Dynamic Cerebral Autoregulation During the Combination of Mild Hypercapnia and Cephalad Fluid Shift

Takuya Kurazumi; Yojiro Ogawa; Ryo Yanagida; Hiroshi Morisaki; Ken-ichi Iwasaki

BACKGROUND: Mild hypercapnia combined with a cephalad fluid shift [e.g., that occurring during spaceflight or laparoscopic surgery with head-down tilt (HDT)] might affect cerebral autoregulation. However, no reports have described the effects of the combination on dynamic cerebral autoregulation. Therefore, we tested the hypothesis that the combination of mild hypercapnia and a cephalad fluid shift would attenuate dynamic cerebral autoregulation.

METHODS: There were 15 healthy male volunteers who were exposed to 4 10-min protocols in which they received air in the supine position (Placebo/Supine), 3% carbon dioxide (CO_2) in the supine position (CO_2/Supine), air with −10° HDT (Placebo/HDT) and 3% CO_2 with −10° HDT (CO_2/HDT). Dynamic cerebral autoregulation was evaluated using a transfer function analysis of the beat-to-beat variability in mean arterial blood pressure (ABP) and mean cerebral blood flow (CBF) velocity.

RESULTS: The phase in the low-frequency range was significantly lower during CO_2/HDT than all other protocols, where CO_2/HDT was −25% lower than Placebo/Supine (CO_2/HDT, 0.49 ± 0.21; Placebo/Supine, 0.65 ± 0.16 radians). The transfer function gain in the low-frequency range was significantly higher during CO_2/HDT than all other protocols, where CO_2/HDT was 26% higher than Placebo/Supine (CO_2/HDT, 1.08 ± 0.34; Placebo/Supine, 0.86 ± 0.28 $cm \cdot s^{-1} \cdot mmHg^{-1}$). However, neither the CO_2/Supine nor Placebo/HDT showed significant differences compared with the Placebo/Supine.

DISCUSSION: Even short-term exposure to 3% CO_2 plus HDT increased synchrony and the magnitude of transmission between ABP and CBF in the low-frequency range. Thus, the combination of mild hypercapnia and a cephalad fluid shift attenuated dynamic cerebral autoregulation.

KEYWORDS: carbon dioxide, cerebral circulation, head-down tilt, transcranial Doppler, transfer function analysis.

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The combination of hypercapnia and a cephalad fluid shift is an extraordinary physiological state that might affect cerebral autoregulation. During spaceflight conditions, astronauts risk being in a state of mild hypercapnia because of the absence of natural air convection in microgravity.[16] In addition, a higher concentration (approximately 0.5%) of carbon dioxide (CO_2) exists aboard the International Space Station (ISS) than under standard Earth conditions.[15] A recent study reported an approximately 6-mmHg elevation in end-tidal carbon dioxide ($P_{ET}CO_2$) during spaceflight compared with on Earth.[10] Microgravity also leads to a cephalad fluid shift, inducing a drastic change in blood distribution in which blood tends to move toward the head.[9] The impairment of cerebral autoregulation and CO_2 reactivity in astronauts after long-duration spaceflight under such conditions was reported.[35] Therefore, the effects of the combination of mild hypercapnia and a cephalad fluid shift on cerebral autoregulation should be investigated by the ground-based approach. On the other hand, the combination is also recognized in certain perioperative

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situations such as laparoscopic surgeries on the pelvic organs requiring intraperitoneal CO_2 insufflation and a head-down tilt (HDT) position.[29] Because the combined effect of mild hypercapnia and a cephalad fluid shift on cerebral autoregulation remains unknown, this type of investigation would also be important for laparoscopic surgery.[17]

Cerebral autoregulation is an important physiological mechanism that enables cerebral blood flow (CBF) to be maintained at an appropriate level to supply oxygen despite changes in arterial blood pressure (ABP). Importantly, dynamic cerebral autoregulation is capable of responding to rapid changes in CBF secondary to rapid changes in ABP.[24] To evaluate dynamic cerebral autoregulation, transfer function analysis has become established as a frequently used method that considers spontaneous fluctuations in ABP and CBF velocity, reflecting the linear relationship between these variables.[4]

The acute effects of hypercapnia on dynamic cerebral autoregulation have been investigated, and moderate-to-severe hypercapnia reportedly impairs dynamic cerebral autoregulation.[25,34] In the case of mild hypercapnia (increases of $P_{ET}CO_2 \approx$ 6 mmHg), however, 10 min of exposure to 3% CO_2 did not impair dynamic cerebral autoregulation.[12] Meanwhile, the effects of an acute cephalad fluid shift on dynamic cerebral autoregulation have been investigated by Cooke et al. These authors concluded that a 10-min exposure to −10° HDT did not affect dynamic cerebral autoregulation.[5]

To date, however, no reports have investigated the effects of the combination of mild hypercapnia and a cephalad fluid shift on dynamic cerebral autoregulation. We tested the hypothesis that short-term exposure to the combination of these factors would attenuate dynamic cerebral autoregulation. Moreover, we compared the combined effect to the individual effects separately.

METHODS

Subjects

Participating in this study were 15 healthy male volunteers (ages 24 ± 3 yr; height 170 ± 5 cm; weight 69 ± 9 kg; mean ± SD). We carefully screened subjects based on their medical history and physical examination. None of the subjects had any known medical problems. Subjects provided written, informed consent. The Ethics Committee of Nihon University School of Medicine approved the entire study protocol (No. 25-8-1), which was registered in the trial registry of the Japan University Hospital Medical Information Network (ID: UMIN000017157).

Instrumentation

Investigations were performed at least 2 h after a meal in a quiet and environmentally controlled laboratory with an ambient temperature of 23–26°C. Subjects refrained from heavy exercise, caffeine, alcohol, and carbonated beverages 12 h prior to the experiment. Subjects had a facemask firmly attached around their mouths and noses to prevent air leaks and lay in a supine position on a tilting bed, initially placed in the horizontal position. Heart rate (HR) was determined using three-lead electrocardiography and oxyhemoglobin saturation (S_pO_2) was measured using pulse oximetry (Life Scope BSM-5132; Nihon Kohden, Tokyo, Japan). Inspiratory gas was sampled from the facemask to confirm the fraction of inspiratory CO_2 using the gas analyzer of the Life Scope. $P_{ET}CO_2$ was analyzed based on expiratory flow using a CO_2 monitor (OLG-2800; Nihon Kohden, Tokyo, Japan). Continuous ABP waveforms in the radial artery were obtained using a tonometry sensor placed on the wrist via a noninvasive ABP monitor (JENTOW 7700; Colin, Aichi, Japan). The absolute value of ABP was calibrated at the level of the heart via intermittent ABP measured using the oscillometric method with a sphygmomanometer cuff placed over the brachial artery at the beginning of each data-collection period. Continuous CBF velocity waveforms in the middle cerebral artery (MCA) were obtained using a 2-MHz probe placed over the right temporal region via transcranial Doppler (TCD) ultrasonography (WAKI; Atys Medical, St. Genislaval, France). The probe was fixed individually at the same position and at a constant angle using a customized molded probe holder made from polymer material to fit each subject's facial bone and ear structures by the same experienced physician. The same depth, power, and sample volume for each individual were used throughout the four protocols. The high consistency of this measuring method has been established.[3,23]

Protocols

The present study included four protocols to cover all possible combinations of 10-min exposures to a specific inhalational gas (either room air or a 3% CO_2 mixture consisting of 3% CO_2, 21% O_2, and 76% N_2) and position (either 0° supine or −10° HDT). The protocols were designed as randomized, crossover, and single-blinded with air regarded as the placebo. The different protocols were labeled Placebo/Supine (exposure to air in the supine position), CO_2/Supine (exposure to 3% CO_2 in the supine position), Placebo/HDT (exposure to air in a −10° HDT position), and CO_2/HDT (exposure to 3% CO_2 in a −10° HDT position). Prior to every protocol, each subject was informed of the relevant position to be employed ("supine" or "−10° HDT"). Subjects were randomly exposed to one of the four protocols at a similar time of day, with an interval of at least 24 h between each protocol to prevent physiological interference. Two 200-L Douglas bags were prepared. The first was filled with air for the purposes of equilibration and the second was filled with either the placebo or 3% CO_2 for the relevant 10-min exposure. After a resting period of ≥15 min and an equilibration period of 10 min, the data were collected during the relevant 10-min exposure period. The difference in height between the tonometry cuff (at the heart level) and the TCD probe (at the level of the MCA) was measured to estimate the ABP at the level of the MCA when HDT was employed. Subjects were studied at rest, breathed of their own accord, and were asked to refrain from sleeping, moving, and speaking during each data-collection period.

Data Analysis

Dynamic cerebral autoregulation was evaluated via a frequency domain analysis and a transfer function analysis using the data collected during the last 6 min of each 10-min exposure period. The waveforms for continuous ABP, CBF velocity, and ECG were recorded at a sampling rate of 1 kHz using Notocord-Hem 3.3 software (Notocord, Paris, France). A fast Fourier transform and transfer function analysis with the beat-to-beat data for mean arterial pressure (MAP) and mean CBF velocity were performed using the analysis algorithm described by Nishimura et al.[23] with DADiSP software (DSP Development Corporation, Cambridge, MA). MAP spectral power and mean CBF velocity spectral power, as well as coherence, phase, and transfer function gain, were calculated in three frequency ranges: very low (0.02–0.07 Hz), low (0.07–0.20 Hz), and high (0.20–0.35 Hz). [23] Coherence expresses the linear relationship between MAP and mean CBF velocity, ranging from 0 to 1. Higher coherence (approaching 1) indicates that MAP and the mean CBF velocity closely covary, and this result is interpreted as CBF having a significantly linear relationship with ABP. Coherence is used to assess the reliability of phase and transfer function gain.[4] Phase is used to estimate the time shift between MAP and mean CBF velocity. It reflects the preceding performance of CBF change to ABP change. A phase value of zero indicates a lack of phase lead and complete synchrony between these two variables, implying the impaired CBF time-response characteristics to ABP oscillation. To avoid phase "wrap-around," negative values under the 0.10 Hz frequency range were removed from the averages.[4] Transfer function gain reflects the magnitude of transmission from MAP to mean CBF velocity, and expresses the amplitude ratio of signal transmission from ABP oscillation to CBF fluctuation. A transfer function gain of a high value implies that CBF fluctuation is amplified because of reductions in suppression capability against transmission from ABP oscillation. Transfer function gain is represented as the absolute value and the relative value normalized with MAP and mean CBF velocity (abbreviated as NGain) in accordance with the recommendation of the white paper.[4]

The MAP and mean CBF velocity values were averaged from the last 6 min of the data collected during each protocol. MAP at the level of the MCA (MAP-MCA) was calculated based on hydrostatic pressure, which was measured as the difference in height between the tonometry cuff and the MCA. Hydrostatic pressure was estimated as the difference in height multiplied by 0.76 mmHg.[32] The cerebral vascular resistance (CVR) index was expressed as MAP-MCA divided by the mean CBF velocity. S_pO_2, $P_{ET}CO_2$, and respiratory rate were recorded manually every minute and calculated by averaging the data of the last 6 min during each protocol.

Statistical Analysis

Data are presented as means and SDs. Data were compared across the four protocols to strengthen the repeated-measures experimental design using the same 15 subjects. For variables with a normal distribution (determined via the Kolmogorov-Smirnov test), one-way repeated-measures of analyses of variance (ANOVA) were performed. When the variables were not normally distributed, Friedman's repeated-measures ANOVA using ranks was performed. To determine where significant differences occurred, the Student-Newman-Keuls method was employed for multiple comparisons. Values of $P < 0.05$ were considered significant. All analyses were performed using SigmaStat version 3.11 (Systat Software, Chicago, IL).

RESULTS

The means and SDs of the protocol-averaged MAP variability and mean CBF velocity variability, as well as the protocol-averaged transfer function indices for each frequency range, are shown in **Table I**. The protocol-averaged spectra of the transfer function analysis in the entire frequency are shown in **Fig. 1** and the means of the transfer function analysis in the low-frequency range are shown in **Fig. 2**.

In the low-frequency range, the phase during the CO_2/HDT protocol was significantly lower than during the other three protocols [$\chi^2 = 8.360$, df = 3, $P = 0.039$ (Friedman), Fig. 2]. However, this index did not differ among the Placebo/Supine, CO_2/Supine, or Placebo/HDT protocols. The absolute value of the transfer function gain in the low-frequency range during the CO_2/HDT protocol was significantly higher than during the other three protocols [$\chi^2 = 9.320$, df = 3, $P = 0.025$ (Friedman), Fig. 2]. However, this index did not differ among the Placebo/Supine, CO_2/Supine, or Placebo/HDT protocols. The protocols did not significantly differ with regard to MAP variability, mean CBF velocity variability, coherence, or NGain in the low-frequency range.

The MAP variability in the very low frequency range during the Placebo/Supine protocol was significantly higher than that during the other three protocols [$F(3.14) = 4.986$, $P = 0.005$ (ANOVA)]. The absolute value of the transfer function gain in the high-frequency range during the Placebo/Supine protocol was significantly lower than the other three protocols [$\chi^2 = 9.564$, df = 3, $P = 0.023$ (Friedman)]. Other indices in the very low and high frequency ranges did not show significant differences across the protocols.

The steady-state cerebral circulation, hemodynamic, and respiratory data for each protocol are shown in **Table II**. MAP and MAP-MCA during the CO_2/HDT protocol were significantly higher than during the other three protocols [$\chi^2 = 8.360$, df = 3, $P = 0.039$ (Friedman), and $\chi^2 = 11.240$, df = 3, $P = 0.010$ (Friedman), respectively]. The mean CBF velocity during the CO_2/Supine and CO_2/HDT protocols were significantly higher than those during the Placebo/Supine and Placebo/HDT protocols, respectively [$F(3,14) = 9.646$, $P < 0.001$, (ANOVA)], and no significant difference was found between the CO_2/Supine and CO_2/HDT results. The CVR index during the CO_2/Supine and CO_2/HDT protocols was significantly lower than during the Placebo/Supine and Placebo/HDT protocols, respectively [$\chi^2 = 17.000$, df = 3, $P < 0.001$ (Friedman)], and no significant difference was found between the CO_2/Supine and CO_2/HDT results. $P_{ET}CO_2$ during the CO_2/Supine and

Table I. Protocol-Averaged MAP Variability and Mean CBF Velocity Variability (with SDs) and a Transfer Function Analysis for Each Frequency.

	PLACEBO/SUPINE	CO_2/SUPINE	PLACEBO/HDT	CO_2/HDT	*P*-VALUE
VLF					
MAP_{VLF} ($mmHg^2$)	7.24 ± 4.69	3.96 ± 3.12*	5.00 ± 2.95*	3.28 ± 2.39*	0.005 (A)
$MCBFv_{VLF}$ (cm^2/s^2)	4.91 ± 3.76	3.31 ± 1.92	3.95 ± 3.02	4.11 ± 3.90	0.422 (A)
$Coherence_{VLF}$ (unit)	0.56 ± 0.21	0.50 ± 0.14	0.55 ± 0.19	0.48 ± 0.19	0.313 (F)
$Phase_{VLF}$ (radians)	1.24 ± 0.38	1.17 ± 0.35	1.23 ± 0.57	1.23 ± 0.55	0.965 (A)
$Gain_{VLF}$ ($cm \cdot s^{-1} \cdot mmHg^{-1}$)	0.61 ± 0.29	0.69 ± 0.26	0.63 ± 0.24	0.79 ± 0.39	0.211 (A)
$NGain_{VLF}$ (%/%)	0.82 ± 0.34	0.77 ± 0.22	0.85 ± 0.29	0.95 ± 0.43	0.475 (A)
LF					
MAP_{LF} ($mmHg^2$)	2.35 ± 1.88	1.85 ± 1.95	1.84 ± 1.53	2.12 ± 2.75	0.379 (F)
$MCBFv_{LF}$ (cm^2/s^2)	1.72 ± 1.19	1.62 ± 0.94	1.76 ± 1.34	1.96 ± 1.68	0.724 (F)
$Coherence_{LF}$ (unit)	0.69 ± 0.12	0.66 ± 0.15	0.61 ± 0.17	0.63 ± 0.16	0.280 (A)
$Phase_{LF}$ (radians)	0.65 ± 0.16	0.52 ± 0.21	0.74 ± 0.33	0.49 ± 0.21†	0.039 (F)
$Gain_{LF}$ ($cm \cdot s^{-1} \cdot mmHg^{-1}$)	0.86 ± 0.28	0.94 ± 0.23	0.91 ± 0.30	1.08 ± 0.34†	0.025 (F)
$NGain_{LF}$ (%/%)	1.16 ± 0.28	1.08 ± 0.24	1.22 ± 0.35	1.33 ± 0.42	0.080 (A)
HF					
MAP_{HF} ($mmHg^2$)	0.24 ± 0.22	0.27 ± 0.22	0.18 ± 0.14	0.29 ± 0.29	0.517 (F)
$MCBFv_{HF}$ (cm^2/s^2)	0.26 ± 0.26	0.40 ± 0.29	0.27 ± 0.22	0.47 ± 0.39	0.092 (F)
$Coherence_{HF}$ (unit)	0.59 ± 0.13	0.63 ± 0.15	0.55 ± 0.16	0.62 ± 0.14	0.271 (A)
$Phase_{HF}$ (radians)	-0.02 ± 0.19	-0.07 ± 0.21	-0.02 ± 0.34	0.04 ± 0.21	0.650 (F)
$Gain_{HF}$ ($cm \cdot s^{-1} \cdot mmHg^{-1}$)	0.88 ± 0.32	1.06 ± 0.28*	0.99 ± 0.19*	1.16 ± 0.42*	0.023 (F)
$NGain_{HF}$ (%/%)	1.20 ± 0.33	1.23 ± 0.32	1.34 ± 0.26	1.39 ± 0.25	0.150 (A)

MAP, mean arterial pressure; MCBFv, mean cerebral blood flow velocity; Gain, transfer function gain; NGain, normalized transfer function gain; VLF, very low frequency (0.02–0.07 Hz); LF, low frequency (0.07–0.20 Hz); HF, high frequency (0.20–0.35 Hz).

Values are means and SDs.

P-values are expressed as (A) ANOVA or (F) Friedman.

† $P < 0.05$ compared with Placebo/Supine, CO_2/Supine, and Placebo/HDT (Student-Newman-Keuls method).

* $P < 0.05$ compared with Placebo/Supine alone (Student-Newman-Keuls method).

CO_2/HDT protocols was significantly higher than that during the Placebo/Supine and Placebo/HDT protocols, respectively [$F(3,14) = 69.235$, $P < 0.001$ (ANOVA)], and no significant difference was found between the CO_2/Supine and CO_2/HDT results. On the other hand, $P_{ET}CO_2$ during the Placebo/HDT protocol was significantly lower than that during the Placebo/Supine protocol [$F(3,14) = 69.235$, $P < 0.001$ (ANOVA)]. S_pO_2 during the CO_2/Supine and CO_2/HDT protocols was significantly higher than that during the Placebo/Supine and Placebo/HDT protocols, respectively [$\chi^2 = 27.942$, df = 3, $P < 0.001$ (Friedman)]. HR and respiratory rate did not vary significantly across the protocols.

DISCUSSION

The major finding of this study is that a 10-min exposure to 3% CO_2 in a −10° HDT position showed an increased transfer function gain as well as a significantly decreased phase in the low-frequency range compared with the other three protocols. The changes were 25% in phase and 26% in transfer function gain compared with a 10-min exposure to air in the supine position. Decreased phase indicates increased synchrony between ABP oscillation and CBF fluctuation, which reflects the impaired CBF time-response characteristics. Increased transfer function gain indicates an increased magnitude of transmission from ABP oscillation to CBF fluctuation. Thus, CBF fluctuation is amplified because of a reduced suppression capability against transmission from ABP oscillation. Together, these indices clearly indicate that dynamic cerebral autoregulation is attenuated in the specific frequency range of 0.07–0.20 Hz, but only when mild hypercapnia (increases of $P_{ET}CO_2 \approx 6$ mmHg) and cephalad fluid shifts are simultaneously present in the short term.

Dynamic cerebral autoregulation in hypercapnia was previously investigated using a transfer function analysis.[1,25,34] Decreased phase has been reported during moderate-to-severe hypercapnia due to the inhalations of 5% CO_2 and 8% CO_2.[1,25,34] However, a recent dose-dependent study by Jeong et al. showed that phase was unchanged during 10 min of 3% CO_2 inhalation (increases of $P_{ET}CO_2 \approx 6$ mmHg).[12] Our result that a 10-min exposure to 3% CO_2 in the supine position did not show statistically significant decreases in phase is consistent with Jeong's report. Because of this dose-dependent relationship, acute mild hypercapnia (3% CO_2) alone should not impair the time-response characteristics. Regarding transfer function gain, previous reports have noted the changes during moderate hypercapnia.[12,34] Some subjects have shown increased transfer function gain in a partial frequency range during 5% CO_2 inhalation[34] and a range of 0.02–0.07 Hz during 6% CO_2 inhalation has been reported.[12] This effect implies that a higher concentration of inhaled CO_2 ($\geq$ 5%) increases transfer function gain. However, in the present study, exposure to 3% CO_2 in the supine position did not change transfer function gain. Thus, acute mild hypercapnia (3% CO_2) alone would not affect dynamic cerebral autoregulation. On the other hand, one study by Cooke et al. has investigated dynamic cerebral autoregulation in an acute cephalad fluid shift.[5] That study used 10 min of −10° HDT and showed that dynamic cerebral autoregulation is preserved. In the present study, 10 min of air

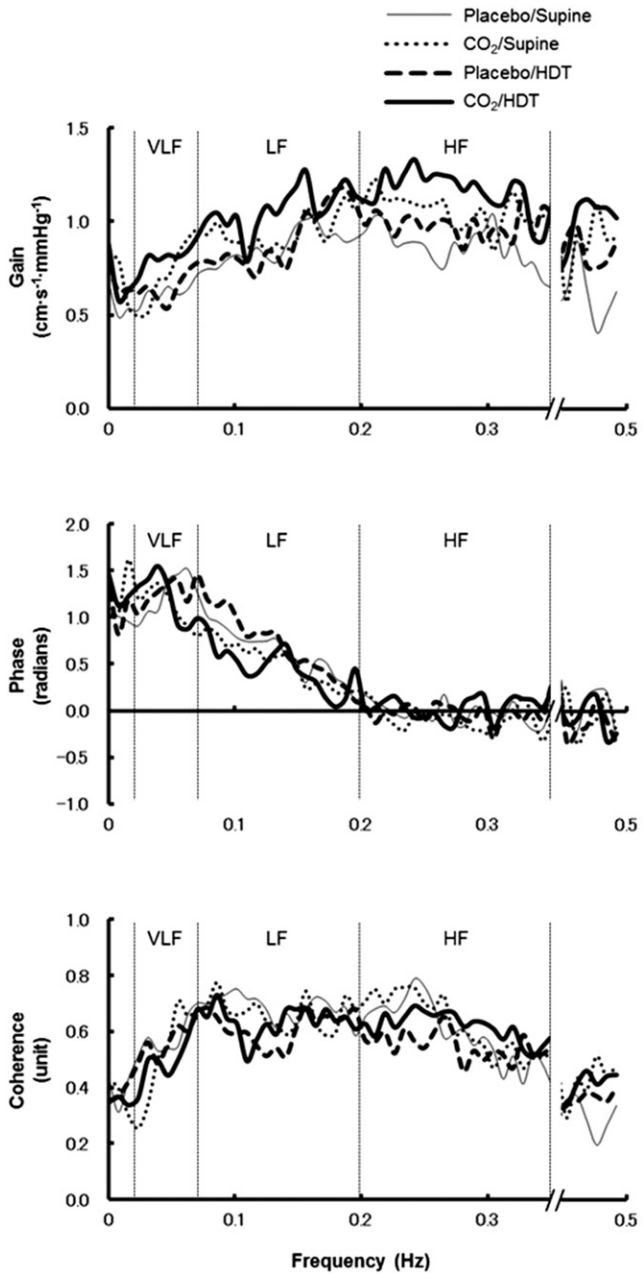

Fig. 1. Protocol-averaged transfer function analysis. Protocol-averaged transfer function analysis via a frequency domain between MAP and mean CBF velocity. Coherence: the coherence function between MAP and mean CBF velocity; Phase: the phase relationship between MAP and mean CBF velocity; Gain: the transfer function gain from MAP to mean CBF velocity; VLF: very low frequency (0.02–0.07 Hz); LF: low frequency (0.07–0.20 Hz); HF: high frequency (0.20–0.35 Hz). Solid line: Placebo/Supine; Dotted line: CO_2/Supine; Bold dotted line: Placebo/HDT; Bold line: CO_2/HDT.

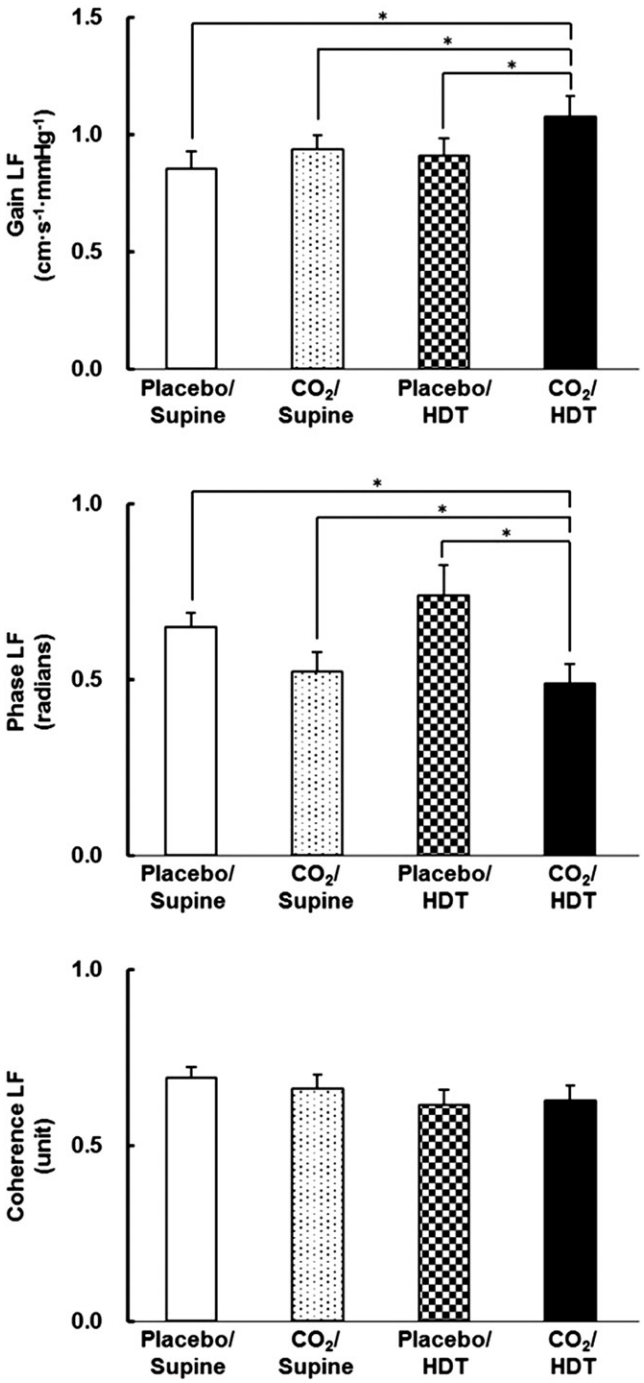

Fig. 2. Means and SDs of coherence, phase, and transfer function gain (Gain) performed via a transfer function analysis between MAP and mean CBF velocity in the low-frequency (0.07–0.20 Hz) range. White bar: Placebo/Supine; Dotted bar: CO_2/Supine; Checkered bar: Placebo/HDT; Black bar: CO_2/HDT. *$P < 0.05$ (Student-Newman-Keuls method).

inhalation in a −10° HDT position was not associated with a significant difference in transfer function indices. The present result and the previous Cooke's result suggest that dynamic cerebral autoregulation is preserved in an acute cephalad fluid shift alone. Our result showing a lower $P_{ET}CO_2$ value during the Placebo/HDT protocol relative to the Placebo/Supine protocol might also have contributed to preserve dynamic cerebral autoregulation.

Despite the present result that each single exposure did not change any of the transfer function indices over the very low and low-frequency ranges, the combination of mild hypercapnia and a cephalad fluid shift uniquely changed two of the transfer function indices in the low-frequency range. This combined effect might impair the time-response characteristics and reduce suppression capability via changes in the physiological

Table II. Steady-State Cerebral Circulation, Hemodynamic and Respiratory Data for Each Protocol.

	PLACEBO/SUPINE	CO_2/SUPINE	PLACEBO/HDT	CO_2/HDT	*P*-VALUE
MAP (mmHg)	79 ± 9	75 ± 12	77 ± 9	81 ± 6†	0.039 (F)
MAP-MCA (mmHg)	79 ± 9	75 ± 12	80 ± 9	84 ± 6†	0.010 (F)
MCBFv ($cm \cdot s^{-1}$)	57 ± 11	65 ± 12#	60 ± 12	69 ± 16#	< 0.001 (A)
CVR index ($mmHg \cdot cm^{-1} \cdot s^{-1}$)	1.4 ± 0.4	1.2 ± 0.3#	1.4 ± 0.3	1.3 ± 0.3#	< 0.001 (F)
HR (bpm)	59 ± 5	58 ± 7	59 ± 7	62 ± 6	0.285 (A)
Respiratory rate (breaths/min)	14 ± 3	14 ± 3	15 ± 4	15 ± 5	0.298 (F)
$P_{ET}CO_2$ (mmHg)	41 ± 3	46 ± 3#	38 ± 3*	47 ± 4#	< 0.001 (A)
S_pO_2 (%)	97 ± 1	99 ± 1#	98 ± 1	99 ± 1#	< 0.001 (F)

MAP, mean arterial pressure; MAP-MCA, MAP at the level of middle cerebral artery (MCA); MCBFv, mean cerebral blood flow velocity; CVR index, cerebral vascular resistance index; HR, heart rate; $P_{ET}CO_2$, end-tidal carbon dioxide; S_pO_2, oxyhemoglobin saturation by pulse oximetry.
Values are means and SDs.
P-values are expressed as (A) ANOVA or (F) Friedman.
† $P < 0.05$ compared with Placebo/Supine, CO_2/Supine, and Placebo/HDT (Student-Newman-Keuls method).
# $P < 0.05$ compared with Placebo/Supine and Placebo/HDT (Student-Newman-Keuls method).
* $P < 0.05$ compared with Placebo/Supine alone (Student-Newman-Keuls method).

mechanisms that modulate dynamic cerebral autoregulation. Cerebral autoregulation is likely modulated by several processes, including metabolic, myogenic, neural, and endothelial cell-related mechanisms.[26] Hypercapnia leads to the metabolically mediated vasodilatation[2] and the release of nitric oxide (NO), which is involved in the cerebral vascular response via endothelial cell-related mechanisms.[14] In addition, the contribution of NO to the myogenic mechanism might also occur.[8] Thus, hypercapnia might render cerebral arteriolar regulation unstable via these three mechanisms. On the other hand, the acute cephalad fluid shift produced by HDT induces a drastic change in the distribution of blood to the head. During HDT, the myogenic response to elevated ABP and the elevated circumferential stress to the endothelium destabilize cerebral arteriolar regulation.[33] The changes in the low-frequency range primarily reflect the physiological changes in the cerebral vascular response.[7] Although we did not attempt to detect the affected mechanism, the observed changes in this frequency range suggest that cerebral arteriolar regulation, modulated by metabolic, myogenic, and endothelial cell-related mechanisms, are attenuated under the combination of mild hypercapnia and a cephalad fluid shift. Intracranial pressure (ICP) should also be considered during simultaneous exposure to hypercapnia and a cephalad fluid shift. Increased cerebral blood volume due to hypercapnia-induced cerebral small vessel vasodilatation, and the cerebral venous or cerebrospinal fluid congestion occurring during a cephalad fluid shift[19] likely change ICP. The interaction between dynamic cerebral autoregulation and ICP is not yet well understood among healthy individuals,[20] including astronauts at risk of visual impairment during spaceflight.[18]

P_aCO_2 is a major modulator of steady-state CBF at rest.[24] In hypercapnia, the vasodilatation of small cerebral vessels results in increased CBF and decreased CVR.[2] No significant differences in CBF and CVR were found between the CO_2/Supine and CO_2/HDT protocols. Thus, a cephalad fluid shift has no additional effect on mild hypercapnia with regard to the steady-state cerebral circulation despite the attenuated dynamic cerebral autoregulation caused by the combined effect. On the other hand, CBF and CVR did not differ between Placebo/Supine and Placebo/HDT protocols, which is consistent with previous studies of short-term HDT.[5,30] The cerebral circulation in a steady state is preserved during a cephalad fluid shift alone.

Microgravity during spaceflight induces a cephalad fluid shift that drastically changes blood distribution toward the head.[9] We found that dynamic cerebral autoregulation was preserved during a cephalad fluid shift alone, but was attenuated during a combination of mild hypercapnia and a cephalad fluid shift. This result is a noteworthy finding for astronauts who experience cephalad fluid shifts during their stay on the ISS. The atmosphere inside the ISS contains higher concentrations of CO_2 than those under standard Earth conditions (approximately 0.5%, a more than tenfold increase).[15] Astronauts are also at risk of being temporarily exposed to much higher CO_2 concentrations due to rebreathing (up to 5% CO_2) and the localization of CO_2, induced by the absence of natural air convection in microgravity.[16] These factors increase the possibility that astronauts will experience mild transient hypercapnia during spaceflight. A recent study revealed that $P_{ET}CO_2$ during spaceflight in the ISS increased by approximately 6 mmHg compared with sitting on Earth.[10] Thus, investigation into the combined effect of 3% CO_2 and HDT on human physiology is required, and thus several experiments are also being conducted.[21] Positive relationships between CO_2 concentrations in the ISS and both the incidence of headache[15] and reduced cerebral CO_2 reactivity after long duration spaceflight have been reported.[35] These data suggest that exposure to CO_2 is an important factor that affects cerebral circulation during spaceflight. A previous report has shown that dynamic cerebral autoregulation is enhanced during Space Shuttle flight,[11] where the concentration of CO_2 is well-controlled below 0.1%.[28] Thus, a cephalad fluid shift on its own, without hypercapnia, would not impair dynamic cerebral autoregulation in space; this supposition is consistent with our results. Thus, the combination of mild hypercapnia (because of exposure to high concentrations of CO_2) and a cephalad fluid shift might impair cerebral autoregulation during spaceflight. This hypothesis suggests that a strict regulation of ambient CO_2 concentrations and avoiding CO_2 localization to prevent hypercapnia might improve astronauts' health.

In the past decade, laparoscopic and robotic surgical techniques employing intraperitoneal CO_2 insufflation have been widely adopted. The combination of hypercapnia and cephalad fluid shift occurs while using these techniques because they frequently employ −10° HDT or steeper position to better visualize the pelvic organs.[29] Recently, certain articles have warned that postoperative visual loss can be a complication of such surgeries.[27] Although permissive mild hypercapnia is frequently employed during laparoscopic surgery, a paucity of studies has investigated the combined effects of mild hypercapnia and a cephalad fluid shift on cerebral circulation.[17] Our study suggested that precise maintenance of normocapnia might be the available countermeasure against the attenuated dynamic cerebral autoregulation in the HDT position.

Changes in MCA diameter represent a common limitation in TCD studies. We estimated the change in CBF based on CBF velocity,[13] assuming that MCA diameter is a constant. A previous report by Serrador et al. showed that MCA diameter remains unchanged under both hypocapnia and hypercapnia conditions.[31] The possibility of underestimating increases in CBF due to the dilatation of the MCA in participants who were exposed to CO_2 concentrations above 6% was recently reported.[6] Although the present study did not measure the actual MCA diameter, this value would not change much in cases of mild hypercapnia (3% CO_2). Even if the present study underestimated the increases in CBF due to MCA dilatation, the actual fluctuation of CBF velocity might exceed the value during our CO_2 protocols. In either case, our findings are consistent because of the significant differences between the CO_2/HDT protocol and the other protocols recognized with regard to both the phase and transfer function gain. It is highly unlikely that this limitation affected the phase results because the phase reflects time-response characteristics. In addition, significant differences in phase and transfer function gain were recognized only in the low-frequency range. Therefore, the changes in these transfer function indices should not be induced only by a change in MCA diameter. Unfortunately, no gold standard test for the assessment of dynamic cerebral autoregulation exists. Although transfer function analysis is the most widely used for assessment of dynamic linear pressure-flow relationships, we should take into account that cerebral autoregulation also includes a nonlinear phenomenon.[22] NGain in the low-frequency range was not significant ($P = 0.080$, ANOVA). Normalizing transfer function gain might require more samples to reach sufficient statistical power. Another limitation might be that the present study only investigated a single-dose combination and short-term effects. Therefore, investigation of dose-dependent and long-term effects should be undertaken in the future.

The present study evaluated the effects of the combination of mild hypercapnia and a cephalad fluid shift on dynamic cerebral autoregulation using a 10-min exposure to 3% inhaled CO_2 to induce mild hypercapnia and −10° HDT to induce a cephalad fluid shift. We found that even short-term exposure to mild hypercapnia combined with a cephalad fluid shift induced both a decreased phase and increased transfer function gain in the low-frequency range, which indicates that this combination attenuates dynamic cerebral autoregulation. Therefore, the adverse effects of the combination of these two factors on dynamic cerebral autoregulation should be taken into account in laparoscopic surgery, as well as when considering astronauts' health during spaceflight.

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The authors have no conflicts of interest to declare.

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