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A pilot study of the effects of suboccipital fascial release on heart rate variability in workers in the clothing industry: Randomized clinical trial



Bruna Luísa Silva ^a, Lara Alves de Oliveira ^a, Camila Medeiros Costa ^a,
Cristiano Queiroz Guimarães ^{b, c}, Leonardo Sette Vieira ^c, Andrei Pereira Pernambuco ^{a, d, *}

^a Centro Universitário de Formiga - UNIFOR-MG, Formiga, Minas Gerais, Brazil

^b Faculdade Ciências Médicas de Minas Gerais - CMMG, Belo Horizonte, Minas Gerais, Brazil

^c Academia Brasileira de Fâscias (ABFâscias), Belo Horizonte, Minas Gerais, Brazil

^d Universidade de Itaúna - UIt, Itaúna, Minas Gerais, Brazil

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ABSTRACT

Introduction: The routine of work inside the clothing industry has been associated with high levels stress perceived by the worker. The parameters of heart rate variability (HRV) are objective markers of the organic response to stress.

Objective: This trial evaluates if suboccipital fascial release (SFR) technique was able to influence HRV parameters in workers in the clothing industry.

Methods: Randomized clinical trial. Forty women who worked in clothing industry were randomized into two groups, untreated group (UG, n = 15) and treated group (TG, n = 25). Heart rate (HR) and HRV parameters were measured using a heart rate monitor. Statistical analysis of data distribution and within group and between groups analysis were performed with α adjusted to 0.05.

Results: Within group analysis in the TG showed significant changes in the following parameters: sympathovagal balance (low frequency/high frequency - LF/HF) ($p \leq 0.01$), mean RR intervals ($p \leq 0.01$), mean heart rate (Mean HR) ($p \leq 0.01$), minimum heart rate (Min. HR) ($p = 0.02$) and maximum heart rate (Max. HR) ($p \leq 0.01$). Between group analysis, on the other hand, showed significant variations in Very Low Frequency (VLF) ($p = 0.04$) and Max. HR ($p = 0.03$) in favor of the TG. No significant changes were observed in the UG during the same period.

Conclusion: The comparison between groups showed that SFR may influence HRV parameters more than rest in workers in the clothing industry and possibly may contribute to the improvement of the stress experienced by them.

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1. Introduction

Work environment and workday may, on their own or together, act as stressors (Amaral et al., 2013). The stress response, also known as fight-or-flight response, is triggered by overactivation of the Hypothalamus - Pituitary - Adrenal (HPA) axis, an essential adaptive mechanism that enables the human body to maintain physiological stability in response to extrinsic or intrinsic stressors (Cozma et al., 2017). The stress response is important for human survival and its changes must last for seconds, minutes or hours.

However, if this response lasts for prolonged times, days, months or years, there will be a series of organic adaptations that will damage the body's homeostasis (Amaral et al., 2013).

Stressors, whether physical or psycho-emotional, can cause both HPA axis hyperactivation resulting in cortisol release, as well as activation of sympathetic pathways that stimulate the adrenal medulla to release adrenaline and noradrenaline (Papini and Pinto 2000). Sympathetic activation and the consequent release of these hormones directly impact on the activity of various body systems, including the cardiovascular system (Sacha 2014). Therefore, cardiac activity is considered one of the main indicators of the presence of stress, and the parameters of heart rate variability (HRV) are objective markers of such activity (Rauschel et al., 2015).

* Corresponding author. 1638, 37280-000, Candeias, MG, Brazil.

E-mail address: brunaluissilva@hotmail.com (A.P. Pernambuco).

HRV is defined as a set of normal and expected periodic and non-periodic oscillations of the heartbeat, which are related to the action of the Autonomic Nervous System (ANS) on the sinus node (Siqueira and Bojadsen 2006). So, HRV behavior and its indicators are indicative of autonomic activity and can be evaluated in different situations, as well as in individuals with different health states (Aubert et al., 2003). HRV is observed on the electrocardiogram as the interval between an R wave and the next R wave and corresponds to a heartbeat (Stein et al., 1994).

The most commonly evaluated HRV parameters are: High Frequency (HF) which is an indicator of the vagus nerve acting on the heart; Low Frequency (LF) which results from the joint action of the vagal and sympathetic components on the heart, with predominance of the sympathetic; Very Low Frequency (VLF) which is an indicator of sympathetic acting. The LF/HF relationship reflects the absolute and relative changes between the sympathetic and parasympathetic components of the ANS, characterizing the sympathovagal balance over the heart (Godoy et al., 2005).

Antagonistic to HPA and ANS axis activity, the Parasympathetic Autonomic Nervous System (PSNS) acts by minimizing stress responses, particularly during rest and relaxation moments (Schleip 2011). In this sense, it is believed that by stimulating parasympathetic activity in workers, it would be possible to minimize the stress response experienced by these individuals.

A possible way to modulate sympathetic autonomic activity and activate parasympathetic autonomic activity would be the application of low-intensity tactile stimuli (gentle touch) in order to stimulate the suboccipital cranial fascia and, consequently, the vagus nerve (Schleip 2011). The fascia is constituted by connective tissue, able to continuously integrate the structures of the human body (Bordoni and Zanier 2015). Suboccipital fascial release (SFR) technique may be able to activate the muscular fascia and consequently the dura mater (Pontell et al., 2013; Enix et al., 2014). In this sense, SFR could be a therapeutic alternative to stimulate PSNS activity. According to the literature, this technique may contribute to the release of endorphins and serotonin, neurotransmitters that have an important action in the psycho-emotional aspect and well-being (Santos and Severiano 2011).

During SFR, the suboccipital region is gently pressed and massaged. Its effects can be explained by three mechanisms: 1) Due to the anatomical proximity to the jugular foramen, point of passage of the vagus nerve (main nerve of the PSNS), it is believed that, by releasing the occipital tensions, it could also release the nerve pathway, decreasing the pressure in the endoneural compartment and thus improving vagal function (Liem, 2005; Schleip 2011); 2) This technique, by touching the muscular fascia, could be able to stimulate the dura mater via the myodural bridges (two fascial connections are described: one between the posterior minor rectus fascia and the dura mater) and another between the nuchal ligament and the dura mater, passing between C1 and C2). In the brain, the dura mater remains in the cerebellum tent and the pituitary tent - which surrounds the pituitary, thus influencing the HPA axis. In addition, the dura mater at the base of the skull is innervated by the vagus nerve (Bordoni and Marelli 2017; Enix et al., 2014; Pontell et al., 2013; Adeeb et al., 2012); 3) It has been proposed that soft and slow techniques have the potential to stimulate the parasympathetic autonomic nervous system (Olausson et al., 2008). The soft and slow techniques stimulate free nerve endings present in the fascia, which send information to the left insular cortex, stimulate the left hypothalamus and generate a parasympathetic response (Strigo and Craig 2016; Schleip et al., 2012).

In the literature, a non-mechanical rather than neurofasciogenic pathway is also described (Tozzi 2015), which could also explain the effects of parasympathetic stimulation. This model integrates nerve tissue with connective tissue, justifying a nonlinear response.

In this way, SFR, described by Olausson et al. (2008), would stimulate the free nerve endings in the skin that would carry the information to the posterior horn of the spinal cord by synapsing in lamina I. This pathway ascends to the superior centers through the spinothalamic tract to the insular cortex, following a pathway different from the proprioceptive pathways that they ascend through the pyramidal tract, which in turn send information to the proprioceptive areas of the brain. Hence the information activates the right insular cortex and stimulates the right hypothalamus with an increase in parasympathetic tone (Strigo and Craig 2016). This would result in a complex action between the afferent and efferent system reshaping the extracellular matrix by decreasing tension through sympathetic inhibition promoted by increased vagal tone (Bordoni and Marelli 2017).

According to the mentioned context, the objective of the study was to assess whether the SFR technique was able to influence HRV parameters in clothing industry workers.

2. Methodology

2.1. Ethical considerations

This study was initiated after approval by the Human Research Ethics Committee, under Opinion n°. 2.439.896. To participate, the volunteers signed an informed consent form (ICF). The research protocol was submitted to the Brazilian Registry of Clinical Trials (REBEC) Clinical Trials Registry and was approved according to number REBEC – RBR – 56yk9m.

3. Type of study

3.1. Two-arm randomized controlled trial

3.1.1. Participants

The non-probabilistic sample was formed by convenience. After active recruitment through contacts with clothing owners from a city in Minas Gerais, Brazil, three companies and 40 employees, of which 15 were enrolled in the untreated group (UG) and 25 enrolled in the treated group (TG).

Inclusion criteria were: clothing factory worker in a specific city in Minas Gerais, Brazil, female, aged between 20 and 50 years, with any educational level, working for 8 h a day, accepting to participate voluntarily in the research and score over 13 points in the Mini Mental State Examination (MMSE), if illiterate, and 19 points, if literate.

Criteria for non-inclusion were: employees who were pregnant or breastfeeding, who were using or who used any non-contraceptive drug in the past month, who did not achieve a minimum score in MMSE (13 points if illiterate or 18 points if literate); workers with history of autoimmune diseases; those who used steroidal anti-inflammatory drugs or anxiolytics in the last year; workers who had deficiencies in the upper and lower limbs (congenital or acquired); those with history of cardiovascular disease; those with endocrine disorders; workers with a history of migraine or recurrent headache; those with type 1 or 2 diabetes and those with more than one job.

Were excluded from the study those who did not feel well during the technique and those who for whatever reason wished to withdraw from the research, regardless of the phase of the study.

3.2. Randomization

The participants recruited for the study were workers from three different factories and were randomly divided into two groups. Randomization was performed by opaque, sealed and

numbered envelopes. With the use of software, a note was randomly generated with the allocation, it was inserted inside the envelopes. Each participant selected an envelope that contained the type of intervention that would be offered to her. Twenty-five participants made up the TG and 15 participants made up the UG. The allocation concealment was conducted, research group and participants were not aware of the type of intervention that would be offered to others.

The imbalance between the number of participants in the groups was generated by the randomization method, as at the beginning of the study the researchers prepared 60 opaque, sealed and numbered envelopes; however, at the presentation of the participants, only 40 volunteers were present. And due to the allocation concealment, the research team was unable to identify the imbalance and correct it, before the intervention period.

The randomization procedure resulted in two groups with similar characteristics at the baseline. The homogeneity between the groups can be seen in Table 1.

3.3. Data collection

Initially, all data were collected by an independent evaluator who was unaware of the study objectives and allocation attributed to participants. Even before randomization, the informed consent form was presented and the participants' signature collected. Those who agreed to participate in the survey completed the identification form and the Mini Mental State Examination (MMSE) for cognitive screening. This questionnaire was used for ethical reasons, in order to determine whether the participant had enough cognitive ability to choose to participate in the research. The MMSE also was used to characterize the sample and check for homogeneity of the participants in the pre-intervention period, as well as Work Stress Scale (WSS) and World Health Organization Disability Assessment Schedule (WHODAS 2.0).

3.4. Instruments

The MMSE is one of the most used and most studied tests in the world. The instrument allows the assessment of cognitive function and tracking of dementia. The original MMSE consists of two sections that measure cognitive functions. The first section contains items that evaluate orientation, memory and attention, totaling 21 points; the second measures naming ability, obedience to a verbal command, and the free writing of a sentence, in addition to a copy of a complex drawing (polygons), making up nine more points. The total score is 30 points and the higher the MMSE score the better the cognitive state of the participant. The following cutoff points are

used: 13 points for illiterate people, 18 points for literate people and 26 points for those who concluded high school (Folstein et al., 1975).

The WSS is an instrument used to measure perceived stress in the occupational environment. It can be applied in different work environments and in different occupations. It was initially proposed with 31 items. In 2004, Paschoal and Tamayo created a reduced version of the Work Stress Scale (WSS), which includes 23 items that represent the main organizational stressors and general psychological reactions. The expanded version has a Cronbach's alpha coefficient equivalent to 0.91, while the reduced version is 0.85. Despite the lower Cronbach's alpha coefficient, the 23-item scale was used in this study. Each item is scored from one to five, the maximum possible score is 115 points. The score should be interpreted as: below 46 points (low stress), 47 to 69 points (medium stress) and 70 to 115 points (high stress). In this sense, higher scores represent worse levels of worker stress (Paschoal and Tamayo 2004).

The WHODAS 2.0 is an assessment tool developed by the World Health Organization (WHO) that covers several items of the International Classification of Functioning, Disability and Health (CIF). WHODAS 2.0 provides the level of functionality/disability after analyzing six domains of life: cognition, mobility, self-care, interpersonal relationships, life activity and participation. The questionnaire consists of 36 questions and each one is scored from one (no difficulty) to five (extreme difficulty or is unable to do). The score is calculated by domains, the sum of all items that are contained in each domain is converted into a percentage value from 0 to 100. The higher the score, the greater the disability presented by the participant. The questionnaire presents the following percentage cutoff points: 0–4% (no difficulty) 5–24% (mild difficulty), 25–49% (moderate difficulty), 50–95% (severe difficulty), 96–100% (complete difficulty/does not). The score can be expressed for each domain and total score on the instrument (De Castro and Leite 2005).

For heart rate and HRV analysis, information was collected during the 5 min preceding the intervention and during the first 5 min after the end of the intervention, with the volunteers sitting quietly in a room next to the work site. The experiments were performed in the morning. Data were measured using a specific heart rate monitor (Polar Heart Rate Monitor V800), which was in the volunteers' lower third of the sternum. After recording the data with the use of the heart rate monitor, the data were transferred to the Polar Flow Sync program and later to the Kubios HRV Standard for HRV data generation. The data obtained were sent to the GraphPad Prism software database, v. 6.0. The following data were recorded: maximum heart rate (max HR), minimum heart rate (min HR), mean heart rate (mean HR), R-R interval average (RR average),

Table 1
Characterization of study participants at baseline (n = 40).

Parameters	UG (n = 15)	TG (n = 25)	P value
Age	36.53 ± 10.52 (30.71–42.36)	33.16 ± 9.87 (29.08–37.24)	0.31
Time in the company (months)	44.73 ± 43.36 (20.72–68.75)	22.00 ± 38.16 (6.24–37.75)	0.02*
MMSE	26.20 ± 2.65 (24.73–27.67)	25.44 ± 2.80 (24.28–26.60)	0.33
WSS	63.47 ± 9.05 (58.45–68.48)	57.24 ± 18.70 (49.52–64.96)	0.23
WHODAS 2.0			
Cognition	19.44 ± 13.05 (12.22–26.67)	27.66 ± 15.40 (21.31–34.02)	0.09
Mobility	6.61 ± 10.33 (0.88–12.33)	12.40 ± 14.37 (6.46–18.33)	0.10
Self-care	5.58 ± 16.80 (–3.72 to 14.89)	2.25 ± 5.67 (–0.09 to 4.59)	0.76
Interpersonal relationships	10.33 ± 9.72 (4.94–15.72)	12.60 ± 12.43 (7.47–17.73)	0.77
Activity	18.75 ± 22.35 (6.37–31.13)	21.63 ± 13.74 (15.96–27.31)	0.20
Participation	20.85 ± 14.96 (12.56–29.13)	23.00 ± 19.35 (15.02–30.99)	0.97
Total	13.32 ± 10.63 (7.44–19.21)	16.59 ± 8.91 (12.91–20.27)	0.17

Untreated Group (UG), Treated Group (TG), Work Stress Scale (WSS), World Health Organization Disability Assessment Schedule (WHODAS 2.0). Data are expressed as mean ± standard deviation and 95% confidence interval (95% CI). The analyses were performed using the independent T test. *p ≤ 0.05.

Very Low Frequency (VLF), RR intervals' standard deviation (STDRR), root mean square successive difference in RR intervals (RMSSD) Low Frequency (LF), High Frequency (HF), Low Frequency/High Frequency (LF/HF) intervals.

3.5. Intervention

SFR was applied only once to each of the TG participants. The intervention lasted 5 min and could supposedly influence certain physiological mechanisms and contribute to neurological or psychophysiological aspects (Schulz et al., 2012).

In this research the technique was actively performed by one of the researchers, skilled and trained for this. The technique used was adapted from the studies conducted by Figueiredo and Oliveira (2011). In the TG, the volunteer remained lying on a stretcher, supine, with hips and knees bent, feet flat, arms beside the body and eyes closed.

The researcher remained seated close to the patient's cephalic region, with her hands positioned in the patient's suboccipital region, between the occipital and the C1 vertebra (see Fig. 1A). The maneuvers used by the researcher included counterclockwise circular sliding and very slow shear techniques, with both hands in prayer position, with the palm up, relaxed and with the volunteer's head resting on the palms of the researcher.

This maneuver is indicated to induce relaxation, as it helps to improve blood and lymphatic flow by reaching both deep and superficial tissues (Rechtien et al., 2002). These movements are capable of generating a pressure that is transmitted to the underlying tissues by means of biotensegrity, a mechanism capable of harmonically and dynamically transmitting force (Levin and Martin 2012). Soft pressure, using the digital spine of the last four fingers of each hand, is a technique used to increase local circulation, stimulate nerve endings, release fascia and suboccipital tissues, and tone soft tissue regions (Kavanag 2006).

UG participants were also absent from their jobs during the day and remained lying down for the same time period and in the same position as the TG participants. However, the suboccipital fascial release technique was not applied to the UG participants (see Fig. 1B). The measurement protocol of the HRV parameters in the UG participants was identical to that adopted for the TG participants.

Regardless of the number of participants completing the study, intention-to-treat analysis would be used. There was no drop out during the study period and all participants were assessed within the original allocation group.

3.6. Statistical analysis

To verify the distribution of data, the Shapiro-Wilk test was used. Descriptive statistics used the average, standard deviation and 95% confidence interval (95% CI) for data with normal distribution, and median and interquartile interval (P25% - P75%) for data

with non-normal distribution. For within group analysis the paired T test or Wilcoxon test was used and for between group analysis the independent T test or the Mann-Whitney test, depending on the data distribution. All analyses were performed using GraphPad Prism v.6 software, with significance level adjusted to $\alpha = 0.05$ ($p \leq 0.05$).

4. Results

Forty women participated in the study, divided into two groups, untreated group (UG) and treated group (TG). Data with normal distribution were represented by the mean $\pm$ 95% confidence interval (95% CI) and data with abnormal distribution were presented as median $\pm$ interquartile interval (P25% - P75%).

In the baseline, the UG consisted of 15 women, aged 36.5 ± 10.5 years, with time in the company of 44.7 ± 43.4 months and who scored 26.2 ± 2.6 points in MMSE. The TG consisted of 25 women, age 33.1 ± 9.8 years, with time in the company of 22 ± 38.1 months. The scores obtained by both groups in the MMSE, WSS and WHODAS 2.0 (domains and total score) are shown in Table 1. Initial analysis showed similarity between the groups at baseline but showed that working time in the company was higher for the UG participants (see Table 1).

The results related to heart rate and HRV parameters are presented in Table 2. In the UG, there was no significant difference in any of the evaluated parameters, when comparing the data obtained in the first and second collection. In the TG, there were significant differences in the following parameters: LF/HF ($p \leq 0.01$), RR average ($p \leq 0.01$), HR average ($p \leq 0.01$), HR min. ($p = 0.02$) and HR max. ($p \leq 0.01$), when comparing data obtained before and after treatment by SFR.

Heart rate data and HRV parameters, together with between group analysis (TG and UG) before and after SFR treatment, are presented in Table 3 and expressed as median $\pm$ standard deviation (95% CI) or as median and interquartile interval. Regarding the time before treatment, no significant differences were observed in any of the evaluated parameters. In relation to the second moment, the parameter VLF and HR max. presented significant difference in relation to both groups, with values ($p = 0.04$) and ($p = 0.03$) respectively.

5. Discussion

This is a pioneering study that demonstrated that a manual therapy technique known as SFR is able to influence HRV parameters and minimize stress experienced by clothing industry workers, since significant differences were observed in the analyzed outcome in favor of the TG when compared to the UG.

The research was conducted with the participation of young adult women (18–35 years) and middle-aged women (36–55 years) (Petry, 2002), who already had professional experience in the function developed. The choice to include only women in the study was

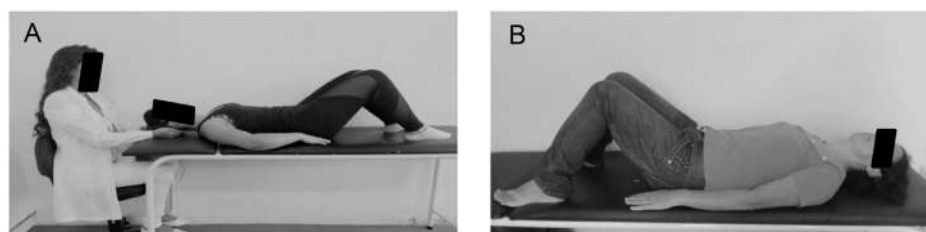


Fig. 1. 1A represents the positioning of the therapist and a treated group participant during the execution of the SFR technique. 1B represents the positioning of an untreated participant who remained lying down for 5 min and did not receive the SFR technique.

Table 2

Within groups analyses of HRV parameters before and after the intervention period.

Parameter	UG			TG		
	Before	After	P value	Before	After	P value
STDRR ^a	37.30 (29.30–52.80)	50.90 (36.40–73.90)	0.09	42.40 (32.25–46.75)	38.50 (28.90–57.45)	0.52
RMSSD ^a	26.20 (23.50–38.50)	43.50 (29.9–57)	0.12	33.30 (26.65–50.35)	35.1 (27.35–57.25)	0.51
VLF ^a	78 (37–164)	183 (54–354)	0.076	89 (47.50–173.50)	64 (41.50–129)	0.29
LF ^a	790 (435–1600)	709 (358–1937)	0.84	929 (394.50–1248)	626 (350.50–981)	0.21
HF ^a	318 (123–603)	719 (159–1040)	0.34	408 (145.50–593.50)	458 (254–1169)	0.31
LF/HF ^a	2.65 (1.36–5.19)	1.67 (0.74–4.54)	0.46	2.43 (1.37–3.24)	1.18 (0.85–2.32)	≤ 0.01*
Average RR ^b	815.40 ± 126.5 (745.40–885.50)	825.30 ± 128 (754.50–896.20)	0.62	795.1 ± 92.62 (756.90–833.40)	849.4 ± 97.44 (809.10–889.60)	≤ 0.01*
Average HR ^b	75.90 ± 10.90 (69.84–81.96)	75.27 ± 11.58 (68.86–81.68)	0.76	76.88 ± 8.69 (73.29 to 80.47)	71.89 ± 8.28 (68.47–75.31)	≤ 0.01*
Min. HR ^b	66.40 ± 8.82 (61.52–71.29)	65.46 ± 9.55 (60.17–70.75)	0.67	66.45 ± 7.52 (63.35–69.56)	63.85 ± 6.93 (60.99–66.71)	0.02*
Max. HR ^b	89.32 ± 17.65 (79.55–99.10)	104.20 ± 42.50 (80.62–127.70)	0.19	91.63 ± 11.64 (86.82–96.43)	83.59 ± 2.49 (78.44–88.74)	≤ 0.01*

^a Results expressed as median and interquartile interval (P25% – P75%).^b Results expressed as median ± standard deviation and 95% confidence interval (95% CI). Within group analysis was performed by Wilcoxon test or paired T test. STDRR (RR interval standard deviation), RMSSD (root mean square successive difference in ms), VLF (very low frequency power in ms²), LF (low frequency power in ms²), HF (high frequency power in ms²), LF/HF (ratio of LF to HF), Mean RR (mean RR intervals), Mean HR (average heart rate - bpm), Min. HR (minimum heart rate - bpm) and Max. HR (maximum frequency heart rate - bpm). RR interval is the interval between two heart beats. ***p** ≤ **0.05**.**Table 3**

Between group analysis of HRV parameters before and after the intervention period.

Parameter	Before			After		
	UG	TG	P value	UG	TG	P value
STDRR ^a	37.30 (29.30–52.80)	42.40 (32.25–46.75)	0.78	50.90 (36.40–73.90)	38.50 (28.90–57.45)	0.07
RMSSD ^a	26.20 (23.50–38.50)	33.30 (26.65–50.35)	0.27	43.50 (29.9–57)	35.1 (27.35–57.25)	0.43
VLF ^a	78 (37–164)	89 (47.50–173.50)	0.44	183 (54–354)	64 (41.50–129)	0.04*
LF ^a	790 (435–1600)	929 (394.50–1248)	0.86	709 (358–1937)	626 (350.50–981)	0.36
HF ^a	318 (123–603)	408 (145.50–593.50)	0.54	719 (159–1040)	458 (254–1169)	0.95
LF/HF ^a	2.65 (1.36–5.19)	2.43 (1.37–3.24)	0.55	1.67 (0.74–4.54)	1.18 (0.85–2.32)	0.56
Average RR ^b	815.40 ± 126.5 (745.40–885.50)	795.1 ± 92.62 (756.90–833.40)	0.56	825.30 ± 128 (754.50 to 896.20)	849.40 ± 97.44 (809.10 to 889.60)	0.50
Average HR ^b	75.90 ± 10.90 (69.84–81.96)	76.88 ± 8.69 (73.29–80.47)	0.75	75.27 ± 11.58 (68.86 to 81.68)	71.89 ± 8.28 (68.47–75.31)	0.28
Min. HR ^b	66.40 ± 8.82 (61.52–71.29)	66.45 ± 7.52 (63.35–69.56)	0.98	65.46 ± 9.55 (60.17–70.75)	63.85 ± 6.93 (60.99–66.71)	0.54
Max. HR ^b	89.32 ± 17.65 (79.55–99.10)	91.63 ± 11.64 (86.82–96.43)	0.62	104.20 ± 42.50 (80.62–127.70)	83.59 ± 2.49 (78.44–88.74)	0.03*

^a Results expressed as median and interquartile interval (P25% – P75%).^b Results expressed as median ± standard deviation and 95% confidence interval (95% CI). Between group analyses were performed using the Mann-Whitney or independent T test. STDRR (RR interval standard deviation), RMSSD (root mean square successive difference in ms), VLF (very low frequency power in ms²), LF (low frequency power in ms²), HF (high frequency power in ms²), LF/HF (ratio of LF to HF), Mean RR (mean RR intervals), Mean HR (average heart rate - bpm), Min. HR (minimum heart rate - bpm) and Max. HR (maximum frequency heart rate - bpm). RR interval is the interval between two heart beats. ***p** ≤ **0.05**.

due to the fact that the clothing industry historically employs more women (Silva et al., 2017) and the fact that female labor has been growing in various sectors of commerce and industry (Proni 2018). In addition, a sample of women alone would avoid gender biases of the participant (Heidari et al., 2017). The inclusion of participants aged between 20 and 50 years aimed to minimize bias related to the effects of climacteric. It is important to note that in Brazil the women begin menopause about 51.2 years old (Pedro et al., 2003). The non-inclusion of inexperienced participants is because the work environment and or the workday may function as a stress-generating mechanism and could interfere with the results (Amaral et al., 2013). After all, it is known that physical or psycho-emotional agents are capable of over activating the HPA axis and ANS, which in turn can alter HRV parameters (Papinie and Pinto 2000).

Although there were no significant differences between study groups at baseline regarding participants' age, MMSE, WSS and WHODAS scores, it was observed that participants of the UG had been working in the company longer than the TG participants. Two hypotheses should be considered in this regard. The first is that by working longer, and being exposed to stressors longer, UG participants would be experiencing higher levels of stress than TG participants. During the early phase of stress, increased HPA and ANS axis activity would result in increased release of cortisol, adrenaline, and norepinephrine (Sacha 2014). These hormones have pleiotropic effects and therefore interfere with the functioning of multiple organ systems, such as the cardiovascular one (Sacha 2014). However, the WSS scores and HRV parameters were

similarly in both groups, suggesting that stress levels were also similar.

The second hypothesis is that because they worked longer in the company and were exposed to stressors for a longer period of time, the participants of the UG were already in the stress phase known as burnout and no longer had the ability to respond to that type of stressor agent. Therefore, they would experience lower stress levels than those presented by the TG participants. The burnout phase is marked by hypocortisolemism and lower circulating adrenaline levels (Silva et al., 2018). In the burnout phase, besides the neuro-endocrine imbalance, there are also a series of psycho-emotional manifestations such as chronic anxiety, nervousness, mental exhaustion and depression (Amenábar 2006). Again, the absence of significant differences between groups in WSS scores and HRV parameters, suggests that the groups' stress levels were similar at baseline.

In this study, we chose to use cardiovascular parameters as variables of interest, as these are objective parameters that cannot be controlled voluntarily by the participant. Most studies that investigate stress among workers still use subjective instruments, such as questionnaires or interviews (Sousa et al., 2017). It should be noted that subjective instruments may suffer interference from the Hawthorne effect, placebo effect and other confounding factors (Plauto 2008). Objective parameters that are independent of the patient's perception, besides minimizing interpretation biases, strengthen the findings of the study (Gonçalves et al., 2011).

No other significant differences were found at baseline when

comparing the two groups of study. The absence of statistical significance in cardiovascular parameters before the experiment demonstrates the homogeneity of the sample and ensures the reliability of the within group and between group analysis. Within group analysis in the UG showed no significant change between the initial and final evaluation. In the TG, significant changes were observed in five of the evaluated parameters: low frequency/high frequency ratio (LF/HF), average RR intervals (Average RR), average heart rate (Average HR), minimum heart rate (Min. HR) and maximum heart rate (Max. HR). These findings together support the hypothesis that the SFR technique applied in the suboccipital region may be able to interfere with autonomic modulation and minimize the organic effects related to stress response. However, it should be pointed out that the changes in the HRV parameters observed in this investigation cannot necessarily be attributed to the fascial release technique used, as it could equally be due to the act of touching. In this sense the inclusion of an appropriate sham intervention would have been informative.

The heart, despite having its intrinsic innervation and therefore being able to regulate its own rhythm, also has its ANS and PSNS modulated functions (Powers and Howley 2000; Carter et al., 2003). Significant changes in important cardiovascular parameters are strong indications that the technique used was able to modulate sympathetic activity of the heart and stimulate parasympathetic activity. Heart rate regulation via ANS is done through norepinephrine which makes the sinoatrial node cell membrane more permeable to potassium ions, which leads to accelerated heart rate (Aubert et al., 2003). The parasympathetic influence on the heart is exerted via the vagus nerve and release of acetylcholine, a neurotransmitter that makes the pacemaker cell membrane less permeable and consequently reduces the heart rate (Aubert et al., 2003).

In this study we evidenced a significant increase in the average RR intervals. According to Kiss (2003) in situations where there is a predominance of parasympathetic activity over sympathetic activity in the heart, there is an increase in the interval between RR waves (Kiss 2003). In addition to this important finding, it was also shown that the TG participants had a significant reduction in the average heart rate, minimum heart rate, maximum heart rate, and low/high heart rate (LF/HF) ratio after the intervention. The LF/HF ratio, also called sympathovagal balance reflex, is an important marker of cardiac activity. High values of this index indicate a sympathetic predominance, while lower values indicate greater parasympathetic activity (Achten and Jeukendrup 2003). These results independently or associated demonstrate that parasympathetic activity in the heart was stimulated by the SFR in the suboccipital region.

It is important to mention that the effects observed on HRV parameters could be due to leaving the work environment, that is, exclusively due to the removal of stressors inherent in the workplace and even the 5-min relaxation period experienced by the study participants. However, no significant changes in cardiovascular parameters were evidenced in the untreated group. This group was also removed from the work environment and remained lying down for the same period as the treated individuals. Thus, it is believed that the SFR technique was able to stimulate PSNS activity on the heart.

It is important to mention that within group differences, although clinically important, cannot be attributed solely to the treatment offered, as it is practically impossible to control some confounding factors such as: placebo effect, Hawthorne effect, return to average, natural history, spontaneous improvement, among others (McCall et al., 2011). However, it should be noted that the outcome used was an objective marker, change of which is not dependent on the participant's opinion or perception regarding the intervention used.

In addition, a non-active control group (untreated group) was used, which allowed fairer interpretations of the results.

In the between group comparison, at the end of the study, it was observed that the VLF parameter presented significant difference between the studied groups, with lower values in the TG. VLF is a component still poorly studied and understood, and seems to be related to thermoregulatory cycles, renin activity and other hormonal cycles (Monteze 2014). VLF has been shown to decrease during a mental stress task and to take longer than other components to return to baseline (Usui and Nishida 2017). In this sense, the VLF values in the treated group would be expected to be similar or significantly higher than those found in the untreated group, which did not occur. In this study, measurements were performed immediately after the intervention ended and, as it is a slow recovery variable, such recovery may not have been detected by the methodology used. It should also be emphasized that this parameter is modulated by both ANS and PSNS and thus it is not possible to state whether SFR benefits the behavior of this variable (Tau 2004).

However, max. HR showed significantly lower values in favor of the TG when compared to the UG. It is believed that this fact is at least partially justified by the vagus nerve PSNS stimulation, with consequent release of acetylcholine and a reduction in the cardiac pacemaker cell membrane permeability, resulting in a reduction in HR and a concomitant reduction in max. HR (Aubert et al., 2003; Powers and Howley 2000; Carter et al., 2003).

The main limitations of the study are related to the small sample size, which may cause type II errors and the absence of a follow-up period, which could contribute to the understanding of long-term responses. Another important flaw of the study is that, although there is an untreated group, there was no group that received a sham treatment. This fact prevents us from claiming that the effects observed in TG were truly provided by the method used. In this sense, there is no way to say whether the technique used would be superior to any other type of manual stimulus in the occipital region, or even for the simple fact that someone holds the head of the participant who is lying down. It is believed that the addition of a placebo group, or even the use of a crossover trial with washout period, would be solutions to the problems identified. Future studies should address these limitations.

6. Conclusion

The findings of this study suggest that SFR may influence HRV parameters more than rest in workers in the clothing industry and possibly may contribute to the improvement of the stress experienced by them. The unprecedented character of this study prevents the confrontation of the data presented here. Studies with greater methodological power, designed to include a sham intervention group, and extended follow-up periods or even a or a crossover trial with a washout period should be conducted to clarify issues not addressed here.

Clinical relevance

- The SFR is superior to rest by influencing heart rate and HRV parameters in clothing industry workers;
- The SFR possibly influences the stress experienced by clothing industry workers.

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Credit author statement

BLS, LAO, CMC and APP designed and performed the experiments, performed the data analysis and wrote the manuscript and CQG, LSV and APP reviewed and edited the paper. All authors read and approved the final manuscript.

Declaration of competing interest

We declare that the authors Cristiano Queiroz Guimarães and Leonardo Sette Vieira own a school that specializes in the study of fascia and that the results of this study may be of economic interest to them. We emphasize, however, that the mentioned authors only had access to the results at the end of the manuscript writing phase and that no data was altered or manipulated in order to satisfy any interest.

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