Beta 1-blockade increases maximal apnea duration in elite breath hold divers

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We hypothesized that the cardioselective $\beta_1$-adrenoreceptor antagonist esmolol would improve maximal apnea duration in elite breath hold divers. In elite national level divers ($n=9$), maximal apneas were performed in a randomized and counterbalanced order while receiving either i.v. esmolol (150μg · kg$^{-1}$ · min$^{-1}$) or volume matched saline (placebo). During apnea, heart rate (ECG), beat-by-beat blood pressure, stroke volume (SV), cardiac output (CO) and total peripheral resistance (TPR) were measured (finger photoplethysmography). Myocardial oxygen consumption (MVO$_2$) was estimated from rate pressure product. Cerebral blood flow through the internal carotid (ICA) and vertebral arteries (VA) was assessed using Duplex ultrasound. Apnea duration improved in the esmolol trial when compared to placebo (356±57 vs. 323±61 seconds; $P<0.01$) despite similar end apnea peripheral oxyhemoglobin saturation (71.8±10.3% vs. 74.9±9.5; $P=0.10$). The HR response to apnea was reduced by esmolol at 10-30% of apnea duration, while MAP was unaffected. Esmolol reduced SV (main effect; $P<0.05$) and CO (main effect; $P<0.05$), and increased TPR (main effect; $P<0.05$) throughout apnea. Esmolol also reduced MVO$_2$ throughout apnea (main effect; $P<0.05$). Cerebral blood flow through the ICA and VA were unchanged by esmolol at baseline and the last 30 seconds of apnea; however, global cerebral blood flow was reduced in the esmolol trial at end apnea ($P<0.05$). Our findings demonstrate that, in elite breath hold divers, apnea breakpoint is improved by $\beta_1$-blockade, likely owing to an improved total body oxygen sparring through increased centralization of blood volume ($\uparrow$TPR) and reduced MVO$_2$.
NEW & NOTEWORTHY

The governing bodies for international apnea competition, the Association Internationale pour le Développment de l’Apnée and La Confédération Mondaile des Activités Subaquatiques have banned the use of beta-blockers based on anecdotal reports that they improve apnea duration. Using a randomized placebo controlled trial we are the first to empirically confirm that beta-blockade improves apnea duration. This improvement in apnea duration coincided with a reduced myocardial oxygen consumption.
INTRODUCTION:

Voluntary apnea elicits an integrative circulatory and neural response to maintain adequate perfusion of vital organs (e.g. the brain), while simultaneously reducing both flow to non-vital organs (e.g. skin and skeletal muscle) and total body oxygen consumption (9, 10, 20). Bradycardia, peripheral vasoconstriction, and centralization of blood volume are the primary cardiovascular responses to voluntary apnea, which together represent the mammalian diving reflex (20). During the initial phase (~25%) of a maximal apnea, the extreme lung volumes mechanically compress the heart while high intra-thoracic pressures impede venous return, resulting in an abrupt drop in stroke volume (SV), cardiac output (CO), and consequently mean arterial pressure (MAP) and cerebral blood flow [CBF; (5, 8, 26)]. Concomitant unloading of the baroreceptors causes a transient increase in muscle sympathetic nerve activity (7), heart rate (22), and total peripheral resistance (TPR). Following this initial phase, bradycardia (via reduced baroreflex output in addition to increases in vagal tone (18)) occurs alongside progressive increases in CO, MAP, TPR (5, 25) and CBF (23, 31). These elevations in CO, MAP, and TPR are thought to be mediated via marked elevations in sympathetic nerve activity (10-20 fold) (14, 29). Along with hypoxemic and hypercapnic mediated dilation of cerebral blood vessels, this increase in MAP aids in augmenting the CBF response to maintain cerebral oxygen delivery throughout the apnea (31).

The precise mechanism(s) for apnea breakpoint remain poorly understood (24), but in the motivated and elite breath hold diver are likely related in part to chemoreflex stress (3, 4) and a critical oxygen level required to maintain consciousness (4, 31). Additional mechanisms have been theorized to contribute to breakpoint in the untrained breath holder such as diaphragmatic afference (24), however, how this translates to the motivated and elite breath holder is unknown.
Esmolol and Maximal Apnea Duration

Beta (β) adrenoreceptor antagonists (β-blockers) have been commonly used in elite apnea competitions due to anecdotal reports of improved maximal apnea duration (personal correspondence, Croatian National Apnea Coach). As a result, the Association Internationale pour le Développment de l’Apnée (AIDA; http://www.aidainternational.org) and La Confédération Mondiale des Activités Subaquatiques (CMAS; http://www.cmas.org) prohibit the use of β-blockers in and out of competition. Given that recent work in elite apnea divers supports the presence of a critical oxygen tension as a major contributor to apnea break point (4), the present theory is that β-blockade improves maximal apnea duration via myocardial oxygen sparing, yet there are no published data demonstrating this effect. In the first randomized and placebo-controlled study of its kind, we determined the effects of β1-blockade on maximal apnea duration and the circulatory parameters that are characteristic of extreme apnea. It was hypothesized that β1-blockade would prolong apnea duration, owing to a reduced heart rate throughout the apnea and a consequently reduced rate of myocardial oxygen consumption (MVO2). We further reasoned that the effects of β1-blockade would be reflected in the cerebral vasculature through a lower CBF during baseline and end apnea compared to the placebo control due to a reduced CO and MAP.

METHODS:

Subjects
Elite breath hold divers (n=9; 1 female) were recruited from the National Croatian Apnea team to participate in this study. The divers were 29±9 years old, weighed 80±13Kg, and were 184±8cm tall (body mass index = 23.5±2.6). Personal record static apnea time was on average 394 seconds (range: 296-496), while divers had a mean of 4 years (range: 2-11) of experience in competitive apnea training and an FVC of 6.9L (range: 5.0-7.8). Following written informed consent, divers visited the laboratory on one occasion for the experimental session. Subjects arrived at either 8am or 12pm having abstained from alcohol, caffeine, and exercise for 24 hours prior to arrival. All subjects were free from any cardiovascular, respiratory, and cerebrovascular disease at the time of study as assessed by a screening questionnaire and spirometry. This study was approved by the University of British Columbia Clinical Research Ethics Board, and by the Ethical Committee of the University of Split School of Medicine and conformed to the Declaration of Helsinki.

**Experimental Design**

Upon arrival to the laboratory subjects first performed a spirometry test (Quark PFT, Cosmed, Rome, Italy) in the upright position. Subjects voided their bladders and were then instructed to rest supine, at which time a 20-gauge intravenous catheter was placed into an antecubital vein under local anesthesia (lidocaine 1.0%). Following cannulation subjects rested for ≥20 minutes during the set-up of experimental monitoring equipment.

This study implemented two experimental conditions: 1) cardiac specific β1-adrenergic receptor blockade (Esmolol; Esmocard, Austria, 10mg · mL⁻¹), and 2) a volume matched placebo (Saline; 0.9% NaCl) condition. Esmolol was first infused at a rate of 500μg · kg⁻¹ · min⁻¹ for one
minute followed by 50μg · kg⁻¹ · min⁻¹ for 5 minutes. This was repeated two additional times prior to a steady maintenance infusion of 150μg · kg⁻¹ · min⁻¹ for the remainder of the drug condition. These doses have previously been established to effectively reduce HR, systolic blood pressure, RPP, and both left and right ventricular ejection fraction during rest and exercise (16). The order of infusions was randomized in a counterbalanced manner and separated by >5 half-lives (60 minutes) (30). The participants and apnea coach were blinded to the experimental condition.

All experimental breath holds (maximal breath holds) were completed in the presence of the breath hold divers’ coach, the Croatian national apnea coach. The coach was present to ensure all divers were attaining a true maximal apnea. Each maximal breath hold was preceded by two practice breath holds. The preparatory breath holds included one at functional residual capacity; lasting until seven involuntary breathing movements (IBMs), and after two minutes of rest, a second breath hold at total lung capacity, lasting until ten IBMs. Subjects then rested quietly for six minutes in preparation for their maximal breath hold. The experimental breath hold was performed at TLC, while the extent of glossopharyngeal insufflation (lung packing) performed was based upon the individual capacity of each subject, but standardized between trials.

**Experimental Measures**

**Cardiovascular Measures.**
All cardiovascular and respiratory variables were sampled continuously at 1000Hz via an analog to digital acquisition system (Powerlab; ADInstruments, Colorado Springs, CO). Finger photoplethysmography (Finometer, Finapres Medical Systems, Amsterdam, Netherlands) was used to measure beat-by-beat blood pressure and estimate cardiac output (CO), stroke volume (SV), and total peripheral resistance (TPR), while standard three-lead electrocardiogram (ECG) was used to measure heart rate. Peripheral oxyhemoglobin saturation (SpO₂) was measured during baseline and throughout the apneas (Poet II, Criticare, USA). The IBM onset was recorded in real time during the apnea by the apnea coach and verified from a chest plethysmography belt. All data were interfaced with LabChart (version 7, ADInstruments) on a laboratory computer were stored for offline analysis.

Myocardial oxygen consumption

For each trial MVO₂ of the left ventricle was estimated from RPP (the product of systolic blood pressure and HR) and left ventricular mass. To calculate MVO₂ per 100g tissue, we used the equation reported by Gobel et al., 1978 (12):

\[
MVO_2 (mL \cdot 100 g^{-1} \cdot min^{-1}) = 0.08 (RPP \cdot 10^{-2}) - 0.15
\]

Left ventricular mass was estimated at baseline using echocardiography from a parasternal short-axis view in accordance with the recommendations of the American Society of Echocardiography (19) using the following equation:
(2) \( LV \text{ Mass (g)} = 0.8 \times 10.4 \times [(IVS + LVID + PWT)^3 - LVID^3] + 0.6g \)

where IVS and PWT are the diameters of the ventricular septum and inferolateral wall during diastole, respectively, and LVID is the internal diameter of the LV during diastole,

To then calculate absolute MVO\(_2\) for each subject, left ventricular mass was incorporated into the following equation on an individual basis:

(3) \( MVO_2 (mL \cdot min^{-1}) = \frac{Left \text{ ventricular mass (g)}}{100g} (0.08 \cdot (RPP \cdot 10^{-2}) - 0.15) \)

The cumulative myocardial oxygen consumption throughout apnea was then calculated by estimating MVO\(_2\) from RPP on a beat-by-beat basis. The volume cost of oxygen per beat was calculated by dividing beat-by-beat MVO\(_2\) by the R-R interval for each heart-beat. Total myocardial oxygen consumption for the entire apnea was calculated as the area under the curve of the beat-to-beat O\(_2\) cost.

Cerebrovascular Measures.

Blood velocity in the right middle cerebral artery (MCAv) and left posterior cerebral artery (PCAv) were measured using a 2MHz transcranial Doppler ultrasound (TCD; Spencer Technologies, Seattle, WA). The TCD probes were attached bilaterally to a specialized commercial headband (model M600 bilateral head frame, Spencer Technologies) and secured in place. Insonation of the MCA and PCA was performed through the trans-temporal window using previously described location and standardization techniques (32).
Blood flow through extra-cranial cerebral vessels was measured using a 10MHz multi-frequency linear array vascular ultrasound (Terason T3200, Teratech, Burlington, MA). The internal carotid (ICA) and vertebral artery (VA) were insonated ipsilateral to the MCA and PCA, respectively. Arterial diameter was measured via B-mode imaging, while peak blood velocity was simultaneously measured with pulse-wave mode. Measurements of ICA diameter and velocity (ICA_v) were acquired ~1.5cm distal to the common carotid bifurcation, with no evidence of turbulent or retrograde flow present during recording. Vertebral artery diameter and velocity (VA_v) were acquired between either the C4-C5 or C5-C6 vertebral segment. This location was determined on an individual basis in an attempt to select the most reproducible measures, with the same location repeated within subjects. Measurement location and insonation angle were standardized within subject and between the esmolol and placebo conditions. An angle correction of 60° was used for all subjects, while the velocity sample volume was placed in the center of the vessel and adjusted to span across the entire luminal diameter. Baseline recordings were made in all subjects (n=9); however, the sample size for VA measures throughout an entire breath hold was reduced (n=6) due to strict exclusion criteria such as obvious angle changes.

Ultrasound images of the extra-cranial vessels were recorded as video files at 30Hz and stored for offline analysis. Concurrent values for arterial diameter and peak blood velocity were acquired at 30Hz, using customized edge detection software designed to mitigate observer bias (33). Blood flow was subsequently calculated for each vessel using the following formula:

\[ Q_{\text{ICA}} \text{ or } Q_{\text{VA}} = \frac{\text{Peak Velocity}}{2} \cdot \pi \left(\frac{\text{Diameter}}{2}\right)^2 \]
Where $Q_{ICA}$ represents ICA blood flow, and $Q_{VA}$ represents VA blood flow. For the maximal apneas $Q_{ICA}$ and $Q_{VA}$ were averaged for a one minute baseline, and the last 30 seconds of apnea. Furthermore, values were also calculated on an individual basis to represent 10% increments in total apnea duration. No flow calculation included less than 12 consecutive cardiac cycles, and where possible included the entire averaging period. Once individual vessel flow was calculated, global cerebral blood flow ($gCBF$) was estimated for each apnea time point using the following formula:

$$gCBF = 2 (Q_{ICA} + Q_{VA})$$

During the latter half of each maximal apnea (i.e., struggle phase), IBM’s resulted in large diaphragmatic movements and neck muscle recruitment (i.e. sternocleidomastoid contraction). As such, reliable velocity traces were no longer attainable. Once measures of $Q_{ICA}$ and $Q_{VA}$ were no longer attainable due to poor velocity traces, flow was estimated using the following formulas in subsequent fashion for the remainder of the apnea:

$$eICAv \text{ or } eVAv = \text{Pre IBM ICAv or VAv} \cdot \%\Delta MCAv \text{ or } \%\Delta PCAv$$

$$Q_{ICA} \text{ or } Q_{VA} = eICAv \text{ or } eVAv \cdot \pi \left(\frac{d}{2}\right)^2$$

Where changes in MCAv and PCAv were used to calculate an estimated ICA and VA velocity ($eICAv \text{ & } eVAv$, respectively; equation 3). The term “Pre IBM ICAv or VAv” represents the average velocity value of the last 30 seconds before the IBMs that precluded the measurement of...
velocity occurred, and “%ΔMCAv or %ΔPCAv” represents the relative change in MCAv and PCAv from the same pre IBM stage. As such, this multiplication gives a reliable estimate of neck vessel blood velocity during the struggle phase of apnea in the event that changes in downstream vessel (MCA & PCA) velocity represent upstream vessel (ICA & VA) velocity changes. As reliable diameter measures \(d\) were still attainable throughout IBMs, changes in vessel diameter were accounted for in our estimation of volumetric blood flow (equation 4). Volumetric flow through the neck vessels (\(Q_{ICA}\) and \(Q_{VA}\)) was then calculated from the estimated velocity values and vessel cross sectional area \(\pi \left( \frac{d}{2} \right)^2\). To exemplify the feasibility of this estimation, we ran a Pearson’s correlation between the % changes in ICAv and VAv, with those of MCAv and PCAv, respectively, throughout apnea until velocity measures were unattainable. The average within subject \(R^2\) values between ICAv and MCAv were 0.88±0.12 and 0.90±0.10 during the placebo and esmolol trials, respectively. For VAv and PCAv the \(R^2\) average was 0.88±0.09 and 0.95±0.03 for the placebo and esmolol trials, respectively. The strong correlation between blood velocity of confluent intra and extracranial blood vessels supports the accuracy of our estimation. Further, we have previously demonstrated that MCAv and ICAv reactivity do not differ statistically over a wide range of end-tidal CO\(_2\) (Rest to +9mmHg) (15), while work in our laboratory also indicates ICAv and VAv reactivity do not differ from MCAv and PCAv reactivity respectively during hypoxia at an SaO\(_2\) of 98%, 90%, 80%, and 70% (unpublished observations).

**Statistical Analyses.**
Differences in apnea duration and time to IBM onset between esmolol and placebo were compared using two-tailed paired t-tests. To analyze the affect of esmolol over the progression of apnea, apnea duration was normalized to 100%, with comparisons being made between placebo and esmolol conditions at 10% relative increments in apnea duration using a two-way repeated measures ANOVA (factors: esmolol & apnea duration). When significant F-ratios were detected, post-hoc comparisons were made using Tukey’s HSD. End apnea variables (final 30sec) in the placebo and esmolol trials were compared using two-tailed paired t-tests (Table 1). Data are presented as mean±standard deviation.

RESULTS

Apnea Duration

Apnea duration increased 10±10% from 323.4±60.6 seconds during the placebo trial to 355.9±56.8 seconds after β₁-blockade with esmolol (P<0.01; Figure 1). The time to IBM onset was delayed by 21±15% [from 160.4±41.3 seconds during the placebo trial to 192.0±45.3 seconds after treatment with esmolol (P<0.01)]. There was no relationship between the absolute ($r^2=0.06; P=0.53$) or percent ($r^2=0.21; P=0.22$) change in IBM onset and apnea duration. Despite the prolonged apnea duration, SpO₂ during the last 30 seconds of apnea was not significantly different between placebo (74.9±9.5%) and (71.8±10.3%) esmolol trials (P=0.10).

Hemodynamics
Values and statistical output for HR, MAP, SV, CO, and TPR during each relative time point throughout apnea are presented in Figure 2. During the initial phase of apnea HR was elevated in both placebo and esmolol trials, while esmolol reduced HR compared to placebo at 10-30% of apnea duration. In both trials MAP was elevated from baseline. Throughout apnea, SV and CO were reduced from baseline, and lower (main effect: both $P<0.01$) during the esmolol compared to placebo trial from 40% to max apnea and 20% to max apnea, respectively. End apnea (final 30 seconds; Table 1) HR and MAP were not different between esmolol and placebo, while SV and CO were reduced by 15% and 18% in the esmolol trial, respectively; however, the difference in CO did not reach statistical significance ($P=0.06$). Esmolol increased TPR (main effect: $P=0.04$) compared to placebo, while within condition TPR was elevated above baseline from 20-100% of apnea duration.

There was a main effect of esmolol on MVO₂ ($P<0.01$; Figure 3), which was lower in the esmolol trial. In both trials MVO₂ was elevated from baseline at 30%, 40%, and 90%. There was a 16.2±4.2% reduction in MVO₂ across all stages (range: 8.0-21.6%) in the esmolol trial. However, there was no difference in the cumulative myocardial oxygen consumption between the placebo (76.3±19.9mL) and esmolol (70.2±21.7 mL) trials ($P=0.27$; Figure 3B) due to the prolonged duration of the esmolol apnea.

**Cerebral Blood Flow**

Measures of $Q_{ICA}$, $Q_{VA}$, $gCBF$, $MCAv$ and $PCAv$ were not different between placebo and esmolol at baseline (Table 2). At the end of apnea (final 30 seconds) the sample size for $gCBF$
and $Q_{VA}$ were reduced to six participants. In these six participants, gCBF was lower at the end of apnea during the esmolol trial compared to placebo (Table 2).

**DISCUSSION**

The primary findings of this study are: 1) $\beta_1$-blockade with esmolol improves maximal apnea duration in elite breath hold divers by ~10%, 2) esmolol reduces MVO$_2$ at each time point throughout maximal apnea by ~16%, while the cumulative myocardial oxygen consumption was unaltered, 3) there are marked changes in the hemodynamic response (TPR & CO) to apnea between esmolol and placebo trials, and 4) end apnea SpO$_2$ was not different despite the prolonged apnea in the esmolol trial. Collectively, these data support the hypothesis that apnea breakpoint is influenced by $\beta_1$-blockade but ultimately governed by a critical oxygen tension.

**$\beta_1$-blockade and Apnea Duration**

It is now recognized that $\beta$-blocker drugs act as an ergogenic aid and are therefore banned from international apnea competition by AIDA. In support of this rule, using a placebo-controlled and blinded design, this study is the first to empirically demonstrate the increased maximal apnea time with $\beta_1$-blockade in elite breath hold divers.

**Hemodynamic response to apnea**
β₁-blockade affects hemodynamics, and thus CO, through chronotropic and inotropic means (2, 16). In the present study we observed a predominately negative inotropic influence of β₁-blockade with esmolol on CO (reduced SV), with only slight chronotropic changes. With the exception of a lower HR during the initial hypotensive phase of apnea (first 10-30% apnea duration), esmolol did not have an appreciable affect on HR; MAP was not different from placebo at any relative time point during the esmolol trial apnea. However, esmolol did cause a reduction in SV throughout the apnea (likely through reduced contractility – i.e., negative inotropy), which caused a concomitant reduction in CO compared to the placebo trial. This reduction in CO would have likely acted to further baroreceptor unloading and augment the reflex increase in sympathetic output (and TPR) throughout the apnea (14) in order to maintain the MAP response. Despite a reduced CO, increased centralization of blood volume (due to ↑ TPR) in the esmolol trial likely reduced non-vital organ perfusion while aiding in the selective maintenance of oxygen delivery to vital organs.

Esmolol reduced left ventricular MVO₂ throughout apnea when compared to the placebo trial, indicating that whole heart MVO₂ would have likely been reduced (Figure 3). Reduced MVO₂ would further spare oxygen and also contribute in part to the prolonged apnea duration associated with β₁-blockade. Despite a lower MVO₂ throughout apnea, the cumulative myocardial oxygen consumption was not different between trials owing to the prolonged apnea in the esmolol trial.

Mechanisms Governing Apnea Termination
The physiological apnea breakpoint (i.e., cessation of breathhold) in untrained breath holders, which is observed as IBM onset in elite breath hold divers, is governed largely by chemoreceptor afferents (6, 11). Breskovic et al., 2012 suggested that IBM onset is related predominantly to a PaCO$_2$ threshold level of $6.5 \pm 0.5$ kPa (48.8mmHg). A similar PaCO$_2$ threshold for IBM onset has been reported in most (1, 21) but not all (4) earlier studies, highlighting the importance of PaCO$_2$ in initiating the physiological breakpoint. Nevertheless, the prevailing oxygen tension will impact the level of PaCO$_2$ at which IBM onset occurs (6). These chemoreflex interactions, along with a modulatory role from lung afferents (reviewed in: (24)) indicate that it is the integration of signals (i.e., peripheral and central chemo-afference) that collectively govern the physiological breakpoint.

Concurrent decreases in O$_2$ depletion and CO$_2$ production, both a consequence of a reduced metabolism, will act to attenuate arterial blood gas changes and hence chemoreflex stimulation. Indeed, our findings show that IBM onset was delayed by ~32 seconds, but nevertheless occurred at the same level of SpO$_2$ between trials (Placebo = 97.8±1.3%; Esmolol = 96.4±2.2%). Under the assumption that the respiratory exchange ratio was unaltered between trials, a similar reduction in SpO$_2$ signifies that metabolic CO$_2$ production likely did not differ and was also similar between trials at IBM onset. Therefore, despite the delayed IBM onset with esmolol treatment, IBMs likely occurred at the same level of chemoreflex stress with no influence from altered chemosensitivity (13) or CO$_2$ washout in the brainstem (i.e., Q$_{VA}$, an index of brainstem flow, was similar between trials). Therefore, it is likely that the delay in reaching a chemoreflex threshold was responsible for delayed IBM onset (or physiological breakpoint). In contrast to the physiological breakpoint, the actual apnea termination in elite breath hold divers appears to be only modestly affected by suppression of peripheral chemoreflex drive (4).
Typically it is accepted that man is incapable of sustaining a voluntary apnea to the point of losing consciousness (27). However, many elite apnea divers are able to suppress their ventilatory drive during an apnea to the loss of consciousness (unpublished observations). This latter point is reflected in that AIDA includes loss of consciousness as a criterion for disqualification during apnea competition. At this point, the apnea ‘breakpoint’ is potentially governed to a large extent by a threshold oxygen tension to maintain cerebral functioning and, therefore, consciousness. Current literature indicates this threshold occurs around 25-35mmHg PaO₂ (4, 31), which coincides with P50. Despite a prolonged apnea duration following esmolol in the current study, SpO₂ was not significantly different between trials indicating similar end apnea hypoxic stress. Based upon the commonly used Severinghaus equation (28) and our pulse-oximetry measures, it appears that end apnea PaO₂ would have been approximately 35-40mmHg in both the placebo and esmolol trials. While the achieved hypoxic stimulus does not appear as severe as previous study indicating the potential of a oxygen threshold for apnea termination (4), the similar end apnea SpO₂ indicates some role of hypoxia in mediating apnea breakpoint in both trials. Of note, data from our previous study (4) where concurrent SpO₂ (finger pulse-oximetry) and SaO₂ (radial artery blood draw) measures were collected, indicate that SpO₂ overestimates SaO₂ by a mean value of ~5%, and that this over estimation is largest below an SpO₂ of 70% (Figure 4). Therefore, it is likely the end apnea hypoxic stimulus was similar to our previous studies in elite breath hold divers (4, 31). These data are similar to our previous findings in elite breath hold divers of similar end apnea PaO₂, despite moderate prolongation (but in some cases reductions) in maximal apnea time following peripheral chemoreflex blunting with low dose dopamine (4).
In the current study, centralization of blood volume and therefore a reduction of non-vital organ perfusion but maintenance of vital organ oxygen delivery, in combination with myocardial oxygen sparring, likely reduced both the hypoxic and acidotic stresses characteristic of extreme apnea. Together, these factors likely underscore the increased apnea duration with esmolol.

Methodological Considerations

In the current study we have used RPP and echocardiographic assessment of left ventricular mass to estimate left ventricular MVO$_2$ from the equation reported by Gobel et al., 1978 (12). Although RPP has been shown as an excellent ($r=0.85$) correlate of MVO$_2$ during exercise pre- and post- β-blockade (propranolol) (17), we acknowledge that this validation has not been made during the specific conditions of prolonged apnea. Despite this, it is unlikely that our estimates of RPP and MVO$_2$ would differ between conditions and discredit our findings. Further, although we used comparable doses to other studies (16), we must acknowledge that we did not assess the completeness of our β$_1$-blockade. However, the presence of an incomplete block would have lead to an underestimation of the magnitude by which esmolol impacts apnea duration.

We used SpO$_2$ to quantify the hypoxic stimulus during prolonged breath hold during placebo (saline) and esmolol infusion. As demonstrated in Figure 4, SpO$_2$ over estimates SaO$_2$ substantially below a SaO$_2$ of ~70%. However, given the over estimation of SaO$_2$, our data should be interpreted as presenting similar end apnea hypoxic stress as previous investigations by our group (4, 31).
**Conclusion**

This study contributes to the recent body of literature (4, 31) supporting the emerging hypothesis that apnea breakpoint in elite breath hold divers is governed to a large extent by an oxygen threshold for the maintenance of consciousness. Cardiac specific β₁-blockade with esmolol improved apnea duration in the current study and thus acts as an effective ergogenic aid in apnea competition. The likely mechanisms by which esmolol increased apnea duration is through increased centralization of blood flow (due to ↑TPR), maintenance of vital organ perfusion, and a reduced MVO₂. Together these two factors would contribute to improved total body oxygen sparring and reduced metabolic CO₂ production.

**Acknowledgements**

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**Conflict of Interest**

The authors declare no conflict of interest, financial or otherwise.
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# TABLES

## Table 1. End Apnea hemodynamic variables.

<table>
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<th>Units</th>
<th>Placebo</th>
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<tr>
<td><strong>HR</strong></td>
<td>b \cdot min^{-1}</td>
<td>54.2±16.9</td>
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<td><strong>MAP</strong></td>
<td>mmHg</td>
<td>139.0±18.6</td>
<td>129.6±19.1</td>
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<tr>
<td><strong>SV</strong></td>
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<td><strong>CO</strong></td>
<td>L \cdot min^{-1}</td>
<td>5.4±2.4</td>
<td>4.4±1.3</td>
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<td><strong>RPP</strong></td>
<td>mmHg \cdot b^{-1} \cdot min^{-1}</td>
<td>13192±3882</td>
<td>10124±3318</td>
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<td><strong>Systolic BP</strong></td>
<td>mmHg</td>
<td>247.9±37.6</td>
<td>195.7±25.8</td>
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<tr>
<td><strong>SpO2</strong></td>
<td>%</td>
<td>74.9±9.5</td>
<td>71.8±10.3</td>
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† denotes a significant difference between placebo and esmolol, P<0.05. HR, heart rate; MAP, mean arterial pressure; SV, stroke volume; CO, cardiac output; RPP, rate pressure product; BP, blood pressure; SpO2, peripheral oxyhemoglobin saturation.

## Table 2. Baseline and end apnea cerebrovascular variables.

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<th>Baseline</th>
<th>Final 30 seconds</th>
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<td><strong>QICA</strong></td>
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</tr>
<tr>
<td><strong>QVA</strong></td>
<td>mL \cdot min^{-1}</td>
<td>74.3±35.5</td>
</tr>
<tr>
<td><strong>gCBF</strong></td>
<td>mL \cdot min^{-1}</td>
<td>583.6±99.8</td>
</tr>
<tr>
<td><strong>MCAv</strong></td>
<td>cm \cdot s^{-1}</td>
<td>57.3±11.5</td>
</tr>
<tr>
<td><strong>PCAv</strong></td>
<td>cm \cdot s^{-1}</td>
<td>41.0±13.4</td>
</tr>
</tbody>
</table>

As noted in the methods section, the values for $Q_{ICA}$, $Q_{VA}$, and $gCBF$ are based off of estimated velocity values combined with real diameter measures. † denotes a significant difference between placebo and esmolol, P<0.05; N=9 unless otherwise specified. $Q_{ICA}$, internal carotid artery blood flow; $D_{ICA}$, internal carotid artery diameter; $Q_{VA}$, vertebral artery blood flow; $D_{VA}$, vertebral artery diameter; $gCBF$, global cerebral blood flow; $MCAv$, middle cerebral artery blood velocity; $PCAv$, posterior artery blood velocity.
FIGURES

Figure 1. Maximal apnea duration during placebo and esmolol trials. Individual data is represented by open circles (○) and solid lines, while mean data is represented by filled squares (■) and the dashed line. Overall there was a 32±25 second increase in apnea duration with esmolol treatment. * denotes a significant difference in apnea duration (P<0.01).

Figure 2. The hemodynamic response to maximal apnea during placebo and esmolol infusion. Data are normalized within subjects to represent 10% increments in apnea duration. Closed circle (●) represent data from the placebo trial. Open squares (□) represent data from the esmolol trial. Data were analyzed using a two way repeated measures ANOVA with the factors of apnea duration and esmolol treatment. * significant difference from baseline P<0.05; † main effect of treatment P<0.05; ‡ interaction between apnea time and treatment. HR, heart rate; MAP, mean arterial pressure; SV, stroke volume; CO, cardiac output; TPR, total peripheral resistance.

Figure 3. Myocardial oxygen consumption (MVO₂) during maximal apnea in elite breath hold divers. A. Data are normalized within subjects to represent 10% increments in apnea duration. Closed circle (●) represent data from the placebo trial. Open squares (□) represent data from the esmolol trial. Data were analyzed using a two way repeated measures ANOVA with the factors of apnea duration and esmolol treatment. * significant difference from baseline P<0.05; † main effect of treatment P<0.05. B. Total MVO₂ throughout apnea was unaltered between placebo and esmolol trials. Individual data are represented by open circles (○) and solid lines, while mean data is represented by filled squares (■) and the dashed line.

Figure 4. A Bland-Altman plot of SaO₂ and SpO₂ at the end of a maximal apnea. This figure demonstrates an over estimation of SaO₂ (radial blood draw) by SpO₂ (finger pulse-oximetry) at the end of apnea in elite breath hold divers using data from our previous study (4). The dotted line represents the mean difference (5.3), while the dashed lines represent the upper and lower limits of agreement (16.1 and -5.6, respectively). Of note, the over estimation of SaO₂ by SpO₂ is most evident below a SaO₂ of 70%. Each data point represent an individual subject, n=14.
Apnea duration (%)

MVO$_2$ (mL/min)$^{-1}$

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**A**

Esmolol: $P=0.005$
Apnea Duration: $P=0.001$
Interaction: $P=0.159$

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**B**

Placebo  Esmolol
Mean of $\text{SpO}_2$ and $\text{SaO}_2$ (%)