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Infradian cardiovascular rhythms and between subject cardiovascular coupling in Mars500 prolonged confinement study

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ABSTRACT

Introduction: The aim of this study was to characterize infradian rhythms in heart rate and blood pressure, as well as investigate potential synchronization of these parameters among participants in the Mars500 study. This study involved a 520-day simulated mission to Mars, during which six crew members were confined for the entire duration of the experiment.

Methods: The study involved daily measurements of blood pressure and heart rate, taken in the morning and evening for each participant. Mean differences were analyzed using a mixed model design, considering journey stage and time of day as fixed factors, and participant as a random factor. The Lomb-Scargle periodogram was used to identify rhythmic patterns in the data. Significant rhythms were described by fitting a cosine curve using a cosinor model. The level of synchronization between participants' measurements was assessed by analyzing signal pairs using the Hilbert transform. Phase differences were measured in one-month bins using the gamma parameter. Surrogate data was employed for comparison with the observed data. Gamma differences between bins were evaluated using a mixed model design.

Results: All signals showed significant changes over time, and most participants exhibited at least one significant infradian rhythm in the analyzed variables. The primary peaks varied between participants and variables, ranging from 22 days to 520 days. Interestingly, there was a consistent period of 173 days observed in the evening heart rate across all participants. Regarding synchronization, the gamma values of evening heart rate differed from surrogate data in all stages, except during the middle part of the mission.

Discussion: The primary finding of this study is the identification of significant infradian rhythms in cardiovascular variables, specifically in evening heart rate, with a shared and synchronized period of 173 days among participants. We propose that affective changes associated with different stages of the mission, along with the cohesive functioning of the crew as a unit, may contribute to these variations.

1. Introduction

The cyclic patterns of physiological and behavioral changes that occur over periods longer than 24 h, known as infradian rhythms, are important in understanding the complex interaction between the environment and the individual. These rhythms are often associated with changes in endocrine or autonomic nervous system regulation and can be modulated by several external factors including light/dark cycles, mood, and physical activity. Blood pressure and heart rate are two of the most studied parameters in the context of infradian rhythms.

Infradian rhythms are characterized by circaseptan, monthly or seasonal periods. Circaseptan periods are often viewed as a reflection of

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a social weekly schedule. However, they are also observed in subjects isolated from society, suggesting that they have endogenous components that are amenable to manipulation by cycles in the environment [1]. Monthly periods are typically observed in women, in whom the menstrual cycle is associated with endocrine variations that contribute to cardiovascular changes [2]. Seasonal changes have been observed in many studies performed in various geographic regions with different climate types. The alternation of seasons is accompanied by changes in weather conditions (air temperature, atmospheric pressure, relative air humidity, and partial density of oxygen in air) and the length of daylight, causing adaptive reactions in the functions of the body. For example, the seasonal affective disorder is thought to be linked to changes in the body's circadian rhythm and the production of melatonin, causing changes in cardiovascular variables [3]. However, in human populations, fluctuations are minimized by means of artificial illumination, room heating, air conditioning, wearing clothes, and changing of the liquid and food consumption [4].

One interesting aspect of infradian rhythms in heart rate and blood pressure is the possibility of synchronization between individuals. Previous research has suggested that the synchronization of short-term physiological rhythms can occur between individuals who are in proximity or have a social connection, and may be attributable to a variety of factors, including a common effect of the environment and social connection on the autonomic nervous system [5–7]. Additionally, the influence of cognitive and behavioral mechanisms may also play a role in the synchronization of physiological rhythms between individuals. These mechanisms are thought to be related to the perception, attitude, and thought processes of an individual, which can influence their physiology and potentially synchronize it with another individual [8,9]. However, little is known about the potential for synchronization between individuals of longer-period rhythms.

This holds particular importance in the realm of long-term human space exploration, especially concerning Mars exploration. The isolation, confined spaces, and altered environmental cycles encountered in space missions requires the understanding of human physiological and psychological adaptability. Long term confinement simulation studies are crucial in this context as they simulate the conditions of long-duration space travel, providing insights into the potential impacts on human health and behavior.

The Mars500 study stands out as a prominent example, involving a 520-day long simulated mission to Mars, in which six crew members were confined in a single module for the duration of the experiment [10]. The experiment provided a unique opportunity to study the effects of long-term isolation on biochemical, physiological, and psychological parameters, as well as the possibility of synchronization of these parameters between the crew members. Thus, the present study was conducted to assess and characterize infradian heart rate and blood pressure rhythms and to evaluate the possible synchronization of these parameters between participants of the Mars500 study.

2. Material and methods

2.1. Participants

Six healthy nonsmoking male participants (mean ± SD age: 34 ± 5 yrs) participated in this 520-d confinement study. The group had diverse scientific and cultural background: three Russians (a surgeon, a physician, and an engineer), one Chinese (an astronaut trainer), and two Europeans (both engineers).

2.2. Design

The Mars500 study (http://www.esa.int/Mars500) was organized by the Institute for Biomedical Problems (IBMP) of the Russian Academy of Sciences and the European Space Agency (ESA). The study simulated a Mars mission in almost all aspects (duration, crew composition, activities, communications, full life support), with the natural exception of microgravity and radiation exposure. The crew was selected from a group of 9600 applicants, following criteria like those applied in astronaut selection. The six members followed an astronaut training program that included survival and team building activities.

Participants were confined in the isolation facility at IBMP, located in the northwest of Moscow (55° 46′ 48″ N, 37° 31′ 25″ E), from the 3rd of June 2010 to the 4th of November 2011. The simulated journey included the undocking from orbital assembly (15 Jun 2010), transfer to heliocentric orbit towards Mars (23 Jun 2010), shifting to spiral orbit towards Mars (24 Dec 2010), entering circular orbit around Mars (1 Feb 2011), undocking and landing on Mars (12 Feb 2011), egresses on simulated Martian Surface (14, 18 and 22 Feb 2011), ascent and docking (23, 24 Feb 2011), entering into spiral orbit away from Mars (2 Mar 2011), transfer to heliocentric orbit towards Earth (7 Apr 2011), and shifting to spiral orbit towards Earth (13 Oct 2011). The crew participated in more than 100 scientific protocols comprising physiological, psychological, and social issues of confinement. Their schedules were organized in 8-h periods of work, spare time, and sleep, with no night shifts. The lighting regime was individually managed by the crewmembers, with lights turned off during the sleep period, typically from 23:30 to 07:30 [11]. During the different phases of the missions, workload was maintained similar.

The isolation facility comprised four pressurized, hermetically sealed, and interconnected habitat modules resembling a spacecraft, with the ability to communicate at any time with Mission Control [11–13]. Ambient temperature was maintained constant at 24 °C, with a relative humidity of 35–45 %, normal barometric pressure, and artificial lighting conditions of 50–300 lux. Units included a habitable module (six compartments, a main control room and a toilet), a medical module (two medical berths, medical and research equipment, and a toilet), a storage module (physical training facilities, food storage, a greenhouse, and a bathroom), a Mars landing module simulator and a Martian surface simulator. The total habitable volume was 550 m^3 [11–13].

Procedure. Throughout the experiment, three consecutive measurements of blood pressure and heart rate were taken using an automatic sphygmomanometer with participants in a sitting position, and then averaged. Measurements were collected twice daily, once in the morning (08:00) and once in the evening (22:00). For all participants and variables, missing observations where less than 5 %, and were replaced by linear interpolation.

2.3. Analyses

Stage and time effects. Windows of different lengths were considered for the analyses of the effect of journey stages: Earth orbit (20 days); heliocentric orbit towards Mars (184 days; divided approximately in three thirds); spiral orbit towards Mars (40 days); circular orbit around Mars, undocking, landing, egresses, ascent, and docking (29 days); spiral orbit away from Mars (36 days); heliocentric orbit towards Earth (185 days; divided approximately in three thirds); spiral orbit towards Earth (26 days). Differences between means were assessed through a mixed model design including stage and time as fixed factors and participant as a random factor [14,15]. The structure of the residuals was analyzed to assess normality and homoscedasticity. Post hoc analyses were conducted using the Tukey correction [16].

Rhythm detection. The Lomb-Scargle periodogram was used to explore the presence of rhythms in heart rate and blood pressure signals detection [14,17]. A peak detection function was used to detect peaks, with a p-value threshold of 0.05 as the minimum amplitude threshold for peak detection.

Rhythm characteristics. To describe the significant rhythms identified in the periodogram a cosine curve was fit to the data using the least squares method to minimize the residual sum of squares, using a single- (when only a significant period was detected) or multiple- (when more than one period was detected) component cosinor model [14]. Thus, for

each identified period, the MESOR (Midline Estimate Statistic of Rhythm), amplitude (half of the difference between the maximum and the minimum of the fitted function), acrophase (the time at which the maximum of the rhythm occurs) and % rhythm (the fraction of the total variance explained by the model) were calculated [18,19]. When similar individual periods were identified, a Rayleigh test was employed to assess if acrophases follow a unimodal deviation from uniformity [20]. Then, a population-mean cosinor procedure was applied to describe the crew rhythm as a whole [18,19,21–23].

Phase coupling. To measure the degree of heart rate and blood pressure phase synchronization between participants, the signals of every pair (n = 15) of participants were analyzed. First, time series were low-pass filtered with a cut-off frequency of 7 days. Then, the Hilbert transform was applied to obtain the instantaneous phase of each time series [Φ participant $_x$ (t_i); Φ participant $_y$ (t_i)]. Basically, the Hilbert transform facilitates the extraction of the instantaneous phase of each time series by creating an analytic signal that encompasses both amplitude and phase information, allowing for a direct and efficient analysis of the signal's time-varying phase characteristics without losing the original temporal information [24,25]. It is considered that a couple of time series are synchronized if the phase difference φ (t_i) = Φ participant $_x$ (t_i) - Φ participant $_y$ (t_i) is constant. The phase difference can be quantified by $\gamma = <\cos(\varphi(t_i) \bmod 2\pi)>^2 + <\sin(\varphi(t_i) \bmod 2\pi)>^2$, where the $<\cdot>$ brackets denote an average in the considered period. Values of γ range from 0 to 1; $\gamma = 0$ when both time series are completely desynchronized and $\gamma = 1$ when the signals are completely synchronized [26]. As synchronized patterns may arise randomly, surrogate data was used to create a random sample of data to compare with the observed

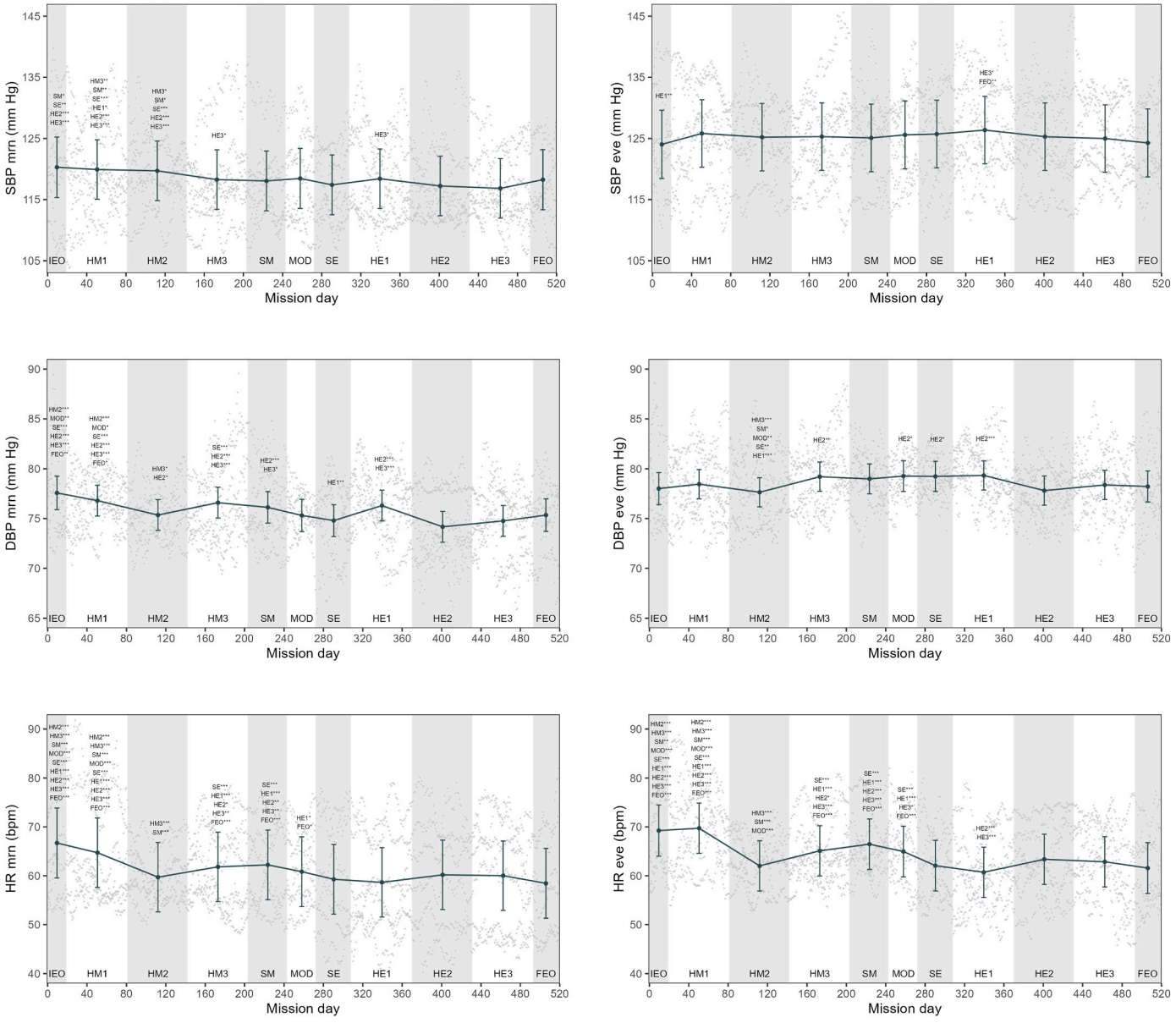

Fig. 1. Morning (left) and evening (right) values of systolic blood pressure (SBP, top), diastolic blood pressure (DBP, middle), and heart rate (HR, bottom) throughout the different stages of the Mars500 mission. Individual values were filtered using a seven-day filter to reduce short-term noise and enhance the visualization of long-term variations. The error bars represent the estimated marginal means and 95 % confidence intervals derived from the model. While all the measurements exhibit significant variations, the most visually noticeable changes are observed in the heart rate. IEO: initial Earth orbit; HM1: heliocentric orbit towards Mars (1st third); HM2: heliocentric orbit towards Mars (2nd third); HM3: heliocentric orbit towards Mars (3rd third); SM: spiral orbit towards Mars; MOD: Mars orbit and descent; SE: spiral orbit towards Earth; HE1: heliocentric orbit towards Earth (1st third); HE2: heliocentric orbit towards Earth (2nd third); HE3: heliocentric orbit towards Earth (3rd third); FEO: final Earth orbit. ; *p < 0.05, **p < 0.01, ***p < 0.001.

data. The surrogate pairs of signals were created leaving unchanged the signal of one of the participants of the pair and randomizing the sequence of the signal of the other participant. This method preserves the distribution characteristics of the signal, such as retaining the original mean and standard deviation of the observations, while entirely disrupting the temporal structure [27]. Gamma values were calculated across the entire duration of the mission and within one-month bins, both for actual and surrogate data. Differences of gamma values between actual and surrogate data for the whole mission were evaluated using independent samples t-tests, with Cohen's d reported as a measure of effect size. Variations of gamma values within the mission stages and between actual and surrogate data were assessed through a mixed model design including stage and data source (actual vs surrogate) as fixed factors, participant pair as a random factor, and an additional factor to model variance differences along stages and between sources [15]. The structure of the residuals was analyzed to assess normality and homoscedasticity. Post-hoc differences between actual and surrogate data were evaluated using Tukey analyses [16].

2.4. Ethical aspects

The study protocol was approved in advance by the ESA Medical Board and the Ethics Committee of the University Hospital Gasthuisberg of Leuven, Belgium, which complied with all guidelines stated in the Declaration of Helsinki. Each participant provided written informed consent before participating. For this study, participant original IDs were randomized to maintain anonymity.

3. Results

3.1. Variations over time

SBP, DBP and HR variations are shown in Fig. 1. Interactive effects of time and stage were significant for all variables: SBP [$F(10,6213) = 6.1$, $p < 0.001$, $R2 = 0.15$], DBP [$F(10,6213) = 6.2$, $p < 0.001$, $R2 = 0.10$], and HR [$F(10,6213) = 2.5$, $p < 0.01$, $R2 = 0.09$].

SBP morning values [$F(10,3104) = 33.8$, $p < 0.001$, $R2 = 0.02$] were higher during the first part of the outbound journey (Earth orbit till 2nd third of heliocentric orbit towards Mars) than during the spiral orbit towards Earth and the two last thirds of heliocentric orbit towards Earth (Fig. 1, top-left panel). SBP evening values [$F(10,3104) = 2.9$, $p = 0.001$, $R2 < 0.01$] remained stable with a minimum at departure from Earth orbit and a maximum during the first part of heliocentric orbit towards Earth (Fig. 1, top-right panel).

DBP morning values [$F(10,3104) = 13.5$, $p < 0.001$, $R2 = 0.04$] showed a local minima during the heliocentric orbit towards Mars and a local maxima during the heliocentric orbit towards Earth (Fig. 1, middle-left panel). DBP evening values [$F(10,3104) = 5.8$, $p < 0.001$, $R2 = 0.02$] showed higher and stable values after the last part of the heliocentric orbit towards Mars until the first part of the heliocentric orbit towards Earth (Fig. 1, middle-right panel).

HR morning values [$F(10,3104) = 33.8$, $p < 0.001$, $R2 = 0.04$] showed a local minimum during the heliocentric orbit towards Mars and during the heliocentric orbit towards Earth (Fig. 1, bottom-left panel). HR evening values [$F(10,3104) = 58.2$, $p < 0.001$, $R2 = 0.09$] showed a similar but more pronounced pattern than morning HR (Fig. 1, bottom-right panel). It was the only model that explained more than 5 % of the observed variance.

3.2. Periodogram

Table 1 shows the results of periodogram analyses for each participant and assessed variable. Periodogram analyses show that most participants have at least one significant infradian rhythm in any of the considered variables with main peaks that range from 22 days to 520 days depending on the participant and variable considered. Two significant infradian rhythms were detected in eight occasions and three significant rhythms were detected in only one participant, in evening HR. Interestingly, evening HR showed peaks with a similar period of 173 days for all participants.

Table 1
Periodogram analysis.

Variable	Participant	Morning		Evening	
		period	power	period	power
SBP	5001	260	0.07	ns	ns
	5002	519	0.08	519	0.17
	5003	31	0.04	ns	ns
	5004	260/43	0.16/ 0.04	519	0.12
	5005	ns	ns	ns	ns
	5006	519	0.13	260	0.05
DBP	5001	130/52	0.05/ 0.04	ns	ns
	5002	519	0.15	519	0.16
	5003	22	0.05	ns	ns
	5004	260	0.09	260	0.05
	5005	519/ 130	0.05/ 0.05	519	0.09
	5006	519	0.24	ns	ns
HR	5001	519/ 173	0.12/ 0.10	173	0.08
	5002	173	0.04	173	0.07
	5003	260/ 130	0.11/ 0.05	173	0.11
	5004	260/74	0.20/ 0.08	173	0.05
	5005	260	0.07	173	0.05
	5006	519/ 130	0.10/ 0.04	173/519/ 104	0.13/0.11/ 0.04

Shown are calculated periods and standardized powers of Lomb's periodograms for each variable and participant. All periods $p < 0.05$ except those denoted as ns. SBP: systolic blood pressure; DBP: diastolic blood pressure; HR: heart rate.

3.3. Cosinor

Table 2 shows cosinor fitting parameters, while Fig. 2 shows cosinor fits for all crew members for the periods calculated with Lombs' periodogram. The acrophases for evening HR ranged from 34 to 79 days (Rayleigh's test statistic = 0.984, $p < 0.001$), with a mean acrophase difference between any given pair of participants of 19.8 days. This difference is less than 1/8 of the variable's 173 day-period, suggesting that individuals share the same phase. Thus, we further calculated the population mean cosinor parameters for this variable for the period of 173 days, being (mean ± SE): mesor = 64.2 ± 2.6; amplitude = 2.2 ± 0.3; acrophase = 52.0 ± 7.2; percent rhythm = 10 %; $F(2,4) = 68.6$; $p < 0.001$.

3.4. Phase coupling

Table 3 presents the phase coupling gamma values (phase synchronization) for the entire journey. While the disparity between the actual and surrogate data was significant for all variables except evening DBP, it is noteworthy that only evening HR exhibited a substantial effect size.

Fig. 3 shows phase coupling gamma values (phase synchronization) of actual and surrogate data along time. All variables exhibited differences from the surrogate data that were dependent on the mission stages.

Morning SBP gamma values [interaction: $F(10, 488) = 2.8$, $p = 0.002$)] were higher than those from surrogate data in the initial Earth orbit, the first and second thirds of the heliocentric orbit towards Mars, and the spiral orbit towards Mars.

Evening SBP gamma values were higher than those from surrogate data [interaction: $F(10, 488) = 14.7$, $p < 0.001$)] in the initial Earth orbit, the first and second thirds of the heliocentric orbit towards Mars,

Table 2
Cosinor fitting parameters.

Var.	Part.	Morning								Evening							
		% var	Mesor		Period	Amplitude		Acrophase		% var	Mesor		Period	Amplitude		Acrophase	
SBP	5001	7	115	(0.3)	260	2.6	(0.4)	134	(6.5)	ns	–	–	–	–	–	–	–
	5002	8	118	(0.2)	519	1.7	(0.3)	277	(12.7)	17	125	(0.2)	519	2.7	(0.3)	266	(8.2)
	5003	4	111	(0.2)	31	1.3	(0.3)	10	(1.1)	ns	–	–	–	–	–	–	–
	5004	20	124	(0.2)	260	3.1	(0.3)	27	(4.0)	12	132	(0.2)	519	2.0	(0.2)	451	(9.7)
					43	1.5	(0.3)	11	(1.4)								
	5005	ns	–	–	–	–	–	–	–	ns	–	–	–	–	–	–	–
	5006	13	115	(0.2)	519	2.3	(0.3)	12	(9.4)	5	122	(0.2)	260	1.3	(0.3)	93	(8.2)
DBP	5001	10	78	(0.2)	130	1.4	(0.3)	58	(3.7)	ns	–	–	–	–	–	–	–
					52	1.2	(0.3)	36	(1.7)								
	5002	15	73	(0.2)	519	2.1	(0.2)	278	(8.7)	16	77	(0.2)	519	2.1	(0.2)	267	(8.4)
	5003	5	78	(0.1)	22	0.9	(0.2)	2	(0.7)	ns	–	–	–	–	–	–	–
	5004	9	75	(0.1)	260	1.4	(0.2)	8	(5.7)	5	77	(0.2)	260	1.2	(0.2)	4	(8.0)
	5005	10	75	(0.3)	519	2.3	(0.4)	216	(15.6)	9	77	(0.3)	519	2.9	(0.4)	250	(11.7)
					130	2.2	(0.4)	75	(4.0)								
	5006	24	75	(0.1)	519	2.7	(0.2)	505	(6.4)	ns	–	–	–	–	–	–	–
HR	5001	22	70	(0.4)	519	4.9	(0.5)	446	(9.3)	8	69	(0.3)	173	3.3	(0.5)	44	(4.1)
					173	4.4	(0.5)	36	(3.4)								
	5002	4	56	(0.2)	173	1.5	(0.3)	79	(6.0)	7	64	(0.3)	173	2.3	(0.4)	79	(4.6)
	5003	16	72	(0.2)	260	2.7	(0.3)	249	(5.0)	11	74	(0.3)	173	3.1	(0.4)	34	(3.5)
					130	1.8	(0.3)	49	(3.8)								
	5004	28	63	(0.2)	260	3.1	(0.3)	186	(3.4)	5	58	(0.2)	173	1.8	(0.3)	57	(5.0)
					74	1.9	(0.3)	9	(1.6)								
	5005	7	52	(0.3)	260	2.5	(0.4)	197	(6.7)	5	57	(0.3)	173	2.4	(0.5)	66	(5.4)
	5006	14	53	(0.2)	519	1.9	(0.2)	483	(10.8)	25	63	(0.2)	173	2.7	(0.3)	46	(2.8)
					130	1.2	(0.2)	6	(4.1)				519	2.5	(0.3)	474	(9.2)
													104	1.5	(0.3)	49	(3.1)

Shown are the percentage of explained variance, mesor [mean(SE)], amplitude [mean(SE)] and acrophase [mean(SE)] as calculated with the cosinor model for the significant periods identified with Lombs' periodogram, for each participant and variable. SBP: systolic blood pressure; DBP: diastolic blood pressure; HR: heart rate.

and the first third of the heliocentric orbit towards Earth. Also, the gamma value was lower than that of surrogate data in the second third of the heliocentric orbit towards Earth.

Morning DBP gamma values were higher than those from surrogate data [interaction: $F(10, 488) = 14.5$, $p < 0.001$)] in the heliocentric orbit toward Mars, the spiral orbit towards Mars, the Mars descent and the third third of the heliocentric orbit towards Earth.

Evening DBP gamma values were higher than those from surrogate data [interaction: $F(10, 488) = 7.8$, $p < 0.001$)] in the outbound Earth orbit, the spiral orbit towards Earth and in the first third of heliocentric orbit towards Earth.

Morning HR gamma values [interaction: $F(10, 488) = 3.7$, $p < 0.001$)] were higher than those from surrogate data in the second third of the heliocentric orbit towards Mars and in the heliocentric orbit towards Earth.

Evening HR gamma values were higher than those from surrogate data [interaction: $F(10, 488) = 22.2$, $p < 0.001$)] in all stages except in the spiral orbit towards Mars (where it was lower), and in the Mars descent, the spiral orbit towards Earth, and the final Earth orbit (where differences were non significant).

4. Discussion

The primary finding of this study is the identification of significant infradian rhythms in cardiovascular variables, specifically in evening heart rate, with a shared and synchronized period of approximately 180 days among participants.

Cardiovascular infradian rhythms were reported in several studies. A meta-analysis of panel studies revealed seasonal variations in blood pressure and heart rate. The findings showed that both systolic and diastolic BP were highest in winter and lowest in summer, while daytime HR was highest in autumn and lowest in spring and summer. These changes were attributed to neurohumoral regulation in response to the influence of day length, air temperature, or atmospheric pressure [4]. Additionally, three-month rhythms were also described in awake and asleep blood pressure, which may also be associated with meteorological fluctuations [28]. However, we did not find evidence of cardiovascular six-month periodicities in the literature that would correspond with our results. It should also be noted that these studies did not involve confined populations with controlled lighting, temperature and atmospheric pressure conditions and, therefore, are not directly comparable to ours. In contrast with these environmental factors, geomagnetic activity is a variable that cannot be controlled. Seasonal fluctuations in geomagnetic activity may have influenced participants' hemodynamic parameters. Geomagnetic activity is, on average, higher in March–October than it is in December–February, and geomagnetic perturbations are more frequent and strong near the vernal or autumnal equinoxes [29,30]. An increase in heart rate would be expected during these periods, potentially reflecting a physiological stress response [31,32]. However, these seasonal variations do not align with our results either, which show peaks in summer and winter of 2010 and summer of 2011.

To identify the possible causes of the observed periodicities, other factors must be analyzed, including autonomic, endocrine, and behavioral changes. In a previous study, we described a decrease in the amplitude of the rest-activity pattern of the parasympathetic function of the autonomic nervous system, with a progressive increase in wake vagal autonomic activity during the day, which was attributed to reduced daylight or mood changes [10]. With respect to endocrine changes, the levels of norepinephrine and serotonin increased significantly between day 168 and day 300, close to the Mars landing period. After that, they began to decline, but they remained higher than the levels observed during the first three days of the study. The level of cortisol showed an increasing tendency with fluctuations, reaching its maximum value on day 510. The level of dopamine exhibited a quasi-significant decline on day 249 and then remained relatively stable until the end of the study [33]. Overall, it can be seen that, while the autonomic and endocrine changes observed in Mars500 crew may contribute to variations in cardiovascular function, they do not appear to account for the observed six-month periodicities.

Salt intake might have also acted as a confounder. As part of a scientific protocol, dietary salt reduction was performed stepwise from 12 g

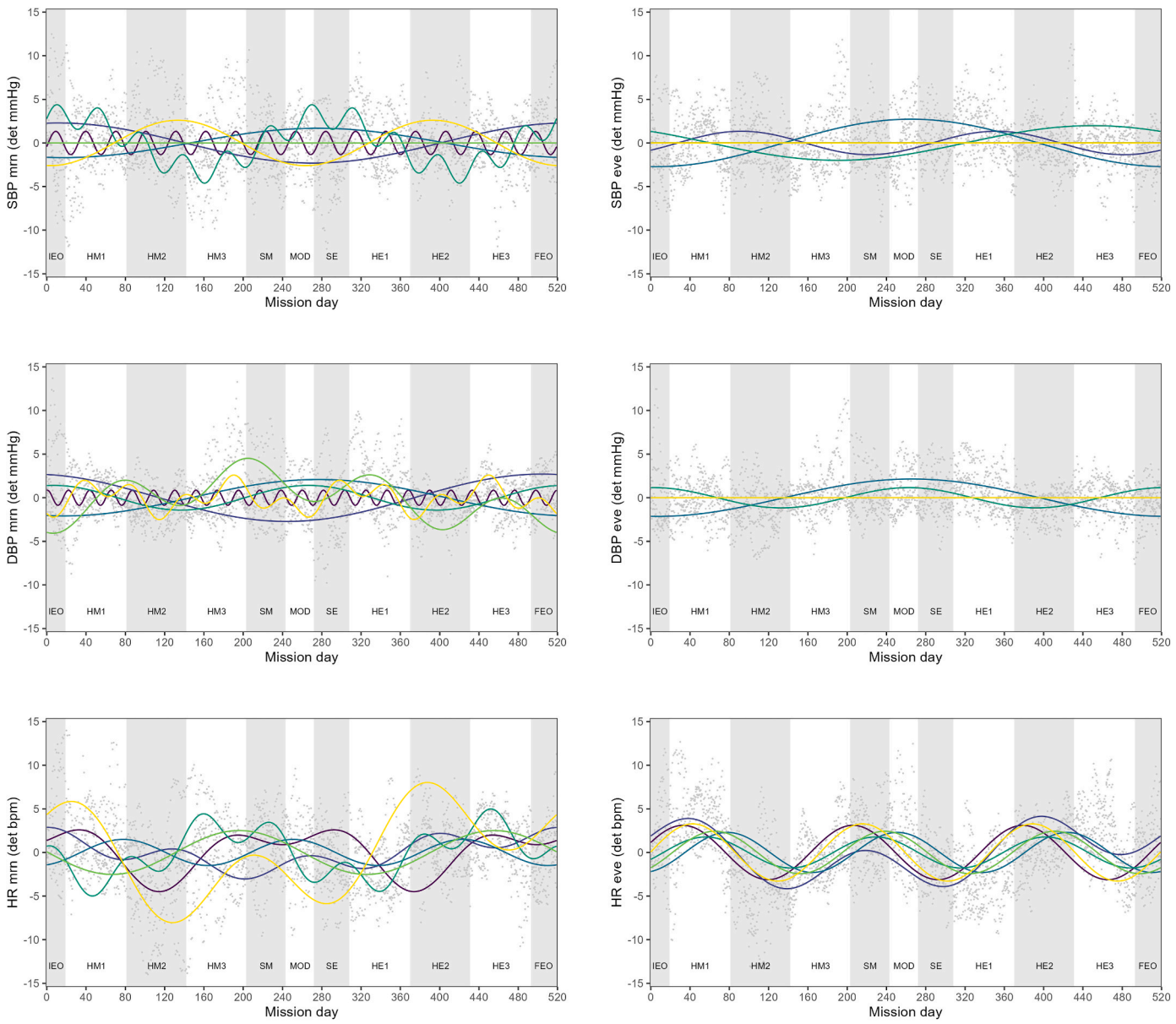

Fig. 2. Cosinor fit for morning (left) and evening (right) detrended values of systolic blood pressure (SBP, top), diastolic blood pressure (DBP, middle), and heart rate (HR, bottom) throughout the different stages of the Mars500 mission. Flat lines represent non-significant fits. Individual values were filtered using a seven-day filter to reduce short-term noise and enhance the visualization of long-term variations. All variables seem to be uncoupled with exception of evening HR. IEO: initial Earth orbit; HM1: heliocentric orbit towards Mars (1st third); HM2: heliocentric orbit towards Mars (2nd third); HM3: heliocentric orbit towards Mars (3rd third); SM: spiral orbit towards Mars; MOD: Mars orbit and descent; SE: spiral orbit towards Earth; HE1: heliocentric orbit towards Earth (1st third); HE2: heliocentric orbit towards Earth (2nd third); HE3: heliocentric orbit towards Earth (3rd third); FEO: final Earth orbit.

Table 3
Global phase coupling.

Variable	γ actual		γ surrogate		Δ	95 % CI		Cohen's d	T(14)	p
	mean	**sd**	**mean**	**sd**		**lower**	**upper**			
SBP mrn	0.04	0.04	0.01	0.01	0.03	0.00	0.05	0.85	2.34	0.032
SBP eve	0.05	0.05	0.01	0.02	0.03	0.01	0.06	0.91	2.50	0.022
DBP mrn	0.06	0.07	0.02	0.02	0.05	0.01	0.09	0.87	2.38	0.030
DBP eve	0.03	0.04	0.02	0.02	0.01	−0.01	0.03	0.30	0.81	0.425
HR mrn	0.05	0.05	0.02	0.03	0.03	0.00	0.06	0.76	2.08	0.048
HR eve	0.13	0.06	0.02	0.01	0.11	0.07	0.14	2.45	6.71	<0.001

SBP: systolic blood pressure; DBP: diastolic blood pressure; HR: heart rate; mrn: morning; eve: evening.

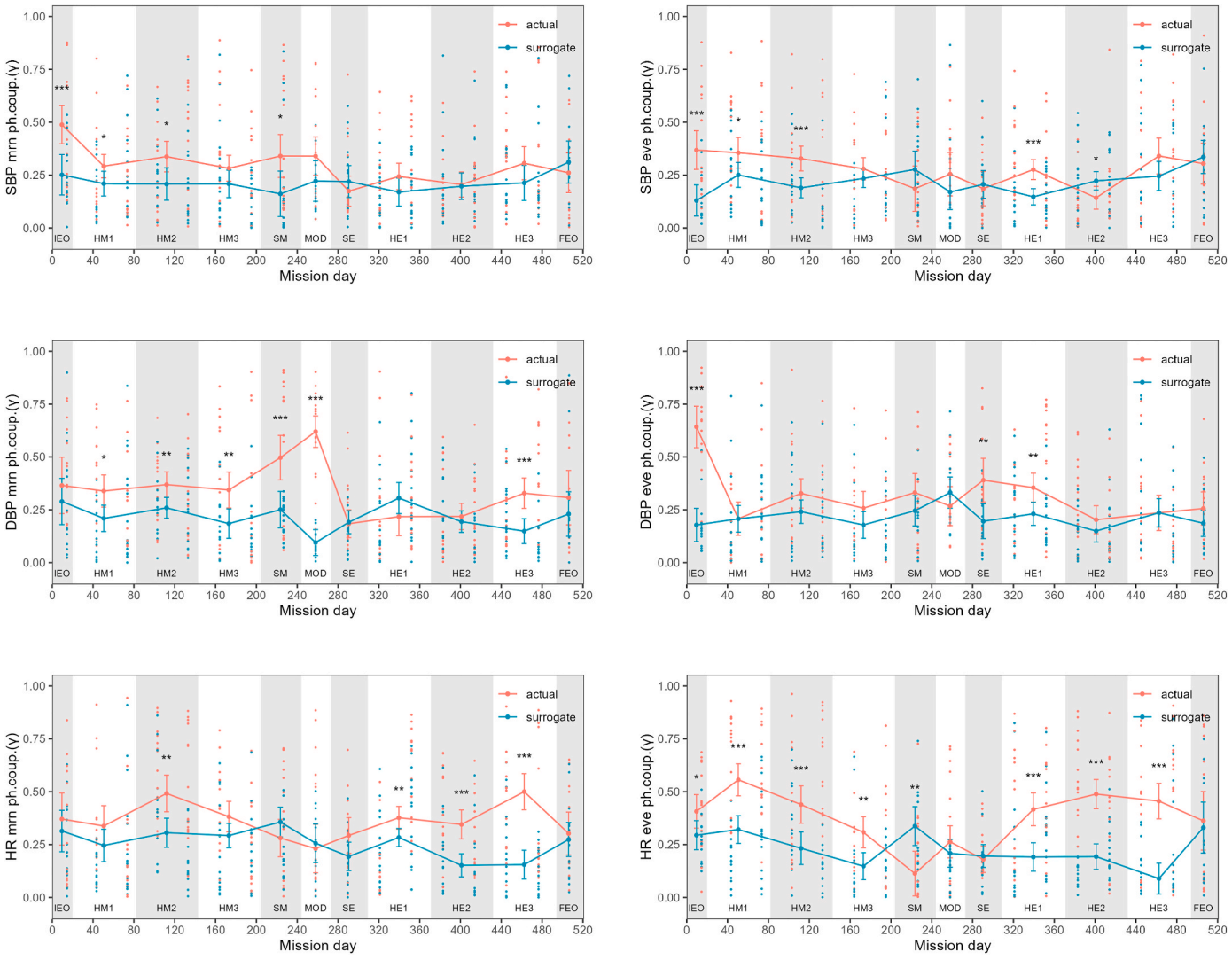

Fig. 3. Phase coupling gamma values (phase synchronization) between participants for both actual and surrogate data. This comparison is depicted for morning (left) and evening (right) values of systolic blood pressure (SBP, top), diastolic blood pressure (DBP, middle), and heart rate (HR, bottom). The dots represent the gamma value between participants of a given pair, calculated in one-month bins. Error bars depict the estimated marginal means and 95 % confidence intervals for each stage, as derived from the model. For all variables, actual data differs from surrogate data depending on the mission stages. IEO: initial Earth orbit; HM1: heliocentric orbit towards Mars (1st third); HM2: heliocentric orbit towards Mars (2nd third); HM3: heliocentric orbit towards Mars (3rd third); SM: spiral orbit towards Mars; MOD: Mars orbit and descent; SE: spiral orbit towards Earth; HE1: heliocentric orbit towards Earth (1st third); HE2: heliocentric orbit towards Earth (2nd third); HE3: heliocentric orbit towards Earth (3rd third); FEO: final Earth orbit. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

to 9 g–6 g salt per day and re-exposed thereafter to 12 g salt per day during the first 205 days of the experiment. Urinary Na + excretion, aldosterone excretion, urinary free cortisol, and urinary free cortisone showed peaks of about six days period length. The last two also showed prominent peaks with monthly period lengths. The results also showed a modest, variable salt effect on blood pressure (a decrease followed by an increase), which was delayed and required around four weeks to stabilize [34]. Fluctuations in dietary salt intake may complicate the study of pure infradian rhythms. However, even if the pattern of salt intake of the experiment appears to align with the first 180-day cycle of heart rate fluctuations reported here, this rhythm persisted beyond day 200. Moreover, the reported periods lengths of urinary Na+, aldosterone and cortisol are different from those observed in our study.

Regarding affective or behavioral changes during the simulation, in a study by Poláčková Šolcová et al., crew members qualitatively differentiated three periods: the journey to Mars, Mars, and the way back [35]. Each period had up and down moments that could explain the 180-day periodicities found in heart rate. The first period lasted two to four months and was dedicated to technical adaptation. During this phase, the crew had to learn new skills and establish rules for the mission. This was followed by a phase of monotony and dissonance, during which the crew performed their duties automatically and with strict adherence to a prescribed diet, as part of a scientific protocol. This phase eased towards the end of 2010, marking the start of the second period. This period included the first off-nominal situation (a power down simulation), a phase of celebrations, and the simulated descent onto Mars (where the crew was separated), all of which had a positive impact on the crew. Indeed, Mars descent was described as the most exciting, challenging, and critical phase of the experiment. After reuniting again in March 2011, as they had completed all the planned activities while experiencing reduced external communications, the crew's good mood turned into the feeling of burnout and a decline in motivation. Towards the end of the isolation, the restoration of communications and the anticipation of the beginning of their lives outside the isolation improved the team's mood and gave them hope that the end was near [35].

Following a quantitative approach, the same group described monthly changes in crew's mood throughout the simulation [36],

utilizing the University of Wales Institute of Science and Technology (UWIST) Mood Adjective Checklist (UMACL) [37]. Individual moods were expressed by the following 12 affective adjectives, categorized as positive (pleased, cheerful, optimistic, contented, satisfied, happy) and negative (low-spirited, dissatisfied, gloomy, depressed, sad, and sorry). Participants were asked to rate their mood on a 5-point scale (very slightly or not at all, a little, moderately, quite a bit, extremely). Authors reported the median intensity of positive, negative and overall mood, as shown in Fig. 4 [36]. Positive and negative mood remained relatively constant throughout their study, except for a decrease of positive mood observed on mission days 203 (December 22, 2010), and 385 (June 22, 2011). The intensity of overall mood exhibited four maxima: one during consecutive measurements of mission days 21 (June 23, 2010) and 49 (July 21, 2010); one on mission day 112 (September 22, 2010); and two others coinciding with the decline in positive mood, driven instead by an increased intensity of negative mood, which is not evident from its median values [36]. As depicted in Fig. 4, in our study, the population cosinor model fit to evening heart rates, displayed three maxima occurring near three of the four aforementioned dates; specifically on mission days 52 (July 24, 2010), 225 (January 13, 2011), and 398 (July 5, 2011).

Findings from another study partially support this hypothesis. Facial expressions such as smiling and laughing are the major manifestations that indicate well-being. Low percentages of facial expressions were an objective behavioral indicator of difficult periods that have been reported by crew members after 6 months (between days 159 and 243) and after 1 year (around days 355 and 369) [38]. Interestingly enough, it was reported that, while immediate physiological responses to positive and negative emotional events might be of the same magnitude, cardiovascular activation lasts longer after negative events [39], and therefore it can explain the delayed maxima seen in our study.

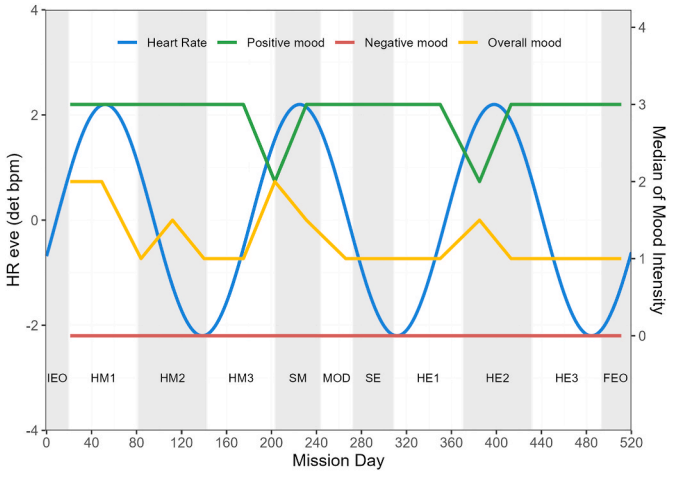

Fig. 4. Population mean cosinor fit to detrended evening heart rates, with positive and negative median mood values overlaid, as reported in Ref. [36]. Positive mood decreased on mission day 203 (December 22, 2010), and on mission day 385 (June 22, 2011). Overall mood exhibited four maxima: one during consecutive measurements of mission days 21 (June 23, 2010) and 49 (July 21, 2010); one on mission day 112 (September 22, 2010); and two others coinciding with the decline in positive mood, driven instead by an increased intensity of negative mood, which is not evident from its median values. The population cosinor model fit to evening heart rates displayed three maxima, occurring near three of the four aforementioned dates; specifically on mission days 52 (July 24, 2010), 225 (January 13, 2011), and 398 (July 5, 2011). HR eve: detrended values of evening heart rate. Mood intensity rates the extent of moods experienced by the crewmembers: 0 = very slightly or not at all, 1 = a little, 2 = moderately, 3 = quite a bit, 4 = extremely. IEO: initial Earth orbit; HM1: heliocentric orbit towards Mars (1st third); HM2: heliocentric orbit towards Mars (2nd third); HM3: heliocentric orbit towards Mars (3rd third); SM: spiral orbit towards Mars; MOD: Mars orbit and descent; SE: spiral orbit towards Earth; HE1: heliocentric orbit towards Earth (1st third); HE2: heliocentric orbit towards Earth (2nd third); HE3: heliocentric orbit towards Earth (3rd third); FEO: final Earth orbit.

The fact that the observed periodicities in evening heart rate were synchronized between individuals requires further discussion. Neural, physiological, and behavioral signals synchronize between human subjects in a variety of settings. Specifically, cardiac synchrony is a crucial component of shared experiences, considered as an objective measure of emotional processes accompanying empathic interactions. Multiple hypotheses have been proposed to explain this interpersonal synchrony. It was observed that the mere presence of other people is enough for cardiac synchrony to occur spontaneously and that it increases in function of a shared and coherent explicit emotional experience, like attending theatrical live performances [6]. In a study in which informative videos were presented to participants in an attentive and distracted condition, intersubjective correlation was observed for electroencephalography, gaze position, pupil size, and heart rate, but not respiration and head movements, suggesting that correlation emerges only for those signals that exhibit a robust brain–body connection, and thus was interpreted as the result of effective cognitive processing [8]. Another study investigated physiological synchrony (cardiorespiratory, skin conductance, and facial muscle responses) across concert audience members while listening to live string quintet performances. The results indicated that specific music features induce similar physiological responses across concert audiences and are linked to arousal, engagement, and familiarity [40]. In addition, it has been theorized that singing practices are an adaptive advantage for humans because they facilitate bonding and cohesion between group members. Investigations into the effects of singing together showed that heart rate variability became more coupled when people make long (>10 s) sounds synchronously compared to short sounds (<1 s) and baseline measurements [7].

Accordingly, the synchronization of heart rate among crew members with a period of 173 days can be attributed to the common and intense experiences associated with long-term confinement, as previously described. Emotional energy and perceived similarity are important factors in group wellbeing. It is plausible that the synchronization of moods and emotions among crew members contributes to this phenomenon by creating a shared reality and shared experience of emotions and moods [36], which in turn determines cardiac synchronization. Notably, the crew members participated in autogenic training both in the morning and in the evening as a self-regulation procedure, which may have enhanced crew cohesion [41]. However, it is unclear why evening heart rate synchronizes between individuals, while morning heart rate does not. We speculate that, in the morning, participants ready themselves for a strict and intense schedule that included different individual activities, whereas in the evening they share a common context to wind down and prepare for sleep [38].

Although the generalization of these results should be taken with caution, they could be applicable to other isolation situations. For example, confined and isolated operational settings, such as surface or deep-sea expeditions, polar missions, or high-altitude exploration, function as largely closed systems with tightly interconnected components. Within these systems, biological and psychosocial elements may interact through direct or indirect bidirectional pathways, offering an integrated perspective on the behavioral biology of teams [42,43]. Furthermore, during COVID-10 prolonged lockdown, a UK population-based study demonstrated that vaccine efficacy is influenced by psychosocial factors, which can be addressed through interventions acting as behavioral adjuvants [44]. In these contexts, our findings further support the link between social experiences and physiological changes.

There are some limitations that must be considered. Firstly, the small number of participants, though typical for this kind of study, reduces the generalizability of the conclusions. Furthermore, the study's applicability to broader demographics is limited by the homogeneous nature of the participant cohort, comprising solely healthy males within a narrow

age range. Additionally, the simulation's inability to mimic critical space travel conditions, such as microgravity and cosmic radiation, further delimits the generalizability of our results to actual space missions. In this regard, a simulated mission may also not fully capture the mental and emotional complexities of a real space voyage, potentially influencing the observed physiological responses. Finally, while the reported results were significant and are consistent with behavioral changes reported by others, a causal relation between them remains speculative.

5. Conclusion

In summary, this study has demonstrated the existence of a significant ~180-day periodicity in evening heart rate, which was found to be synchronized across participants. It is proposed that this result may be attributed to affective changes associated with the various stages of the mission, as well as the cohesive unit functioning of the crew in response to these changes. Understanding infradian rhythms of individuals living in isolated and confined environments, such as those found in space missions, can help develop strategies to promote their health and well-being, considering that physiological variables may have long-term variations. In addition, the finding of synchronization of physiological variables between individuals helps to elucidate the mechanisms underlying the interplay of social behavior and biological processes. These insights advance our knowledge of human adaptability in extreme space conditions and may have implications for human health on Earth in conditions of social isolation and confinement.

CRediT authorship contribution statement

Daniel Eduardo Vigo: Writing – review & editing, Writing – original draft, Visualization, Software, Formal analysis, Data curation, Conceptualization. **Francis Tuerlinckx:** Writing – review & editing, Methodology, Conceptualization. **Mariano Scaramal:** Writing – review & editing, Formal analysis. **Rafael Reimann Baptista:** Writing – review & editing, Data curation. **Evgeny Bersenev:** Writing – review & editing, Project administration, Conceptualization. **Omer Van den Bergh:** Writing – review & editing, Project administration, Funding acquisition, Conceptualization. **André E. Aubert:** Writing – review & editing, Project administration, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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