Generation of Reactive Oxygen and Nitrogen Species during Hyperbaric Stress in Experienced Divers versus Naïve Subjects

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Introduction

Disturbances in blood flow or membrane homeostasis have been reported to promote production of reactive nitrogen and oxygen species (RNS; ROS) within the vasculature. When chronically imposed, disruptions in membrane homeostasis have also been shown to stimulate biochemical and morphological remodelling in vascular responses to these disturbances. Recent studies suggest that venous gas emboli following decompression indirectly affect membrane homeostasis via flow disturbance or mechanical injury to vascular endothelia cells (ECs). We examined the effects of decompression stress on the production of ROS/RNS and the activity of an anti-oxidant surperoxide dismutase (SOD) in naïve subjects and experienced divers following dives for 30 minutes at 18, 30 and 45 MSW.

Statistical Analysis

Data are expressed as means ± SEM.

One way ANOVA followed by Tukey test was used to examine for significant differences (*) between the naïve subjects and experienced divers, p £ 0.05.

Methods

Following medical screening and a short introductory dive, blood samples were taken from naïve subjects (n=10) and experienced divers (n=12) before and after decompression (DCIEM schedules) in a hyperbaric chamber for 30 minutes on air at pressures equivalent to 18 MSW, 30 MSW and 45 MSW, (interval between dives 7 < days > 10; *Figure 1*). Total nitric oxide production was spectrophotometrically measured @ 540 nm after assay of plasma samples with a Griess reagent kit (Assay Designs). Reactive oxidant generation by PMNs was determined cytometrically using the Bursttest (Phagoburst) commercial kit (Orpegen Pharma) containing the fluorogenic substrate, dihydrorhodamine (DHR) 123. Nitrated tyrosine peptides (N-Tyr) were measured spectrophotometrically @ 450 nm after assay of plasma samples (OxisResearch). Plasma SOD activity was assayed (Cayman Chemicals) and measured spectrophotometrically @ 450 nm.

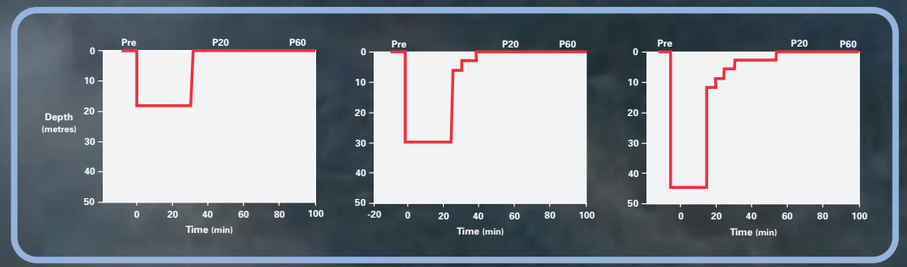

Figure 1
Blood sampling protocols before (Pre), 20 (P20) and 60 (P60) minutes after air dives for 30 minutes to 18, 30 and 45 MSW.

Results

VGE levels were generally higher in the experienced divers.

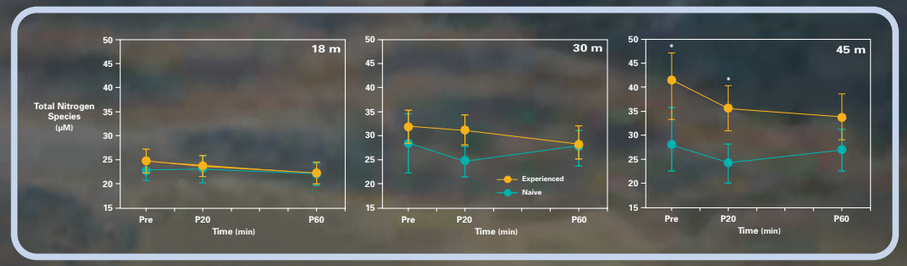

Figure 2
Nitric oxide production was higher in the plasma of experienced divers before and after the 45 m dive.

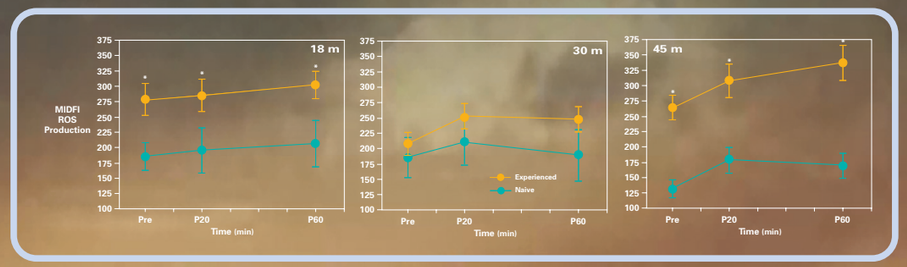

Figure 3
Production of reactive oxygen species was higher in PMNs of experienced divers than in naïve divers.

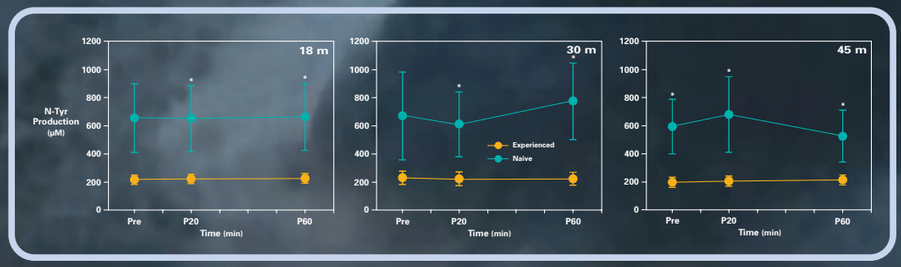

Figure 4
Levels of nitrated tyrosines were higher in the plasma of naive divers reflecting increased production of ONOO-.

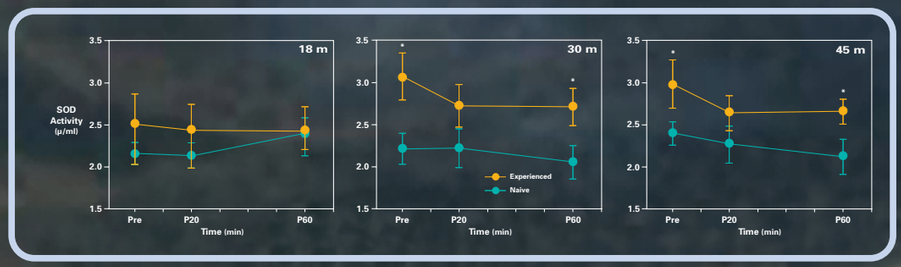

Figure 5
SOD activity was higher in the plasma of experienced divers following dives to 30 and 45 MSW.

Conclusions

In experienced divers, although decompression stress promoted higher ROS and .NO levels, ONOO-production was lower, possibly due to higher SOD activity.

Repeated disturbances in blood flow and/or low level injury to venous endothelium by VGE following decompression may confer the experienced diver with advantageously higher antioxidant levels to counter ROS/RNS production.

Although not previously considered similar to well-known EC injurious agents that can promote vascular remodeling, such as bacteria, parasites, antibiotics, interferons, etc., VGE may potentially cause more injury to endothelial cells in a shorter period of time.

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