taken into account. Whereas perceptual processing accounts for a relatively constant amount of time for a given stimulus condition, the processes of mapping particular stimuli onto the appropriate behaviour and preparing the motor response provide flexibility but introduce delay and variability in reaction time.[24] Therefore, one of the causes for the difference in the reaction time could obviously be the nature of the task itself.[25] There are probably fewer processing stages for automatic attention to act upon in simple tasks (PVT) than in complex ones or in mechanisms less sensitive to automatic attention (MathProc and Ptrail). The idea that very different mechanisms mediate simple and complex tasks is certainly not new.[26] Also, automatic attention mechanisms could be set to operate at a lower gain level in simple tasks than in complex tasks as a consequence of the usual particular demands of these tasks. There is evidence for an adjustment of automatic attention mechanisms to task demands.[27,28]

Finally, there could be less room for automatic attention to reduce reaction time in simple tasks than in complex tasks because of the greater previous preparation to process the target stimulus in the former tasks than in the latter ones.[29] In simple terms, any increase in the number of synapses involved in a certain task would account for increased response time – each synaptic connection adding a delay of approximately 1 ms. The neural pathway for the PVT tests might involve fewer neural connections. Therefore, even if the conduction between neurons is increased because of the increased partial pressure of oxygen, this effect will not be noticeable since the number of synaptic connections involved is too small. Conversely, if many neural connections are involved, the integration over the whole path will yield a shorter time for the test, as shown for Ptrail and MathProc or a higher CFFF.

Conclusion

We conclude that the CFFF test provides an assessment of cognitive function that is similar to some tests from PEBL but requires a less complicated set up and could be used under various environmental conditions. Using CFFF, it would thus be possible to conveniently measure cognitive performance underwater. Further research is needed to assess the combined effects of increased pressure and variations in inspired gas mixtures during diving.

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