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Author(s): Maria Paula Bonomini, Mikel Val Calvo, Alejandro Diaz Morcillo, Florencia Segovia, Jose Manuel Ferrandez Vicente, Eduardo Fernandez-Jover

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## THE EFFECT OF BREATH PACING ON TASK SWITCHING AND WORKING MEMORY

MARIA PAULA BONOMINI†

*Instituto Argentino de Matemáticas "Alberto P. Calderón" (IAM), CONICET  
Saavedra 15, CABA, Argentina.*

*Instituto de Ingeniería Biomédica, Fac. de Ingeniería, Univ. de Buenos Aires,  
Paseo Colón 850, CABA, Argentina.  
E-mail: paula.bonomini@conicet.gov.ar*

MIKEL VAL CALVO

*Dpto. de Inteligencia Artificial, Universidad Nacional de Educación a Distancia (UNED),  
Juan del Rosal, 16, 28040, Madrid, Spain.*

*Dpto. Electrónica, Tecnología de Computadoras y Proyectos, Univ. Politécnica de Cartagena,  
Cartagena, Spain.*

ALEJANDRO DIAZ MORCILLO

*Dpto. Tecnologías de la Información y las Comunicaciones, Univ. Politécnica de Cartagena,  
Cartagena, Spain.*

FLORENCIA SEGOVIA

*Sanatorio Guemes, CABA, Argentina  
Cartagena, Spain.*

JOSE MANUEL FERRANDEZ VICENTE

*Dpto. Electrónica, Tecnología de Computadoras y Proyectos, Univ. Politécnica de Cartagena,  
Cartagena, Spain.*

EDUARDO FERNANDEZ-JOVER

*Instituto de Bioingeniería, Univ. Miguel Hernández,  
Elche, Spain.*

The cortical and subcortical circuit regulating both cognition and cardiac autonomic interactions is already well established. This circuit has mainly been analyzed from cortex to heart. Thus, the heart rate variability (HRV) is usually considered a reflection of cortical activity. In this work, we investigate whether HRV changes affect cortical activity. Short-term local autonomic changes were induced by three breathing strategies: spontaneous (Control), normal (NB) and slow paced breathing (SB). We measured the performance in two cognition domains: executive functions and processing speed. Breathing manoeuvres produced three clearly differentiated autonomic states, which preconditioned the cognitive tasks. We found that the SB significantly increased the HRV low frequency power (LF) and lowered the power spectral density (PSD) peak to 0.1 Hz. Meanwhile, executive function was assessed by the working memory test, whose accuracy significantly improved after SB, with no significant changes in the response times. Processing speed was assessed by a multitasking test. Consistently, the proportion

of correct answers (success rate) was the only dependent variable affected by short-term and long-term breath pacing. These findings suggest that accuracy, and not timing of these two cognitive domains would benefit from short-term SB in this study population.

*Keywords:* HRV; neurovisceral integration model; cognitive functions; breath control.

## 1. Introduction

The link between the prefrontal cortex, mainly involved in executive functions, and the autonomic drive of the heart is already well established.<sup>1,2</sup> In particular, Thayer et al. associated heart rate variability (HRV) to a set of neural structures implicated in executive functions when setting the basis for the neurovisceral integration model.<sup>3</sup> Thus, a change in prefrontal cortex will affect HRV and viceversa. In fact, cognitive function worsens under conditions of autonomic dysfunction.<sup>4,5</sup> Recently, resting HRV was reported to act as an index of cognitive function, finding a positive relationship between baseline HRV and cognitive performance.<sup>6,7</sup> Moreover, long-term manoeuvres aimed at increasing resting HRV have reflected improvements in many executive domains such as working memory<sup>6,8</sup> and processing speed.<sup>7,9</sup> Breathing is intimately linked with autonomic function. In fact, paced breathing at 6 breaths per minute increases HRV levels.<sup>10,11</sup> The importance of respiration relies on the possibility of voluntary control. Supporting evidence has found increases in the coherence of gamma activity and respiratory signals during interoceptive and exteroceptive attention to breathing.<sup>12</sup> Here, we made use of respiratory pacing as a manoeuvre to produce immediate HRV changes, in order to investigate whether the aforementioned HRV-prefrontal cortex interactions would hold for acute (short-term) changes.

Finally, the use of wearable devices allows us to monitor the blood volume pulse (Bvp) in a noninvasive, ambulatory way. Because it is possible to obtain the HRV from the Bvp, physiological wristbands can allow a complete HRV characterization during both breathing and cognitive load phases. Hence, the aim of this work was to characterize parasympathetic tone during three clearly differentiated states originated from different respiratory frequencies (spontaneous, normal and slow) and to investigate whether the latter would enhance executive functions in the short-term.

## 2. Materials and Methods

### 2.1. Study population and experimental paradigm.

The topics related to this work were extensively discussed in<sup>13,14</sup> and preliminary findings covering working memory were published in.<sup>15</sup> The actual piece of work extended the analysis to processing speed and breath control experience.

Two study populations were defined in this work. Group 1 was enrolled in Protocol 1 to study the short-term effect of breathing on executive functions. The participants within this group had no experience in breathing techniques. Meanwhile, Group 2 included people who had attended a breath and meditation programme from *The Art of Living* ([www.artofliving.org](http://www.artofliving.org)), with experience in breath control, specifically the Suddarshan Krya technique. They participated in Protocol 2, which aimed to investigate the long-term and short-term effects of breath control on processing speed.

All cognitive tests were obtained from Psyc-toolkit<sup>16,17</sup> and were run on a laptop. Responses were implemented as key presses in the keyboard, with response time measured as the time of key press.

### 2.2. Protocol 1.

Young healthy subjects ( $n=21$ ) aged  $34.4\pm 7.2$  years old (12 males) were enrolled. From this population, two subjects were discarded due to noisy respiratory phases and one subject was discarded due to invalid recordings in the cognitive task. All of the subjects carried out three respiratory strategies: spontaneous breathing (Control), paced breathing at a normal frequency, about 12 bpm (NB) and paced breathing at a slow frequency, below 6 bpm (SB).

During respiratory phases NB and SB, the subjects were asked to close their eyes. Following NB and SB, the subjects completed a cognitive task consisting of the 2-Back task ( $2B_{NB}$  and  $2B_{SB}$ ). In short, in the 2-Back task the participants were presented with a sequence of stimuli one-by-one. For each stimulus, they

needed to decide if the current stimulus was the same as the one presented two trials ago. The 2-Back test is a particular implementation of the N-Back test, which aims to assess working memory.<sup>18</sup> From this test, the response times and the success rates were obtained. The order of the respiratory sessions was randomized to avoid bias due to training. From this test, two measures were computed: the average $\pm$ SD of the response times, defined as the time of key press and the success rate, defined as the proportion of correct answers for all trials. In summary, five points in time were defined in this protocol: Control, NB, SB,  $2B_{NB}$  and  $2B_{SB}$ , at which HRV parameters (LF power, HF power and PSD peak) were measured. Meanwhile, working memory measures — response times and success rate — were obtained at three different points in time: Control,  $2Back_{NB}$  and  $2Back_{SB}$ .

### 2.3. Protocol 2.

Healthy subjects (n=66) attending a breath and meditation programme from *The Art of Living* were enrolled. This population was split into three groups: Suddarshan Kria group (SK): 25 subjects aged  $49.4\pm 14.6$  years old (6 male) with a 3-month or longer experience in Suddarshan Kryia technique, Breathing group (Br): 26 novice subjects aged  $35.8\pm 8.2$  years old (3 male) attending the programme for the first time and a control group (Controls): 15 subjects aged  $33.5\pm 10.3$  years old (7 male) without experience in Suddarshan Krya. SK and Br groups completed a multitasking test and immediately after they were asked to close their eyes and take 20 breaths at a frequency lower than 6 bpm (SB). Afterwards, they completed a second multitasking test. In order to assess differences between multitasking performances due to respiration, and not test habituation, the control group watched neutral images (a static sign in the monitor) instead of breath pacing in the time elapsed between *initial* and *final* multitasking tests.

Briefly, multitasking tests consist of two tasks, A and B, and participants have to randomly alternate between both.<sup>19</sup> This actual implementation used a dice as stimulus with two tasks associated: *position* of the dice (horizontal/rotated) and *dots* in it (two/three). In turn, the stimulus was presented at the top/bottom of the stimulus presentation area. The location of the stimulus acted as the cue (top:

task *position*, bottom: task *dots*), while the imperative stimulus was presented at exactly the same time. It was configured a delay of 100 ms seconds between correct answers and a 3 seconds timeout. After a wrong answer, a sign reminding the rules was shown for 1 second. This paradigm was implemented in two blocks: one of pure trials (a battery of 30 only task A trials followed by a battery of 30 only task B trials) and a second block of 60 mixed trials (trials randomly alternating tasks *position* and *dots*). The average slow down shown in the second block (mixed trials) with respect to the first one (pure trials) was defined as the mix cost, and it was obtained by subtracting the average response times:

$$Mix_{cost} = \sum_{i=0}^N RT_P(i) - RT_M(i) \quad (1)$$

where  $RT_P$  and  $RT_M$  are the response times of pure trials and mixed trials respectively, and N accounts for total trials of the mixed block.

Within mixed blocks, the difficulty to switch between tasks is expressed as the switch cost, which is defined as the slowing down on trials that immediately followed a task switch with respect to a task repeat.

$$Switch_{cost} = \sum_{i=0}^M RT_S(i) - RT_R(i) \quad (2)$$

where  $RT_S$  is the response time of the switching task,  $RT_R$  is the response time of the repeating task and M expresses total switching trials within the mixed block.

Finally, the success rate (SR) was calculated as the average of correct answers for all trials within the mixed block. The switch cost function, mix cost function and success rate were computed for both initial and final multitasking tests. To avoid cumbersome notation, a unique terminology was adopted for *initial* and *final* multitasking tests for every group, followed by specification of the variable that it refers to in any case. Thus,  $SK_1$ ,  $Br_1$  and  $C_1$  may refer to switch cost, mix cost or success rate for the *initial* multitasking tests of SK, Br and Control groups respectively. Analogously,  $SK_2$ ,  $Br_2$  and  $C_2$  may refer to switch cost, mix cost or success rate for the *final* multitasking tests of SK, Br and Control groups respectively.

To assess the differences between multitasking tests due to respiration and not test habituation, the control group watched neutral images instead of breath

spacing in the time elapsed between initial and final multitasking tests.

#### 2.4. *Parasympathetic tone: HRV computation.*

Parasympathetic (vagal) tone was assessed by spectral analysis of HRV, following guideline recommendations.<sup>20</sup> From this analysis, the high frequency power (HF), low frequency power (LF) and spectral peak of the power spectral density (PSD) of normal-to-normal (NN) inter-interval beats were computed. Inter-interval beats (tachograms) were obtained from the blood volume pulse (Bvp) signal of the Empatica E4 wrist-band.<sup>21</sup>

The E4 wristband implements a modified working principle of classic pulse oximetry and it relies in the reflective mode principles: light absorbance and reflection. In short, the E4 wristband emits green and red lights toward the skin, which are absorbed by the blood in different ways. A portion of the light is then reflected back and measured by the light receiver. The measured light during green exposure contains most of the information on the pulse wave because oxygenated blood absorbs energy in that wavelength range. Therefore, blood oxygen content acts as an indirect measure of the cardiac cycle, producing clearly defined diastolic and systolic points in the Bvp signal, from which inter-beats intervals can be obtained. Meanwhile, red light is insensitive to oxygen but is used to cancel motion artifacts.

HRV was computed as follows: minima of the Bvp (diastolic points) were detected, and the  $n$ -th pulse-to-pulse interval (PPI) was measured as the temporal distance between the  $n$ -th and  $(n+1)$ -th blood pulse minima. From these PPI series, ectopic beats were discarded and normal-to-normal (NN) inter-beat intervals series were constructed. These series were then lowpass filtered at 2 Hz (zero-phase Butterworth filter, order 4th) and detrended. Afterwards, resampling at 4 Hz was accomplished to obtain a NN function with evenly spaced samples,<sup>22</sup> on which the power spectral density was estimated by the periodogram, as follows:

$$PSD_{(w)} = \frac{1}{L^2} \sum_{k=0}^{L-1} NN_{(k)} e^{-jwk} \quad (3)$$

where  $L$  is the length of the NN function. Spectral power of the high frequency (HF) around 0.15-0.40 Hz and low frequency (LF) band around

0.04-0.15 Hz were computed and log transformed. Figure 1 shows an example of the (Bvp) signals acquired with the E4 wristband, together with their tachograms and power spectral densities (PSD). Notice the shift of the breath peak from 0.3 Hz in the NB phase, to 0.1 Hz in the SB phase and significant increase in energy, following the greatest oscillations of the tachogram.

#### 2.5. *PPG derived respiratory signals and coherence.*

Respiratory signals were obtained from the PPG signals according to.<sup>23</sup> Since PPG is secondarily modulated by respiration, AM modulation approach was implemented. To compute coherence, the respiratory signals were resampled to 4 Hz and crosscorrelated to the tachogram, on which the normalized cross power spectral density was computed as the normalized cross-spectrum between both signals:<sup>22</sup>

$$Coh_{(w)} = \frac{|\sum_{k=0}^{L-1} R_{rb}(k) e^{-jwk}|^2}{\sum_{k=0}^{L-1} r_{(k)} e^{-jwk} \sum_{k=0}^{L-1} b_{(k)} e^{-jwk}} \quad (4)$$

where  $R_{rb}(k)$  is the cross-correlation between the respiratory and the Bvp signals,  $r_{(k)}$  is the respiratory signal,  $b_{(k)}$  is the Bvp signal and  $L$  is the length of both  $b_{(k)}$  and  $r_{(k)}$ .

All those subjects who failed to show a coherence peak about 0.1 Hz were discarded from further analysis. Figure 2 presents an example showing both heart rate (top left) and respiration (middle left) signals together with their power spectral densities (PSD) on the right. The coherence signal (bottom) shows the spectral agreement between both respiration and heart rate for the SB phase.

HRV computation, extraction of respiratory signals, as well as coherence estimation was performed using Matlab.<sup>24</sup>

#### 2.6. *Statistical analysis*

##### 2.6.1. *Protocol 1*

To test the longitudinal association of HRV and breathing strategy, we used a non parametric test for repeated measures, since PSD peak values were not normally distributed. Thus, a non-parametric test aimed to compare multiple related samples, the Friedman test,<sup>25</sup> was used to compare differences in LF power, HF power, PSD peak among

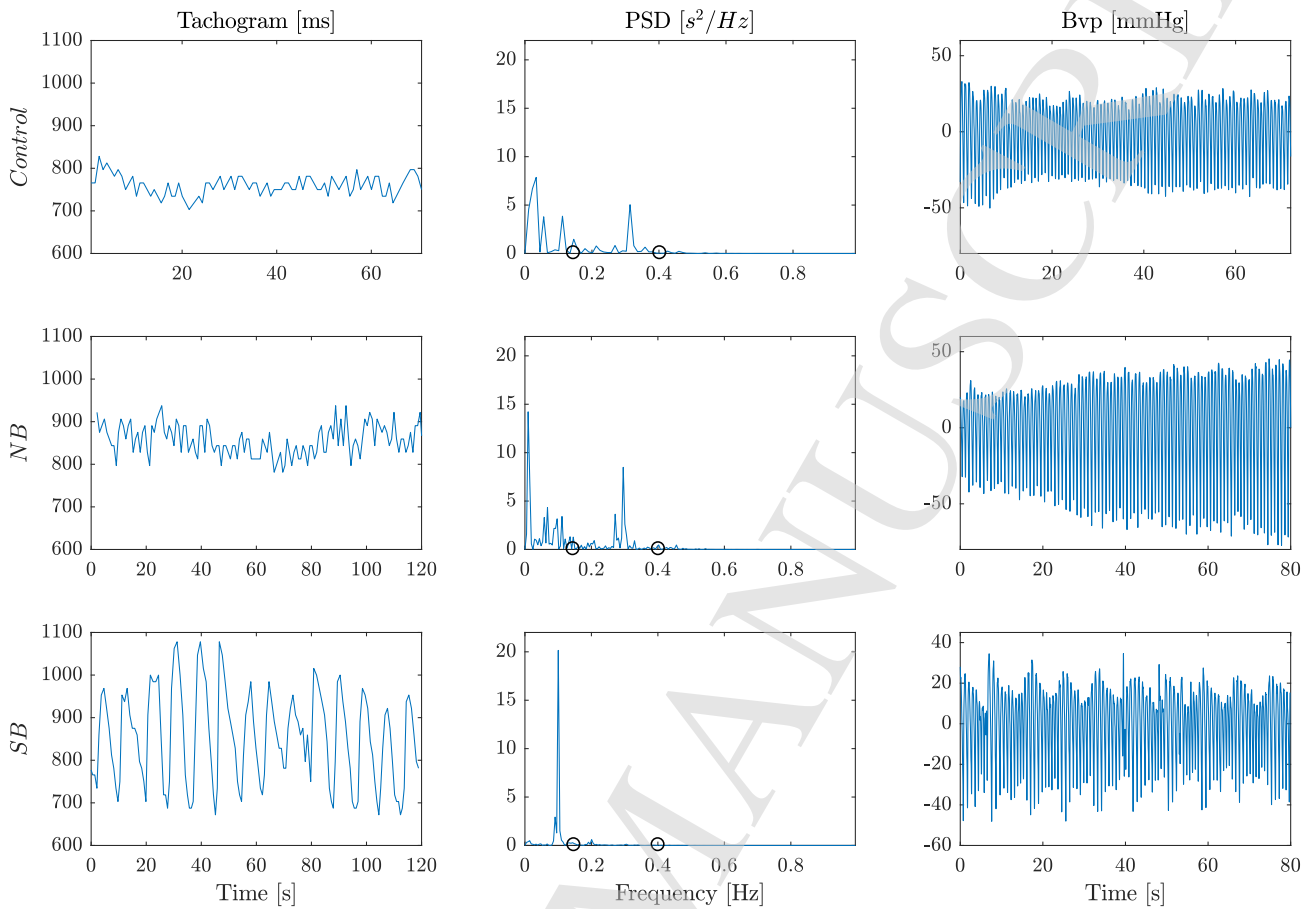


Figure 1: NN inter-interval beats (tachograms), power spectral density of HRV (PSD) and Blood volume pulse (Bvp) signals obtained with the E4 wristband for a representative subject at three breathing strategies (Control, NB and SB). From PSDs, spectral HRV was obtained, expressed in the HF/LF power and PSD peak parameters. Circles delimit frequency bands: LF (0.04-0.15 Hz) and HF (0.15-0.4 Hz). Notice the oscillatory pattern of the heart rate and the Bvp at SB (bottom left-hand and right-hand panels), with a markedly shift of the PSD peak (attributed to respiration) to the SRA resonance frequency: 0.1 Hz (bottom centered panel).

the different points in time they were measured:  $T_0$  : Control,  $T_1$  : NB,  $T_2$  : SB,  $T_3$  :  $2Back_{NB}$  and  $T_4$  :  $2Back_{SB}$ . Pairwise comparisons were performed by the Wilcoxon signed rank tests followed by Bonferroni correction for multiple comparisons.<sup>26,27</sup> Bonferroni correction was applied to the ten comparisons originated from having five test intervals (Ctrl-NB; Ctrl-SB; Ctrl- $2Back_{NB}$ ; Ctrl- $2Back_{SB}$ ; NB-SB; NB- $2Back_{NB}$ ; NB- $2Back_{SB}$ ; SB- $2Back_{NB}$ ; SB- $2Back_{SB}$ ;  $2Back_{NB}$ - $2Back_{SB}$ ). Analogously, to test the association of working memory and breathing strategy, differences in response times and success rate were assessed by the Friedman test for the points in time  $T_0$  : Control,  $T_3$  :  $2Back_{NB}$  and  $T_4$  :  $2Back_{SB}$ . Here, p-values were obtained from a Chi-

squared statistic. In this case, Bonferroni correction was applied to three comparisons (Ctrl-NB; Ctrl-SB; NB-SB).

### 2.6.2. Protocol 2

In this case, we tested the longitudinal and cross-sectional associations of processing speed using two-way repeated measures ANOVA (SK vs Br vs C Groups X Pre vs Post SB). Variables were log-transformed in order to obtain normal distributions and the Lower Bound (LB) correction for compound symmetry (sphericity) was applied. Here, p-values were obtained from an F statistic. Statistical significance was defined for  $p < 0.05$ . In addition, in order

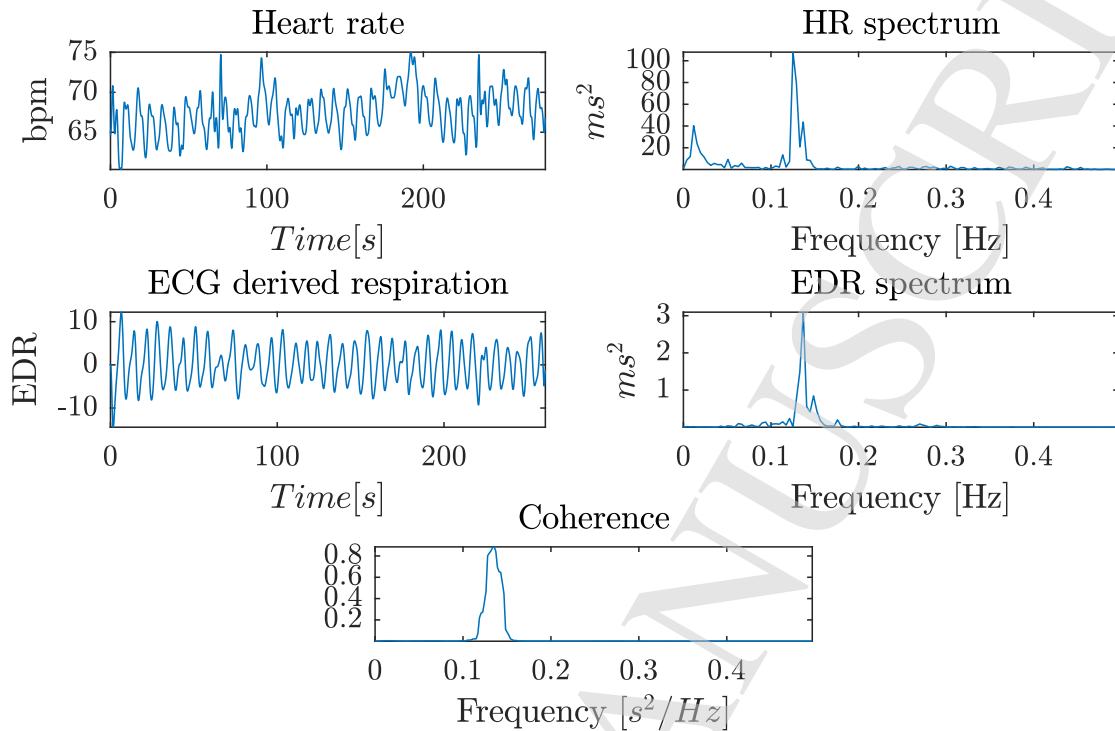


Figure 2: Coherence at the SB breathing strategy. Left-hand top and middle panels: ECG-derived Respiratory signal and Heart Rate; Right-hand top and middle panels: individual Respiratory PSD (EDR spectrum) and Heart Rate PSD (HR spectrum). The respiratory signal was obtained from the Bvp signal by AM modulation from a representative subject. Centered bottom panel shows the coherence spectrum between both respiration and heart rate signals. Note the coherence peak centered around the respiratory frequency at the SRA resonance frequency (0.1 Hz).

to assess long-term vs short-term interactions on the dependent variables, the control group was removed and a repeated measures ANOVA (SK vs Br group X Pre vs Post SB) was defined.

Finally, an analysis of covariance (ANCOVA) was included to cancel possible regression to the mean effects, since baseline levels differed across groups. Thus, follow-up measures from each subject were adjusted according to their baseline measurement.

### 3. Results

#### 3.1. HRV parameters

Figure 3 shows the vagal indices across the respiratory phases and the cognitive load periods for Group 1. The HF power during NB and SB was similar, and in both cases significantly higher than Control ( $p < 0.005$  and  $p < 0.0005$  respectively). However, the LF power exhibited significant increases with respect to Control ( $p < 0.0005$ ),  $2Back_{NB}$  ( $p < 0.0005$ )

and  $2Back_{SB}$  ( $p < 0.0005$ ). Here, the trend for NB was also similar to SB, although less markedly (NB vs Control:  $p = 0.0009$ , NB vs  $2Back_{NB}$ :  $p = 0.041$ ). The main difference between NB and SB was evidenced by the differential increase of the LF power. In addition, PSD peaks showed a downward trend for the SB,  $2Back_{NB}$  and  $2Back_{SB}$ , although failed to produce statistical significance. Notice that SB showed the greatest gain in energy in the LF band, accompanied by a shift towards 0.1 Hz of the PSD peak. This fact is in line with the cardiac coherence during the SB phase, where the PSD peak moved towards lower frequencies, showing breath dominance on the spectral content of HRV during SB (Fig. 2, bottom panel). Supporting this fact, Fig. 1 shows PSD peaks around 0.3 Hz for both Control and NB phases (top and middle center panels), and a PSD peak shift to 0.1 Hz for the SB phase (bottom center panel). In this case, it is also clear the oscillatory nature of the Bvp signal during SB (bottom right-hand

panel). Here, not only Bvp, but also respiration and heart rate oscillate at the same frequency (0.1 Hz), as is depicted in Fig. 2.

It is worth noting that during cognitive load—that is, at  $2Back_{NB}$  and  $2Back_{SB}$ —both HF and LF power remained similar to control values, at lower levels than NB and SB. However, the PSD peak remained close to that of the SB phase and moreover, moved to lower frequencies during NB (top right-hand panel). This suggests vagal withdrawal during cognitive load, since HRV power moved away from the parasympathetic frequency range (0.15–0.40 Hz).

### 3.2. Cognitive performance

Figure 4 shows the cognitive performance for subjects belonging to Protocol 1. The response time failed to produce statistical significance either for NB or SB (control:  $679 \pm 91$  ms, NB:  $611 \pm 93$  ms and SB:  $688 \pm 91$  ms,  $p=NS$ ). However, the success rate did improve significantly for the SB phase with respect to the control ( $p=0.036$ ), while the NB phase did not (Control:  $84 \pm 9$ , NB:  $89 \pm 6$  and SB:  $92 \pm 8^*$ ,  $p < 0.05$ ). Table 1 shows Friedmann test results for Success Rate and Response Time for Protocol 1. Notice that Success Rate, and not Response Time significantly improved at SB.

Regarding Protocol 2, Figure 5 shows the Switch and the Mix Cost for the multitasking test before ( $SK_1$  and  $BR_1$ ) and after ( $SK_2$  and  $BR_2$ ) SB and placebo ( $C_1$  and  $C_2$ ). Results from the repeated measures ANOVA tests showed that the factor "Group" (SK, Br and Control) exerted a significant effect on success rate:  $F(2,63)=6.72$ ,  $p=0.002$ . Analogously, the slow breathing manoeuvre "SB" (Pre vs Post), exerted an effect on success rate too:  $F(1,63)=13.91$ ,  $p < 0.0004$ . Also occurred a Group:SB interaction effect:  $F(2,63)=3.71$ ,  $p=0.029$ . However, in the case of the Switch Cost, only factor Group turned out to be statistically significant ( $F(2,63)=4.27$ ,  $p=0.018$ ), probably due to differences in baseline values. Finally, none factor (SB or Group) affected the Mix Cost.

In the second design, when analyzing the long-term vs short-term interaction and hence, ruling out the control groups, neither factor affected the switch nor the mix cost function, while the Group factor (SK vs Br Group) accounting for long-term ef-

fects, produced a significant effect on success rate ( $F(1,49)=11.12$ ,  $p=0.001$ ). The same held for the short-term factor SB (pre vs post SB), with similar strength ( $F(1,49)=11.59$ ,  $p=0.001$ ). Finally, the long-term:short-term interaction exerted an effect on success rate as well ( $F(1,49)=6.22$ ,  $p=0.01$ ), suggesting that breath pacing training facilitated the effect of the short-term SB manoeuvre.

Finally, ANCOVA results are shown in Table 2. Here, SK vs Control groups were analyzed for the outcomes *Success Rate*, *Switch Cost* and *Mix Cost*. The regression equation was the following:

$$Follow_{up} = a_0 + a_1 * (baseline - \mu_b) + a_2 * group \quad (5)$$

where  $\mu_b$  was defined as the mean of the baseline measurements.

Notice that only *Success Rate* presented significant treatment effect and group differences, while the *Switch Cost* only presented group differences while the *Mix Cost* did not present any statistical difference.

## 4. Discussion

Most of the studies relating HRV and cognitive function rely on resting HRV; that is, HRV values at baseline. In this work, we have investigated the link of cognition and HRV induced by three different breathing strategies: spontaneous and paced at normal and slow rate. These breath manoeuvres produced three clearly differentiated ANS states, which in turn, preconditioned the performance on executive functions and processing speed.

The methodology in this study was devised such that breathing strategies acted as preconditioners for the cognitive tests that were run immediately after each breathing phase. Consequently, voluntary breath did not compete for neural resources at the cognitive load periods, as reported in Nierat et al.<sup>28</sup> Here, timed up-and-go tests diminished performance as long as breath control was superimposed to test execution. This was mainly attributed to recruitment of prefrontal areas for breath pacing instead of cognitive function.

### 4.1. Breath-induced autonomic tone

There is a clear consensus on the inverse relationship between HRV and respiratory frequency, increasing



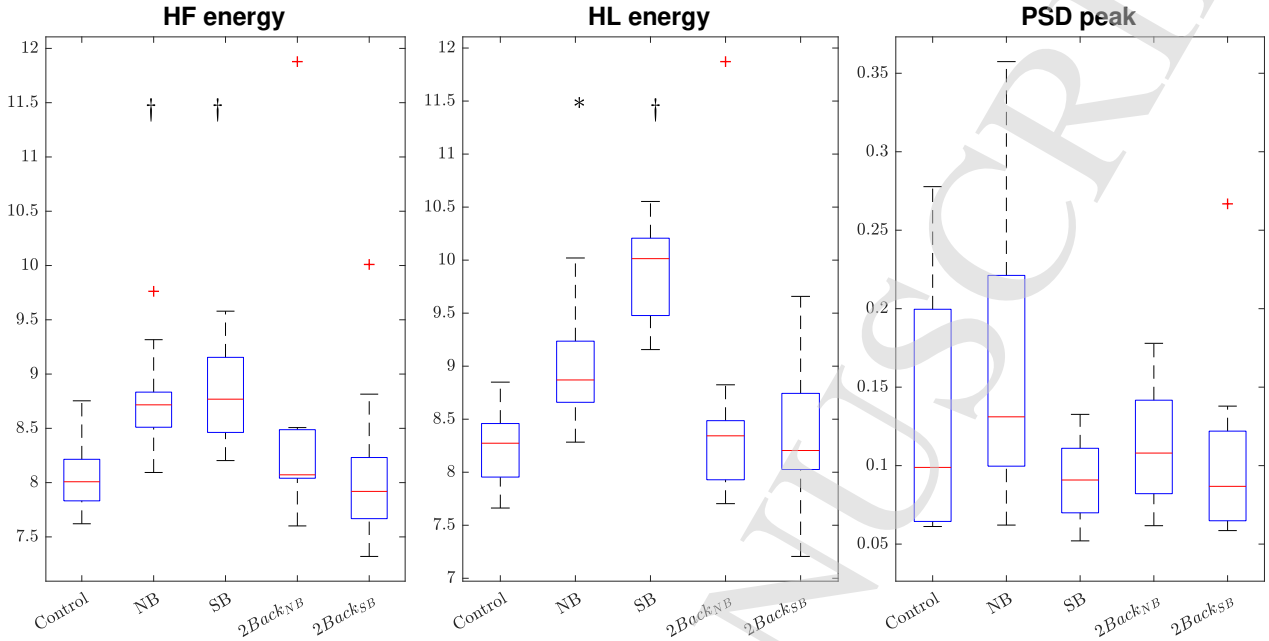


Figure 3: Boxplots of HRV parameters for Group 1 at the three breathing strategies (Control, NB and SB) and during working memory tests ( $2Back_{NB}$  and  $2Back_{SB}$ ). On each box, the central mark is the median, the edges of the box are the 25th and 75th percentiles and the whiskers extend to the most extreme data points the algorithm considers to be not outliers, while outliers are plotted individually (red marks). p-values were obtained from the Friedman test with pairwise comparisons by the Wilcoxon signed rank test and Bonferroni-corrected for multiple comparisons: \* $p < 0.005$  vs Control, † $p < 0.005$  vs Control,  $2Back_{NB}$  and  $2Back_{SB}$ .

Table 1: Friedman results for Success Rate and Response Time in Protocol 1. SS:sum of squares; df: degree of freedom; MS: mean squares.

	Success Rate					Response Time				
	SS	df	MS	$Chi^2$	$p > Chi^2$	SS	df	MS	$Chi^2$	$p > Chi^2$
<b>Columns</b>	5.71	2	2.85	6.10	0.04	2.62	2	1.31	2.62	0.26
<b>Error</b>	24.28	30	0.80			29.37	30	0.97		
<b>Total</b>	30	47				32	47			

Table 2: ANCOVA results for SK vs Control. SE: standard error; t-Stat: t statistical.

	Success Rate				Switch Cost				Mix Cost			
	Estimate	SE	t-Stat	p	Estimate	SE	t-Stat	p	Estimate	SE	t-Stat	p
<b>a0</b>	4.57	0.02	226.67	9.3e-60	6.45	0.17	35.86	2.4e-30	6.87	0.10	66.40	4.3e-40
<b>a1</b>	0.45	0.04	9.88	6.23e-12	-0.08	0.05	-1.68	0.10	-0.01	0.03	-0.40	0.690
<b>a2</b>	-0.02	0.01	-2.06	0.04	-0.32	0.12	-2.62	0.01	-0.12	0.07	-1.76	0.085

HRV while decreasing respiratory rates.<sup>29,30</sup> In particular at 6 breaths per minute, the heart rate oscillates with respiration exactly in phase, maximizing gas exchange. This is due to maximization of the respiratory sinus arrhythmia (RSA), which consists of increasing the heart rate at inspiration and decreasing it at expiration.<sup>31</sup> This phenomenon regulates the

efficiency of gas exchange at the alveoli, providing a greater respiratory blood flow when the lungs are with maximum content of oxygen (inspiration) and diminishing it when the alveolar content is richest in carbon dioxide (expiration). In normal breathing, however, there is a delay between RSA and respiration, producing a moderated gas exchange efficiency.

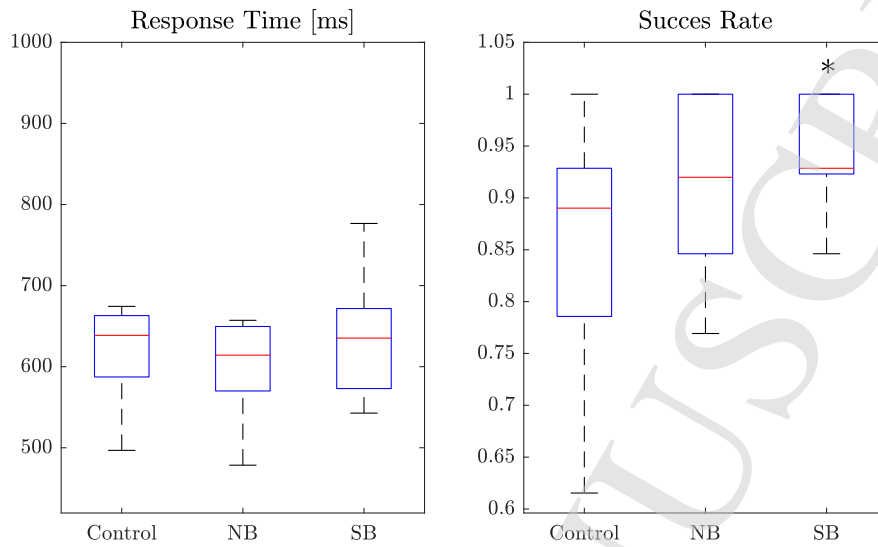


Figure 4: Working memory test performance for Group 1. Boxplot representations for Response Time and Success Rate. On each box, the central mark is the median, the edges of the box are the 25th and 75th percentiles and the whiskers extend to the most extreme data points the algorithm considers to be not outliers. Response times did not change with breathing strategies (Control, NB and SB) while the success rate did significantly improved for SB. p-values were obtained from the Friedman test with pairwise comparisons by the Wilcoxon signed rank test and Bonferroni-corrected for multiple comparisons: \* $p < 0.05$  vs control.

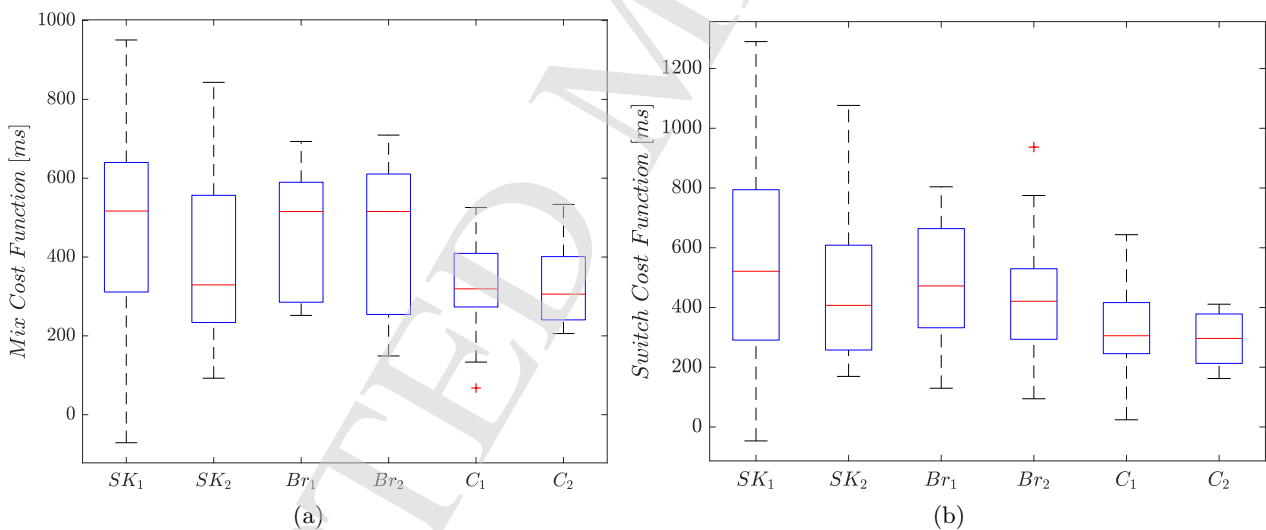


Figure 5: Multitasking response times. Boxplots of the Mix cost (left) and Switch cost (right) functions for the *initial* and *final* multitasking tests for the three groups participating in Protocol 2: SK group (SK), Breathing group (Br) and Controls (C). On each box, the central mark is the median, the edges of the box are the 25th and 75th percentiles and the whiskers extend to the most extreme data points the algorithm considers to be not outliers, while outliers are plotted individually (red marks).

At SB, this delay turns to zero, putting the RSA and heart rate oscillations together, at  $0^\circ$  phase relationship, maximizing this way gas exchange.<sup>32</sup>

However, controversy remains about which part of HRV power increases with slow breathing. Some authors reported LF increases,<sup>30,33,34</sup> while others re-

ported increases in HF power instead.<sup>35,36</sup> In this work, we have found increases in the HF power during NB and SB respiratory phases with respect to control in roughly the same proportion. However, LF power at SB presented a differential increase with respect to NB and control, producing the highest LF power (Figure 3). We speculate that this HF upward trend in both NB and SB could be caused by eye closure. Meanwhile, the outstanding LF power at SB over all other phases, including the NB, with already increased LF power, could be attributed to the respiratory frequency because it is the differential action between NB and SB.

#### 4.2. *Breath-induced changes in executive functions*

Protocol 1 investigated the role of breathing strategies on executive functions in the short term (Figure 4). Here, the success rate after the slow breathing phase was significantly higher than control (spontaneous breathing). These results are consistent with the literature, mostly reporting long-term associations between baseline HRV values and the success rate found in working memory tests across young populations<sup>6</sup> or the elderly.<sup>7</sup>

Nevertheless, response times after SB presented an upwards trend, even though it was not statistically significant. These results contrast with the previously mentioned reports. As a matter of fact, Mahinrad et al. found that higher HRV values at baseline were correlated to shorter response times in an elderly population.<sup>7</sup> In line with this, Hansen et al. also presented decreases in response times of executive functions for a 4-week trained group of marines, where physical training appeared as a manoeuvre to increase baseline HRV.<sup>6</sup> However, Britton et al. did not find an association between autonomic function and cognitive performance in a middle-aged population.<sup>37</sup> At this point, we speculate that the gain in success rate obtained in Protocol 1 could be at the expense of a longer response time, so that no net gain could be attributed to the short term effect of a paced breathing strategy on the working memory test performance.

#### 4.3. *Breath-induced autonomic changes and processing speed*

The results from Protocol 2 show an increase in success rate, a decrease in the switch cost function and no change in the mix cost function for subjects with experience in the Suddarshan Kryia technique (Figure 5). This is in line with Mahinrad et al, who found associations between lower HRV at baseline and worse performance in processing speed emerging from a Letter-Digit Coding test.<sup>7</sup> The fact that a gain in switch times, as well as success rate, was present for higher preconditioned HRV values by the SB strategy in the group with experience in breath control (SK group) was expected because the switch cost function measures the flexibility of the neural system before changing stimuli in a task switching paradigm. This versatility involves inhibition and sustained activity processes, both taking place at the prefrontal cortex, which is a constituting block of the neurovisceral integration model.<sup>3</sup> Then, the finding of a net gain in a task switching test suggests that the SB manoeuvre did affect model structures upstream. However, this observation was only true for the SK group, meaning that short term modulation by means of respiration would exert some effect in the presence of a certain HRV background.

#### 4.4. *Study limitations*

The SK and Br groups presented some heterogeneity with respect to Controls in Protocol 2. First, the Control group was comprised of college students while the SK and Br groups included older people. Second, the gender for the SK and Br groups were predominantly female. Finally, there could also be a bias regarding personality features, such as patience, self-control or introspection that could act as self-selective in the SK group. However, in the long-term vs short-term analysis of Protocol 2, the Control group was ruled out and these methodological flaws were compensated, since both SK and Br groups presented a similar gender imbalance, average age and baseline measures. In addition, ANCOVA results, computed to compensate for baseline differences between SK and Controls, supported the findings from ANOVA results, with the slow breath pacing exerting significant effects on success rate and not Switch nor Mix costs. Nevertheless, future methodology will aim to recruit college students for all of the

groups, with a special focus on training a group of students in breath control to obtain an homogeneous SK group.

## 5. Conclusions

The short-term SB manoeuvre exerted an effect on the success rate, and not timing, of two cognitive domains tested in this population, namely, executive functions and processing speed. In addition, when testing for the SK and BR groups, experience did exert an effect on the success rate, suggesting that people trained in slow paced breathing would be the ones who benefit most from the short-term SB manoeuvre.

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