### Abstract

**Objective:** The intervention of yoga has been shown to improve autonomic conditioning in humans and better adaptability to orthostatic challenges. Similarly, slow breathing at 0.1 Hz akin to pranayama also increases baroreflex sensitivity (BRS). Hence, we intended to investigate whether yoga practitioners have different autonomic responses at rest, during slow deep breathing as well as during 6° head down tilt (HDT) compared to naive group individuals. Aim: The aim of the study was to evaluate the acute effects of slow breathing on cardiovascular variability during HDT in yoga practitioners compared to yoga-naïve individuals. Settings and Design: This was a comparative study with repeated measures design conducted in Autonomic Function Test lab of the Department of Physiology, All India Institute of Medical Sciences, New Delhi, India. Materials and Methods: Time domain and frequency domain parameters of heart rate variability, blood pressure variability (BPV), and BRS were evaluated during 6° HDT and slow breathing at 0.1 Hz on forty yoga-naïve individuals and forty yoga practitioners with an average age of  $31.08 \pm 7.31$  years and  $29.93 \pm 7.57$  years, respectively. All of the participants were healthy. Statistical Analysis Used: General Linear Mixed Model ANOVA was applied with yoga experience as a between-group factor in repeated measures. Independent sample t-test was applied for between group comparison of respiratory rate, demographic, and anthropometric data. P < 0.05 is considered statistically significant. **Results:** Between-group comparison during HDT with spontaneous breathing has shown a significantly lower heart rate (P = 0.004) with higher RR interval (RRI) (P = 0.002) and pNN50% (P = 0.019) in yoga practitioners. The sequence BRS (P < 0.0001) and  $\alpha$  low frequency (LF) of spectral BRS (P = 0.035) were also significantly higher in the yoga group compared to the naïve group. Similarly, during HDT with slow breathing, the heart rate was lower (P = 0.01); with higher RRI (P = 0.009); pNN50% (P = 0.048). Standard deviation of successive RR interval difference of systolic BPV was lower (P = 0.024) with higher sequence BRS (P = 0.001) and  $\alpha$  LF of spectral BRS (P = 0.002) in yoga group than naïve group. Conclusion: The yoga experienced individuals exhibit higher resting parasympathetic activity, lower systolic BPV, and higher BRS than naïve to yoga individuals. It is inferred from the findings that yoga practitioners were better adapted to transient cephalad fluid shift that happens during 6° HDT. Furthermore, acute slow breathing during 6° HDT reduced the systolic blood pressure in all the participants suggesting the beneficial role of slow breathing during exposure to extreme conditions such as microgravity which might help in the prevention of adverse effects of cephalad fluid shift during long-term weightlessness and maintain the astronaut health. Future mechanistic studies with active yoga intervention are necessary to understand the adaptive mechanisms involving central and vascular modulations contributing to either attenuation or accentuation of the cardiovagal baroreflex during HDT and slow breathing in healthy individuals.

**Keywords:** Autonomic regulation, baroreflex sensitivity, blood pressure variability, cardiovascular, head down tilt, heart rate variability, yoga, microgravity

## Introduction

The baroreflex is a primary mechanism for the regulation of blood pressure. It gets activated due to the changes in the blood pressure through the baroreceptors located in the carotid sinus and aortic arch. Baroreflex sensitivity (BRS) was shown to

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be increased with slow breathing.<sup>[1,2]</sup> A slow and deep breathing with eyes closed was used as one of the strategies to remarkably reduce heart rate voluntarily.<sup>[3]</sup> Slow and deep breathing at 0.1 Hz is known to increase the respiratory sinus arrhythmia which is a marker for parasympathetic

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activity,<sup>[4]</sup> with synchronous increase in cardiovascular oscillations. Similarly, the intervention of yoga was also shown to increase the BRS.<sup>[5]</sup>

Yoga training provides a relatively better adaptability to orthostatic challenge as evident from the higher delta change (decrease) in parasympathetic activity and BRS with maintenance of systolic blood pressure (SBP).<sup>[6,7]</sup> Head down tilt (HDT) of 6° is one of the well-established analog and ground-based model for simulating the microgravity of space. Recent studies were found to be focused on head down bed rest of different durations for exploring the head-ward fluid shifts and the cardiovascular adaptations akin to those found in spaceflight and microgravity<sup>[8]</sup>, despite the fact that the tilt angle is 6°, comparing the effects of head down bed rest with immediate effect of HDT is unjustifiable. Moreover, transient HDT is a part of normal daily activities. HDT at various degrees is also practiced in Yoga that involves the practice of physical postures called asanas which are performed with precise coordination of the breath and Pranavama, a vogic breathing technique that involves predominantly slow deep breath inspired through the use of the abdominal muscles and the diaphragm. From the literature search, the cardiovascular variability during 5 min of 6° HDT, during simultaneous slow breathing, and 6° HDT in healthy subjects remains to be explored.

Therefore, the present study aimed to explore the heart rate variability (HRV), blood pressure variability (BPV), and BRS during transient 6° head-down tilt and slow breathing in healthy individuals with and without yoga experience. Considering the experience similar to both HDT and slow breathing in yoga practitioners, the present study aimed to investigate whether yoga experienced individuals exhibit differences in cardiovascular variability during spontaneous breathing and slow breathing during 6° HDT compared to yoga naïve individuals.

# **Materials and Methods**

This was a comparative study with repeated measures design conducted in the Autonomic Function Test lab of the Department of Physiology, All India Institute of Medical Sciences, New Delhi, India. The institutional ethics committee approved the study (IESC/T-464/28.10.2015) and was registered in the clinical trial registry, India (CTRI/2020/06/025730). The protocol and the informed consent forms were approved by the institutional ethics committee. Accordingly, the study included only those participants who have given voluntary and informed consent. The results collected during 6° HDT were part of a larger research study where other experimental protocols, such as head-up tilt, lower body negative pressure and slow breathing were also investigated in both groups of participants. The sample size was calculated by considering the change in BRS (outcome variable) after the intervention of slow breathing as  $5 \pm 3$  ms/mmHg in healthy individuals<sup>[1]</sup> and the anticipated change in yoga

practitioners as  $3 \pm 1.5$  ms/mmHg from the baseline values of BRS<sup>[9]</sup> with the power at 90% and the level of significance of 5% with consideration of 20% loss to follow-up.<sup>[10]</sup>

We recruited forty subjects in each group and the total subjects were eighty. All participants were healthy and normotensive, with normal resting electrocardiogram (ECG). Subjects with cardiovascular or respiratory illness or any other systemic illness, smokers, alcoholics, trained athletes, and those with varicose veins were excluded from the study. In the present study, the participants in naïve group are healthy individuals who did not have prior experience of yoga and regular exercise schedule. The recruited yoga practitioners are healthy subjects trained in yoga institutes or under the supervision of a yoga instructor for at least 3 months, with regular practice of asana and pranayama for a minimum of 3 days a week (at least 20 min of practice per day). The ability and ease of performance of the asana and pranayama from Column 1 and Column II [Table 1] were practically ascertained for voga practitioners before data collection. The selected asana involves forward-bending, backward-bending, and head-down postures that require practice to perform with steadiness, concentration, breath coordination, and flexibility. Yoga practitioners practiced classical yoga that includes asana, pranayama, meditation, and krivas, reflecting the comprehensive effect of yoga. Therefore, integrated voga was practiced in the context of the present study.

An appeal to participate in the study was circulated. Initial eligibility screening was done over the telephone. We screened 65 healthy individuals for the naïve group and 53 healthy trained yoga practitioners. Forty yoga-naïve individuals and forty trained yoga practitioners who had satisfied the inclusion criteria and gave consent for participating were ultimately recruited into this study.

# **Experimental protocol**

# 6° head down tilt protocol

Subjects lie down with their heads on the footboard side of the table, which is tilted from a supine posture to an angle of  $6^{\circ}$  at a rate of 2.3°/s. For 5 minutes, the subject was kept in the 6° HDT posture.

yoga practitioners						
					Column I (Asanas) Column II (Pranayama)	
Halasana	Anuloma viloma (alternate nostril					
Sarvangasana	breathing) and Bastrika (bellows					
Ustrasana	breath)					
Chakrasana						
Sirsasana						
Padahastasana						
Paschimottanasana						

Table 1. Asanas and pranavama demonstration by the

### Slow breathing protocol

The subjects performed slow breathing with 5-s inhalation and 5-s exhalation at a frequency of 0.1 Hz in sync with the continuous repetitive instructions of slow breathing. The participants were briefly trained to inhale for 5 s and exhale for 5 s in synchrony with the instructions from the audiotape with simultaneous visualization of the color-filled image. During data recording participants followed audio instructions and breathed comfortably without rapid and forceful inhalation and exhalation. The instructions were uniform to all the participants and all the participants could perform slow breathing.

### Study design

All the recordings were performed in a controlled ambient temperature of  $24^{\circ}C \pm 1^{\circ}C$  in the Autonomic Function Testing lab in the Department of Physiology, All India Institute of Medical Sciences, New Delhi. All the subjects were abstained from caffeine-containing beverages for 24 h and had light breakfast before 3 h of the testing. Yoga practitioners avoided the practice of yoga on the day of data recording. After instrumentation, all the participants were made to relax in supine rest on the tilt table for 15 min.

After baseline recording, all the participants were tilted to  $6^{\circ}$  HDT for 5 min followed by a recovery period of 5 min. The procedure was repeated, while the participants were slow breathing at 6 cycles per minute during supine and  $6^{\circ}$  HDT [Figure 1]. Lead II ECG, beat to beat blood pressure, and respiratory movements were recorded simultaneously and continuously for the entire duration of the experiment. Data recording of each participant were completed in a single session.

### **Data collection**

Lead II ECG was recorded simultaneously along with the beat-to-beat blood pressure (Model ML 283, AD Instruments). Respiratory movements were recorded using a respiratory belt transducer (Model MLT-1132, ADInstruments) connected to the analog to digital converter (Model 15T. ADInstruments). End-tidal carbon dioxide is a noninvasive measurement of the partial pressure of carbon dioxide (mmHg) during expiration, measured using CapnoTrue (Bluepoint Technologies, Germany). A nasal cannula was placed in the nostrils and connected to the Capnotrue. Lead II ECG was recorded at a sampling frequency of 1 kHz. The digital bandpass filter had a low cutoff frequency of 0.5 Hz and a high cutoff frequency of 35 Hz. Blood pressure and respiratory movements were recorded at a sampling frequency of 200 Hz. All the signals were recorded in Lab chart 8 (AD Instruments) and saved for offline analysis of HRV and BPV in time domain and frequency domain methods, and the BRS was computed by sequence method and spectral method<sup>[11-13]</sup> using Nevrokard analysis/version 6.2.0 software (Nevrokard, Izola, Slovenia). The segments of ECG and blood pressure from the same time period were selected for the analysis. Data from two naïve to yoga participants could not be analyzed.

### Statistical analysis

The group data were averaged and expressed as mean  $\pm$  standard error. General Linear Mixed Model ANOVA was applied with yoga experience as a between-group factor in repeated measures. Independent sample *t*-test was applied for between-group comparison of respiratory rate, demographic, and anthropometric data. The *P* < 0.05 is considered statistically significant. Statistical analysis was performed using SPSS (IBM Corp. Version 20.0. Armonk, NY, USA: IBM Corp Released 2011).

## Results

The autonomic modulation of HRV, BPV, and BRS was evaluated from the oscillations of heart rate and blood pressure during 6° HDT in yoga practitioners (yoga group) and in naïve to yoga individuals (naïve group).

The study participants were age matched. Both male and females were enrolled for the study. Body mass index and respiratory rate were comparable between groups. The average yoga experience was  $2.31 \pm 1.18$  years in yoga practitioners [Table 2].

### Heart rate variability between study groups

At supine rest, the yoga group has exhibited lower heart rate, higher RR interval (RRI), and PNN50 than the naïve group. Similarly, during HDT with spontaneous breathing, slow breathing in supine, and slow breathing in HDT, the heart rate was significantly lower, while RRI and pNN50% were significantly higher in yoga group. The respiratory rate was comparable between groups during supine rest and HDT. End-tidal carbon dioxide was significantly higher in yoga group than naïve group during slow breathing in supine, whereas end-tidal carbon dioxide was observed to be comparable during supine rest and HDT at both breathing frequencies [Tables 2 and 3].

# Blood pressure variability and baroreflex sensitivity between study groups

In supine rest, the yoga group was shown to exhibit a significantly lower standard deviations of normal to

Table 2: Demographic and anthropometriccharacteristics of naïve group and yoga group					
Variables	Total subjects				
	Yoga experienced group ( <i>n</i> =40)	Yoga naïve group ( <i>n=</i> 40)			
Age (years)	31.08±7.31	29.93±7.57			
Gender (male/female)	27/13	25/15			
Weight (kg)	63.15±7.01	60.50±7.04			
BMI (kg/m <sup>2</sup> )	23.43±1.79	22.85±1.40			
Respiratory rate (bpm)	15.89±3.39	16.17±7.01			

Data expressed as mean±SD. SD: Standard deviation

	Table 3: Co	o <mark>mparison of</mark> h	eart rate variability	between study groups		
Variable	Posture	Breathing	Yoga naïve group	Yoga experienced group	F	Р
Heart rate (bpm)	Supine	Spont br	69.350±1.35	63.218±1.31	10.557	0.002*
		Slow br	70.035±1.32	65.443±1.29	6.162	0.015*
	6° HDT	Spont br	69.044±1.4	63.052±1.41	8.758	0.004*
		Slow br	69.424±1.33	64.472±1.3	7.068	0.010*
Respiratory rate (br/min)#	Supine	Spont br	16.12±0.50	$14.74 \pm 0.41$	-0.269	0.789
	6° HDT	Spont br	15.15±0.43	14.11±0.48	0.797	0.428
Etco <sub>2</sub>	Supine	Spont br	35.553±0.66	36.246±0.67	0.531	0.469
-		Slow br	34.582±0.93	38.105±0.94	6.997	0.010*
	6° HDT	Spont br	35.326±0.68	36.784±0.69	2.233	0.140
		Slow br	34.785±0.88	35.098±0.90	0.061	0.805
RRI (ms)	Supine	Spont br	894.166±18.86	966.824±18.39	7.604	0.007*
		Slow br	884.202±17.52	944.402±17.07	6.054	0.016*
	6° HDT	Spont br	891.977±20.03	980.459±19.52	10.001	0.002*
		Slow br	887.664±18.44	956.417±17.97	7.129	0.009*
PNN50 (%)	Supine	Spont br	11.11±1.38	16.03±35	6.484	0.013*
		Slow br	12.342±1.19	16.99±1.16	7.811	0.007*
	6° HDT	Spont br	12.14±1.46	17.04±1.42	5.771	0.019*
		Slow br	12.43±1.12	15.59±1.09	4.027	0.048*
SDNN (ms)	Supine	Spont br	54.980±4.29	68.629±4.18	5.182	0.026*
		Slow br	84.305±5.494	96.848±5.35	2.673	0.106
	6° HDT	Spont br	61.788±4.63	73.544±4.517	3.301	0.073
		Slow br	82.804±5.343	93.046±5.20	1.884	0.174
LF (nu)	Supine	Spont br	48.503±3.422	46.491±3.33	0.177	0.675
		Slow br	84.368±2.72	81.065±2.65	0.754	0.388
	6° HDT	Spont br	45.740±3.40	47.353±3.32	0.115	0.735
		Slow br	80.889±2.95	78.072±2.87	0.466	0.497
HF (nu)	Supine	Spont br	46.566±3.17	48.452±3.08	0.182	0.671
		Slow br	13.614±1.94	14.844±1.89	0.206	0.651
	6° HDT	Spont br	49.400±3.16	47.049±3.08	0.284	0.596
		Slow br	16.782±2.19	17.748±2.14	0.099	0.754
LF/HF	Supine	Spont br	1.948±0.45	$1.487{\pm}0.44$	0.536	0.466
	-	Slow br	10.472±1.50	10.185±1.46	0.019	0.892
	6° HDT	Spont br	1.482±0.33	1.630±0.32	0.100	0.753
		Slow br	7.572±1.25	9.25±1.22	0.919	0.341

\*P<0.05 considered statistically significant compared to naïve group, <sup>#</sup>Independent sample *t*-test was applied. Values expressed as mean±SE. SDNN=Standard deviations of normal to normal R-R intervals, RMSSD=The square root of the mean of the sum of the squares of differences between adjacent NN intervals, pNN50=NN50 count divided by the total number of all NN intervals, LF=Low frequency, HF=High frequency, Etco<sub>2</sub>=End tidal carbon dioxide=nu normalized units, spont=Spontaneous, br=Breathing, HDT=Head down tilt, SE=Standard error, RRI=R-R interval

normal R-R interval (SDNN), standard deviation of successive RR interval difference (SDSD) of systolic BPV, and a significantly higher sequence BRS and  $\alpha$  low frequency (LF) and  $\alpha$  high frequency (HF) of spectral BRS than the naïve group. During HDT with spontaneous breathing, the SDSD and HF power of BPV were significantly lower, while sequence BRS and  $\alpha$ LF and  $\alpha$  HF of spectral BRS were significantly higher in yoga group. During slow breathing in supine, the sequence BRS,  $\alpha$  LF, and  $\alpha$  HF of spectral BRS were significantly higher in yoga group with a comparable BPV between groups. During slow breathing in HDT, the SDNN and SDSD of systolic BPV were significantly lower and sequence BRS and  $\alpha$  LF of spectral BRS were significantly higher in yoga group with a comparable BPV between groups. During slow breathing in HDT, the SDNN and SDSD of systolic BPV were significantly lower and sequence BRS and  $\alpha$  LF of spectral BRS were significantly higher in yoga group [Table 4].

Within group comparison of delta change in the indices of heart rate variability, blood pressure variability, and baroreflex sensitivity during 6° head down tilt while subjects performed spontaneous breathing or slow breathing

The  $6^{\circ}$  HDT compared to supine rest, a significant increase in SDNN of HRV with a significant decrease in LF power of systolic BPV was observed in naïve group.

Table 4: Comparison of systolic blood pressure variability and baroreflex sensitivity between study groups						
Variable	Posture	Breathing	Yoga naïve group	Yoga experienced group	F	Р
SBP (mm Hg)	Supine	Spont br	118.396±2.16	115.755±2.10	0.764	0.385
		Slow br	119.594±2.09	114.131±2.04	3.482	0.066
	6° HDT	Spont br	119.703±2.12	$115.880 \pm 2.07$	1.658	0.202
		Slow br	116.649±2.32	113.278±2.26	1.078	0.302
MBP (mm Hg)	Supine	Spont br	80.074±1.60	77.730±1.56	1.090	0.300
		Slow br	77.873±1.64	75.491±1.59	1.081	0.302
	6° HDT	Spont br	79.599±1.69	78.095±1.64	0.405	0.526
		Slow br	77.259±1.75	75.771±1.71	0.368	0.546
DBP (mm Hg)	Supine	Spont br	62.339±1.45	61.081±1.41	0.386	0.536
		Slow br	61.388±1.47	58.550±1.43	1.903	0.172
	6° HDT	Spont br	62.752±1.54	60.741±1.50	0.872	0.353
		Slow br	60.214±1.51	58.894±1.47	0.390	0.534
SDNN (mm Hg)	Supine	Spont br	6.232±0.29	5.235±0.29	5.684	0.020*
		Slow br	6.011±0.279	5.375±0.272	2.666	0.107
	6° HDT	Spont br	6.194±0.31	5.411±0.305	3.212	0.077
		Slow br	6.667±0.44	5.155±0.429	6.058	0.016*
SDSD (mm Hg)	Supine	Spont br	2.569±0.09	$2.203 \pm 0.08$	8.264	0.005*
		Slow br	2.736±0.162	2.383±0.158	2.449	0.122
	6° HDT	Spont br	2.818±0.148	2.383±0.14	4.453	0.038*
		Slow br	2.771±0.13	2.352±0.127	5.292	0.024*
SBP-LF (nu)	Supine	Spont br	70.284±2.29	73.046±2.24	0.741	0.392
		Slow br	87.819±1.57	88.739±1.53	0.174	0.677
	6° HDT	Spont br	65.871±2.57	72.367±2.51	3.258	0.075
		Slow br	85.158±1.77	87.301±1.72	0.752	0.388
SBP-HF (nu)	Supine	Spont br	25.167±2.34	22.427±2.28	0.700	0.406
	-	Slow br	9.491±1.44	9.030±1.40	0.052	0.820
	6° HDT	Spont br	28.389±2.53	21.313±2.46	4.012	0.049*
		Slow br	11.891±1.65	10.078±1.61	0.613	0.436
BRS (ms/mm Hg)	Supine	Spont br	16.252±1.60	25.888±1.54	18.700	0.000*
	*	Slow br	20.156±1.60	28.233±1.53	13.220	0.001*
	6° HDT	Spont br	18.238±1.86	28.526±1.79	15.845	0.000*
		Slow br	20.596±1.87	29.267±1.80	11.132	0.001*
α LF (ms/mm Hg)	Supine	Spont br	3.889±0.38	5.721±0.37	11.742	0.001*
( U)	1	Slow br	5.745±0.50	7.877±0.48	9.364	0.003*
	6° HDT	Spont br	4.389±0.44	5.712±0.42	4.605	0.035*
		Slow br	5.728±0.44	7.692±0.43	10.017	0.002*
α HF (ms/mm Hg)	Supine	Spont br	5.826±0.76	10.326±0.73	18.101	0.000*
(	τ	Slow br	7.349±1.07	$11.373 \pm 1.03$	7.330	0.008*
	6° HDT	Spont br	6.288±1.83	13.365±1.76	7.718	0.007*
		Slow br	8.082±1.29	11.550±1.24	3.741	0.057

\*P<0.05 considered statistically significant compared to naïve group. Values expressed as mean±SE. SBP=Systolic blood pressure, MBP=Mean blood pressure, DBP=Diastolic blood pressure, SDNN=Standard deviations of beat to beat systolic blood pressure, RMSSD=Root mean square of successive beat to beat systolic blood pressure differences, LF=Low frequency, HF=High frequency, BRS=Baroreflex sensitivity, spont=Spontaneous, br=Breathing, HDT=Head down tilt, SE=Standard error, SDSD=Standard deviation of successive RR interval differences

Similarly, a significant increase in RR interval and root mean square of successive differences between normal heartbeats (RMSSD) and  $\alpha$ -HF of spectral BRS was observed in yoga group [Table 5]. Slow breathing in supine resulted in a significant increase in SDNN, RMSSD, LF power, and LF/HF ratio as compared to supine rest. There was a significant decrease in HF power of HRV and mean blood pressure, as well as a significant rise in LF power of systolic BPV. The

number of sequences, sequence BRS, and LF of spectral BRS increased significantly within both groups [Table 5].

The slow breathing during 6°HDT compared to slow breathing in supine, a significant increase in the HF power, significant decrease in LF/HF of HRV, systolic and diastolic blood pressure, LF and HF power of systolic BPV was observed in naïve group. In yoga group, a significant increase in RRI and RMSSD with a decrease in pNN50 was

Variable Naïve group					Yoga experienced group			
	Spont br	Slow br	Slow br in	Slow br HDT	Spont br in	Slow br	Slow br in	Slow br HDT
	in HDT	in supine	HDT versus	versus spont	HDT versus	in supine	HDT versus	versus spont
	versus	versus spont	slow br in	br in HDT	spont br in	versus spont	slow br in	br in HDT
	spont br in supine	br in supine	supine		supine	br in supine	supine	
Heart rate	$-0.30\pm0.45$	0.68±0.59	-0.61±0.52	0.38±0.89	-0.16±0.44	2.22±0.58	$-0.97 \pm 0.51$	1.42±0.8
RRI (ms)	$-2.18\pm5.53$	$-9.96\pm8.29$	3.46±4.87	$-4.31\pm9.22$	13.63±5.39*	$-22.42\pm8.40$	12.01±4.75*	$-24.04 \pm 8.98*$
SDNN (ms)	$6.80 \pm 2.76*$	29.32±4.21*	$-1.50\pm2.34$	21.01±4.03*	4.91±2.69	28.21±3.76*	$-3.80{\pm}2.28$	19.50±3.93*
RMSSD (ms)	4.21±2.30	10.68±3.31*	$3.19 \pm 2.20$	$-4.31\pm9.22$	13.63±5.39*	13.43±3.71*	12.01±475*	$-24.04 \pm 8.98*$
pNN50 (%)	$1.02{\pm}0.74$	$1.23 \pm 1.03$	$0.09 \pm 0.43$	$0.29{\pm}1.09$	$1.01 \pm 0.72$	0.961±1.14	$-1.40{\pm}0.427*$	$-1.45 \pm 1.06$
LF <sub>RRI</sub> (nu)	$-2.76\pm2.30$	35.86±3.46*	$-3.47{\pm}1.84$	35.14±1.52*	$0.862 \pm 2.24$	34.57±3.37*	$-2.993{\pm}1.80$	30.71±3.43*
HF <sub>RRI</sub> (nu)	$2.83 \pm 2.20$	$-32.95 \pm 3.09*$	3.16±1.53*	$-32.61 \pm 3.17*$	$-1.40\pm2.14$	$-33.60{\pm}3.01*$	$2.90{\pm}1.49$	$-29.30 \pm 3.09*$
LF/HF	$-0.46 \pm 0.32$	8.52±1.51*	$-2.90{\pm}1.28*$	6.09±1.2*	$0.14 \pm 0.31$	8.96±1.47*	$-0.931 \pm 1.25$	7.62±1.19*
SBP (mm Hg)	$1.30{\pm}0.70$	1.19±1.17	$-2.94{\pm}0.94{*}$	$-3.05 \pm 1.38*$	$0.12 \pm 0.68$	$-1.62 \pm 1.14$	$-0.85 \pm 0.92$	$-2.60\pm1.34$
MBP (mm Hg)	$-0.47 \pm 0.97$	$-2.20\pm0.98*$	$-0.61\pm0.67$	$-0.36 \pm 0.94$	$0.36 \pm 0.94$	$-2.24 \pm 0.95*$	$0.28 \pm 0.65$	$-2.32\pm1.15*$
DBP (mm Hg)	$0.41 \pm 0.79$	$-0.95 \pm 0.93$	$-1.17 \pm 0.55*$	$-2.53 \pm 1.04*$	$-0.34 \pm 0.77$	$-2.53 \pm 0.90$	$0.34 \pm 0.54$	$-1.84{\pm}1.01$
SDSD (mm Hg)	$0.24{\pm}0.12*$	0.16±0.15	0.03±0.13	$-0.04 \pm 0.12$	0.17±0.12	$0.17 \pm 0.14$	$-0.03\pm0.12$	$-0.03\pm0.11$
LF <sub>SBP</sub> (nu)	$-4.41 \pm 1.59*$	17.53±2.44*	$-2.66 \pm 1.16*$	19.28±2.53*	-0.67±1.55	15.69±2.38*	$-1.43{\pm}1.13$	14.93±2.47*
HF <sub>SBP</sub> (nu)	$3.22 \pm 1.63$	-15.67±2.36*	$2.4 \pm 0.98*$	$-16.49 \pm 2.43*$	-1.11±1.59	$-13.39{\pm}2.30*$	$1.04 \pm 0.95$	-11.23±2.37*
Number of	$1.63 \pm 3.05$	21.13±2.82*	$-2.27\pm1.48$	17.22±3.06*	$-3.23\pm2.93$	15.71±2.71*	$-1.10\pm1.46$	17.84±2.94*
baroreflex								
sequences								
BRS (ms/mm Hg)	$1.98 \pm 1.65$	3.90±1.81*	0.43±1.11	$2.35 \pm 1.83$	2.63±1.58	2.34±1.74	$1.03 \pm 1.07$	0.74±1.76
α-LF (ms/mm Hg)	$0.50\pm0.35$	$1.85 \pm 0.51*$	$-0.01\pm0.32$	1.33±0.45*	$-0.009 \pm 0.34$	2.15±0.49*	$-0.18\pm0.30$	1.97±0.43*
α-HF (ms/mm Hg)	$0.46{\pm}1.44$	$1.52 \pm 0.85$	0.73±0.90	1.79±1.37	3.03±1.39*	1.04±0.82	0.17±0.86	-1.81±1.32

Fable 5: C	<b>Comparison of</b>	heart rate <b>v</b>	variability, bloo	d pressure	variability, and	barore	lex sensitivity	within study	Į
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\**P*<0.05 considered statistically significant compared to naïve group. Values expressed as mean difference±SE. SDNN=Standard deviations of normal to normal R-R intervals, RMSSD=The square root of the mean of the sum of the squares of differences between adjacent NN intervals, pNN50=NN50 count divided by the total number of all NN intervals, LF=Low frequency, HF=High frequency, BRS=Baroreflex sensitivity, Spont=Spontaneous, br=breathing, SE=Standard error, SDSD=Standard deviation of successive RR interval differences, SBP=Systolic blood pressure, MBP=Mean blood pressure, DBP=Diastolic blood pressure, RRI=R-R interval, HDT=Head down tilt

observed [Table 5]. Slow breathing during HDT compared to HDT with spontaneous breathing, a significant increase in SDNN, LF power, and LF/HF with significant decrease in HF power of HRV was observed within both the groups. A significant increase in the  $\alpha$ -LF of spectral baroreflex sensitivity with a significant increase in SBP oscillations at low frequency with concomitant decrease in high frequency was also observed in systolic blood pressure variability within both the groups. In addition, RRI and RMSSD were observed to be significantly reduced only in yoga group [Table 5].

## Discussion

The HRV, BPV, and BRS were evaluated in supine and during 6° head-down tilt while breathing spontaneously and also while performing slow breathing at 6 cycles/ min at 0.1 Hz in yoga practitioners compared to naïve group individuals. Yoga practitioners exhibited higher parasympathetic activity and BRS with lower systolic BPV than naïve during supine rest. During HDT with spontaneous breathing and slow breathing, the HRV was higher with the predominance of parasympathetic activity along with lower systolic BPV and higher BRS. During

slow breathing in supine, the parasympathetic activity of HRV and BRS was higher with comparable BPV.

# Within-group comparison of cardiovascular variability during spontaneous breathing and slow breathing in supine and during 6° head down tilt

During HDT compared to supine rest [Table 4], the heart rate, blood pressure, and arterial BRS did not change during the HDT within naïve individuals which are following an earlier study.<sup>[14]</sup> Whereas head stand exercise with and without assistance resulted in an increase in sympathetic activity of HRV.<sup>[15]</sup> The disparity in findings could be due to differences in the angle of the tilt, in addition to active involvement of musculoskeletal system while performing the head stand.<sup>[15]</sup> HDT was also observed to reduce the oscillations of SBP oscillations in LF significantly in naïve group which is in agreement with earlier findings.<sup>[16]</sup> Conversely, there was no significant change in BRS during HDT compared to supine. Our findings were in contrast with earlier findings where an increase in BRS was observed during HDT. This might also be due to differences in the degrees of tilt in the head-down tilt. The present study evaluated at 6°, while the earlier study has evaluated at 25° head-down tilt<sup>[16]</sup> and a significant increase in  $\alpha$ -HF of spectral BRS was observed in yoga group at 6° HDT similar to 25° HDT.<sup>[16]</sup> When both studies are viewed together, we are inclined to believe that the BRS can be augmented significantly in yoga group even with 6° HDT, while in naïve group who are healthy individuals without yoga practice may require 25° of HDT for 10 min to have a significant increase in BRS.

This differential response might be due to an anticipatory response in yoga group due to prior experience of yoga which may have helped in the accentuation of the baroreflex activity even with a minimum 6° HDT. Slow breathing in supine compared to spontaneous breathing has significantly increased the parasympathetic activity with comparable SBP within both groups during slow breathing in supine compared to supine rest [Table 4]. Although the mean of SBP was comparable within both groups, the oscillations of systolic and diastolic blood pressure, baroreflex sequences, and  $\alpha LF$  of spectral BRS were significantly increased within both groups. The parasympathetic activity has significantly increased with slow breathing in supine which is in confirmation with earlier findings.<sup>[1]</sup> In the present study, the frequency of slow breathing at six cycles per min was at 0.1 Hz, and accordingly, the respiratory peak was observed to be shifted to LF of the spectrum. The spectral analysis has shown a sharp decline in HF power and the rise of LF power, respectively, during slow breathing. By documenting the findings of the previous studies,<sup>[17,18]</sup> we confirm the enhancement of efferent vagal activity during slow breathing at 0.1 Hz in the supine position. Slow breathing was also shown to acutely increase the parasympathetic activity independent of yoga practice<sup>[1]</sup> and was also found to increase the BRS at both symmetrical breathing of 5 s of inspiration and expiration in yoga beginners.<sup>[19]</sup> In accordance with earlier studies, the present study has also shown that slow breathing can acutely reduce the mean blood pressure within both groups independent of yoga practice.<sup>[20]</sup> The oscillations of systolic and diastolic blood pressure at LF power and spectral BRS at  $\alpha LF$  were also significantly increased with slow breathing.[10,21] The cyclical changes in intrathoracic pressure due to slow breathing may have contributed to the oscillations in arterial blood pressure, heart rate, stroke volume, and cardiac output. These changes may have sensed by carotid sinus and aortic baroreceptors, thereby activating the BRS within the respiratory cycle.[22] The parasympathetic activity has significantly increased during HDT with slow breathing compared to HDT with spontaneous breathing within both groups [Table 4]. However, the systolic and diastolic blood pressure has significantly decreased within naïve group only. The plausible explanation could be that naïve group individuals are new to the breathing maneuver and when combined with HDT, might have generated a greater

reduction in intrathoracic pressure with slow breathing during HDT. This might have contributed to a significant reduction of SBP in naïve group. In contrast, comparable blood pressure in yoga group could be due to adaptation to the downward tilt and slow breathing during yoga practice that includes asanas and pranayama comprising hyperventilation, hypoventilation, and apnea though the angle of the tilt and duration may not be precise with  $6^{\circ}$ HDT. Nonetheless, the oscillations of systolic and diastolic blood pressure at LF with a corresponding increase in the baroreflex sequences and the  $\alpha$  HF of spectral BRS was observed within both groups [Figure 3]. It is interesting to note that the RRI was comparable during all the phases in the naïve group. The SBP oscillations at LF and HF have significantly increased and decreased, respectively, during slow breathing in supine (vs. spontaneous breathing in supine) and slow breathing during HDT (vs. HDT with spontaneous breathing) within both the groups. These findings suggest an increase in systolic BPV with slow breathing following earlier findings.<sup>[10]</sup> The present study has also shown a significant increase in BRS during slow breathing in supine and HDT compared to spontaneous breathing in all the participants signifying the role of slow breathing in enhancing the BRS at LF. While the rise in BRS was higher in yoga group than naive group.

## Between-group comparison of cardiovascular variability during spontaneous breathing and slow breathing in supine and during 6° head down tilt

In the yoga group, the parasympathetic nervous activity and BRS were significantly higher during supine rest, HDT, slow breathing in supine, and slow breathing during HDT. The systolic BPV was significantly lower in supine rest and slow breathing during HDT. Whereas, the BPV was observed to be comparable between groups during HDT and during slow breathing in supine. Regular increase of parasympathetic activity through slow vogic breathing, asanas and meditation may have contributed to the accentuation of the parasympathetic activity and BRS in the voga group compared to the naïve group. The regular practice of yoga especially asanas and pranayama might have increased the sensitivity for blood pressure changes as well.<sup>[23]</sup> There might also be an anticipatory increase in parasympathetic activity in the yoga practitioners, resulting in enhanced BRS which is inversely related to the BPV contributing to lower BPV in the voga group.<sup>[24]</sup> The parasympathetic activity was higher in the yoga group which was in confirmation with earlier studies that suggested that heart rate can be voluntarily reduced with yoga practice.<sup>[25]</sup> In yoga-experienced individuals, the RRI was observed to be significantly increased with HDT and the rise in RRI was independent of slow breathing [Figure 2a]. In contrast, the RRI was observed to be comparable in naïve group during all the phases of the experimental protocols. The SBP was observed to be comparable during HDT in all the participants.



Figure 1: Study design. HDT = Head down tilt, Br = Breathing, HRV = Heart rate variability, BPV = Blood pressure variability, BRS = Baroreflex sensitivity



Figure 2: RR interval (a) and systolic blood pressure (b) during supine, 6° HDT, Slow breathing in supine and slow breathing during 6° HDT. a = HDT (vs.) supine; b = HDT + slow breathing (vs.) supine slow breathing; c = HDT (vs.) HDT + slow breathing. \*P < 0.05 statistically significant difference between groups and <sup>†</sup>P < 0.05 statistically significant difference within yoga group; <sup>§</sup>P < 0.05 statistically significant difference within naive group. HDT = Head down tilt



Figure 3: Baroreflex sensitivity-sequence method (a) and  $\alpha$  LF of spectral BRS (b) during supine, 6° HDT, slow breathing in supine and slow breathing during 6° HDT. c = HDT (vs.) HDT + slow breathing; d = slow breathing vs. spontaneous breathing in supine. \**P* < 0.05 statistically significant difference between groups and †*P* < 0.05 statistically significant difference within yoga group; \**P* < 0.05 statistically significant difference within naive group. LF = Low frequency, BRS = Baroreflex sensitivity, HDT = Head down tilt

A significant fall in SBP was observed with slow breathing during HDT within naïve group but not within naïve group [Figure 2b]. A maintenance of relatively stable blood pressure in yoga group suggests an adaptive response of the HDT practiced during yoga. Moreover, the beat-to-beat oscillations of SBP were significantly lower in yoga group during HDT at both respiratory frequencies [Table 4].

Interestingly, acute slow breathing has reduced the SBP in all the participants, with a significant reduction within naïve group, suggesting the beneficial role of slow breathing during exposure to extreme conditions such as microgravity. From the findings, we are inclined to believe that slow breathing practice might help in the prevention of adverse effects of cephalad fluid shift during long-term weightlessness<sup>[26]</sup> and thereby help in the maintenance of the cardiovascular health<sup>[27]</sup> of astronauts.

The mechanism of yoga was postulated from the direct and indirect associations of the practice of yoga with the indices of cardiovascular variability. Flexibility is one of the important attributes acquired with regular practice of yoga.<sup>[28]</sup> The intervention of Yoga increases the parasympathetic activity,<sup>[29,30]</sup> reduces body mass index,<sup>[31]</sup> and increases the BRS.<sup>[5]</sup> The body mass index is inversely related to HRV<sup>[32]</sup> and BPV.<sup>[33]</sup> Similarly, BRS and BPV are also inversely related.<sup>[24]</sup> In addition, the central arterial stiffness was reported to be lower with yoga practice,<sup>[34]</sup> while a decrease in arterial stiffness was observed with regular static muscle stretching<sup>[35]</sup> which is a part of yoga for acquiring flexibility. However, it is pertinent to mention that there is no available study on yoga practitioners that have compared the effects of transient 6° HDT and slow breathing on cardiovascular variability. Thus, we could not draw a clear conclusion regarding the inherent mechanism for the higher BRS in rest, during HDT and slow breathing in voga experienced individuals. Thus, future mechanistic studies with active yoga intervention are necessary to understand the adaptive mechanisms involving central and vascular modulations contributing to either attenuation or accentuation of the cardiovagal baroreflex during HDT and slow breathing in healthy individuals.

#### Conclusion

The yoga experienced individuals exhibit higher resting parasympathetic activity, lower systolic BPV, and higher BRS than naïve to yoga individuals. It is inferred from the findings that yoga practitioners were better adapted to transient cephalad fluid shift that happens during 6° HDT. Furthermore, acute slow breathing during 6° HDT reduced the SBP in all the participants suggesting the beneficial role of slow breathing during exposure to extreme conditions such as microgravity which might help in the prevention of adverse effects of cephalad fluid shift during long-term weightlessness and maintain the astronaut health.

## Limitations

Our study has certain limitations. Yoga practice by the individuals in yoga group could not be monitored personally. However, the ability and the ease of performance of yogasanas and pranayama have been ascertained before calling for data collection. Although the participants in yoga group self-reported about regularity in yoga, there is also a lack of precise data on how often or frequently the trained yoga practitioners have practiced yoga due to recall bias. Moreover, other forms of activity, lifestyle in all the participants could not be controlled.

### Ethical clearance

The institutional ethics committee approved the study (IESC/T-464/28.10.2015).

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### **Conflicts of interest**

There are no conflicts of interest.

### References

- 1. Paprika D, Gingl Z, Rudas L, Zöllei E. Hemodynamic effects of slow breathing: Does the pattern matter beyond the rate? Acta Physiol Hung 2014;101:273-81.
- Radaelli A, Raco R, Perfetti P, Viola A, Azzellino A, Signorini MG, *et al.* Effects of slow, controlled breathing on baroreceptor control of heart rate and blood pressure in healthy men. J Hypertens 2004;22:1361-70.
- Raghavendra B, Telles S, Manjunath N, Deepak KK, Naveen K, Subramanya P. Voluntary heart rate reduction following yoga using different strategies. Int J Yoga 2013;6:26-30.
- Katona PG, Jih F. Respiratory sinus arrhythmia: Noninvasive measure of parasympathetic cardiac control. J Appl Physiol 1975;39:801-5.
- Bowman AJ, Clayton RH, Murray A, Reed JW, Subhan MM, Ford GA. Effects of aerobic exercise training and yoga on the baroreflex in healthy elderly persons. Eur J Clin Invest 1997;27:443-9.
- Anasuya B, Deepak KK, Jaryal AK. Autonomic tone and baroreflex sensitivity during 70° head-up tilt in yoga practitioners. Int J Yoga 2020;13:200-6.
- Anasuya B, Deepak KK, Jaryal A. Yoga practitioners exhibit higher parasympathetic activity and baroreflex sensitivity and better adaptability to 40 mm Hg lower-body negative pressure. Int J Yoga Therap 2021.doi: 10.17761/2021-D-20-00030. Epub ahead of print. PMID: 34280261.
- 8. Hargens AR, Vico L. Long-duration bed rest as an analog to microgravity. J Appl Physiol (1985) 2016;120:891-903.
- Branka N, Sanja P, Milovanović B. "Autonomous Control of Cardiovascular Function in Yoga Instructors and Effects of Energetic Renewal on Modulation of Autonomic Function." Int Sci Yoga J Sense 2011;1:99-109. Available from: https://yoga-sci. eu/wp-content/uploads/2014/06/journal-2011.pdf#page=99. [Last accessed on 2020 Jun 18].
- 10. Anasuya B, Deepak KK, Jaryal AK, Narang R. Effect of slow

breathing on autonomic tone and baroreflex sensitivity in yoga practitioners. Indian J Med Res 2020;152:638-47.

- Tamuli D, Kaur M, Boligarla A, Jaryal AK, Srivastava AK, Deepak KK. Depressed baroreflex sensitivity from spontaneous oscillations of heart rate and blood pressure in SCA1 and SCA2. Acta Neurol Scand 2019;140:350-8.
- Parati G, Di Rienzo M, Castiglioni P, Ulian L, Mancia G. "Cardiovascular regulation and analysis of blood pressure-heart rate variability interactions." Fundam Clin Pharmacol. 1999;13:11-5. doi: 10.1111/j.1472-8206.1999.tb00315.x. PMID: 10027083.
- Bertinieri G, di Rienzo M, Cavallazzi A, Ferrari AU, Pedotti A, Mancia G. "A new approach to analysis of the arterial baroreflex." J Hypertens Suppl 1985;3:S79-81.
- London GM, Levenson JA, Safar ME, Simon AC, Guerin AP, Payen D. Hemodynamic effects of head-down tilt in normal subjects and sustained hypertensive patients. Am J Physiol 1983;245:H194-202.
- Manjunath NK, Telles S. "Effects of Sirsasana (headstand) practice on autonomic and respiratory variables." Indian J Physiol Pharmacol 2003;47:34-42.
- Porta A, Marchi A, Bari V, Catai AM, Guzzetti S, Raimondi F, et al. Directionality in cardiovascular variability interactions during head-down tilt test. Annu Int Conf IEEE Eng Med Biol Soc 2014; p. 6008-11.
- 17. Chang Q, Liu R, Shen Z. Effects of slow breathing rate on blood pressure and heart rate variabilities. Int J Cardiol 2013;169:e6-8.
- Kromenacker BW, Sanova AA, Marcus FI, Allen JJ, Lane RD. Vagal mediation of low-frequency heart rate variability during slow yogic breathing. Psychosom Med 2018;80:581-7.
- Mason H, Vandoni M, Debarbieri G, Codrons E, Ugargol V, Bernardi L. Cardiovascular and respiratory effect of yogic slow breathing in the yoga beginner: What is the best approach? Evid Based Complement Alternat Med 2013;2013:743504.
- Adler TE, Coovadia Y, Cirone D, Khemakhem ML, Usselman CW. Device-guided slow breathing reduces blood pressure and sympathetic activity in young normotensive individuals of both sexes. J Appl Physiol (1985) 2019;127:1042-9.
- 21. Radaelli A, Raco R, Perfetti P, Viola A, Azzellino A, Signorini MG, *et al.* Effects of slow, controlled breathing on baroreceptor control of heart rate and blood pressure in healthy men. J Hypertens 2004;22:1361-70.
- 22. Blaber AP, Hughson RL. Cardiorespiratory interactions during fixed-pace resistive breathing. J Appl Physiol (1985) 1996;80:1618-26.
- Novak V, Novak P, de Champlain J, Le Blanc AR, Martin R, Nadeau R. Influence of respiration on heart rate and blood pressure fluctuations. J Appl Physiol (1985) 1993;74:617-26.
- 24. Laitinen T, Hartikainen J, Niskanen L, Geelen G, Länsimies E. Sympathovagal balance is major determinant of short-term blood pressure variability in healthy subjects. Am J Physiol 1999;276:H1245-52.
- 25. Telles S, Joshi M, Dash M, Raghuraj P, Naveen KV, Nagendra HR. An evaluation of the ability to voluntarily reduce the heart rate after a month of yoga practice. Integr Physiol Behav Sci 2004;39:119-25.
- 26. Norsk P. Adaptation of the cardiovascular system to weightlessness: Surprises, paradoxes and implications for deep space missions. Acta Physiol (Oxf) 2020;228:e13434.
- 27. Chaddha A. Slow breathing and cardiovascular disease. Int J Yoga 2015;8:142-3.
- 28. Ray US, Mukhopadhyaya S, Purkayastha SS, Asnani V, Tomer OS, Prashad R, et al. Effect of yogic exercises on

physical and mental health of young fellowship course trainees. Indian J Physiol Pharmacol 2001;45:37-53.

- Muralikrishnan K, Balakrishnan B, Balasubramanian K, Visnegarawla F. Measurement of the effect of Isha Yoga on cardiac autonomic nervous system using short-term heart rate variability. J Ayurveda Integr Med 2012;3:91-6.
- Tyagi A, Cohen M. Yoga and heart rate variability: A comprehensive review of the literature. Int J Yoga 2016;9:97-113.
- Chauhan A, Semwal DK, Mishra SP, Semwal RB. Yoga practice improves the body mass index and blood pressure: A randomized controlled trial. Int J Yoga 2017;10:103-6.
- 32. Koenig J, Jarczok MN, Warth M, Ellis RJ, Bach C,

Hillecke TK, *et al.* Body mass index is related to autonomic nervous system activity as measured by heart rate variability--a replication using short term measurements. J Nutr Health Aging 2014;18:300-2.

- 33. Chen H, Zhang R, Zheng Q, Yan X, Wu S, Chen Y. Impact of body mass index on long-term blood pressure variability: A cross-sectional study in a cohort of Chinese adults. BMC Public Health 2018;18:1193.
- Duren CM, Cress ME, McCully KK. The influence of physical activity and yoga on central arterial stiffness. Dyn Med 2008;7:2.
- Nishiwaki M, Yonemura H, Kurobe K, Matsumoto N. Four weeks of regular static stretching reduces arterial stiffness in middle-aged men. Springerplus 2015;4:555.