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Pain–autonomic interaction after work-induced sleep restriction

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Background and purpose: Poor sleep is commonly associated with alterations in pain perception. However, there is a lack of studies that address work-associated sleep restriction (SR) and changes in non-nociceptive perception and autonomic responses after work-induced SR.

Methods: This study was performed with 19 medical students after a normal-sleep night (NS phase) and after a night shift at the local emergency room (SR phase). We performed clinical assessment, quantitative sensory testing for electrical and temperature sensation, RR interval analysis, and recorded sudomotor skin responses (SSRs).

Results: The total mean duration of sleep was 436 ± 18 min in the NS group and 120 ± 28 min in the SR group ($P < 0.001$). The anxiety scores were higher following the SR phase compared with those after the NS phase ($P < 0.01$). After SR, there was a decrease in heat-pain threshold, but neither warm nor electrical thresholds were affected. Following SR, subjects showed higher SSR amplitudes and an increased number of double responses at an interstimulus interval of 2 s. We also observed a moderate inverse correlation between heat-pain thresholds and SSR amplitude ($r = -0.46$; $P < 0.01$). However, there was no correlation between anxiety scores and SSR parameters.

Conclusions: The effects of SR in the context of work stress on pain are specific and appear unrelated to general changes in sensory perception. Hyperalgesia was associated with abnormal autonomic responses, but not with increased anxiety, which suggests an association between the nociceptive and autonomic nervous systems that is independent of the emotional state.

Introduction

Lack of sleep is strongly related to alterations in pain perception in normal subjects and frequently leads to hyperalgesia [1–7]. Sleep restriction (SR) can also affect the autonomic nervous system [8], which in turn has been implicated in C fiber sensitization [9–11] and pain perpetuation [12,13]. Indeed, autonomic nuclei are located at the brainstem and are close to the ascending activated reticular system that is involved in sleep processing [14]. Therefore, sympathetic activity may play a role in poor-sleep-induced pain. Surprisingly, although there are several studies analyzing the effect of sleep deprivation and stress on autonomic function [15–17], there are none that assess pain perception together with autonomic tests in sleep-restricted patients. Furthermore, most of the studies analyzing pain perception in sleep-restricted individuals used artificial methods of SR that are unrelated with real-life conditions.

In this study, we assessed sensory thresholds and autonomic function and correlated these data with clinical data gathered from 19 healthy subjects. We evaluated these subjects at two timepoints: after normal sleep (NS phase) and after SR phase (a laborious night of medical work in an emergency room).

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Methods

Nineteen healthy right-handed male volunteers (mean age, 24.3 years; range 22–30) participated in the experiment. All subjects were fifth-year medical students and had good sleep quality [Pittsburgh Sleep Quality Questionnaire Index (PSQQI) <5]. All subjects gave their written informed consent for this study, which was approved by the Ethical Committee of the Hospital de Clínicas de Porto Alegre, Brazil.

Exclusion criteria

We excluded subjects who fulfilled the following criteria: poor sleep (index >5); antidepressant use; analgesic use; caffeine or other drug use that could potentially affect the nociceptive or autonomic nervous system in the last 8 h, that is, beta blockers, tricyclic antidepressants, anticholinergics; the presence of any known disease potentially involving the autonomic nervous system, that is, diabetes mellitus, cerebrovascular diseases; and the presence of chronic pain complaints. Because mood disorders can affect pain perception and autonomic responses, we also excluded subjects with such conditions using previous history and the results of clinical baseline assessment.

Clinical assessment

Apart from demographic information (see Experimental procedure), to assess psychiatric symptoms, we conducted the Hamilton scale and the State-Trait Anxiety Inventory (STAI) adapted to the Brazilian language [18]. Finally, sleep status was assessed using the PSQQI [19]. After the night shift in the emergency room, we also asked about the duration of sleep during the previous working night.

Psychophysical assessment

Thermoalgesic stimuli

All thermoalgesic stimuli were applied using a Peltier-type contact rectangular thermode (Heat Pain Stimulator-1.1.10, Brazil) with a stimulating area of 30×30 mm^2 [20]. We determined the individual's warm- and heat-pain threshold using the method of limits and standardized procedures [21]. Subjects were instructed to press a button on the device as soon they felt any change in temperature. Thresholds were defined as the mean value of three stimuli separated by an interstimulus interval (ISI) of at least 40 s [22].

Electrical stimuli

Thresholds for perception of electric shocks were obtained using the method of levels [21], in which electrical pulses of 0.2-ms duration were delivered to the median nerve in the wrist at increasing intensities with ISIs of at least 5 s. Subjects were then asked to say 'yes' when they felt electrical non-painful and electrical painful shocks. We stopped the assessment when reproducible thresholds were achieved for each subject.

Autonomic assessment

Sudomotor skin responses (SSRs) were recorded using surface electrodes. The active electrode was attached to the palm and the reference electrode to the dorsum of the hand [23] on the side ipsilateral to the stimulus. To evaluate the recovery of SSR excitability, we applied double electrical stimuli separated by ISIs of 1, 2, and 3 s [24]. The intensity of the stimulus (in mA) for all trials was defined as the intensity of motor threshold, that is, when median nerve stimulation caused a slight twitch of the abductor pollicis brevis. The order of the specific ISIs was random. We performed three double stimuli for each ISI, thus 18 electrical single stimuli in total. To maintain full attention to each stimulus and to retrospectively calculate the habituation index of sensory perception (see Data reduction and statistical analysis), we asked the subjects to gauge the intensity of each electrical shock using a visual analogue scale (VAS) that ranged from 0 (no sensation) to 10 (worst imaginable pain).

Analysis of the RR interval was also conducted using superficial electrodes, but they were located to the chest (V5 precordial derivation) with continuous recording of the heart rate during two provocative maneuvers: deep inspiration–expiration movements (three full cycles) and standing up [25]. This procedure lasted approximately 10 min.

Experimental procedure

Initially, we conducted an interview to assess demographic (age, weight, and height) and clinical data, such as duration of sleep in the preceding night, sleep and psychological scales. Psychophysical and autonomic tests were then performed by two independent examiners (LD and PS, respectively). Prior to their assessments, both examiners were blinded to the results of the sleep status, clinical evaluations, and autonomic profiles. All subjects were assessed twice: once following a night of usual sleep (NS phase) and once following the night of SR phase. The order of evaluations (after normal or restricted sleep) was

balanced. For the SR session, we invited the students the morning after a 12-h duty in the local emergency room. The duties were always on Mondays, Tuesdays, or Wednesdays. On the chosen days, the duties were characterized to have approximately 20 visits every night (approximately 2 per hour). The medical students made the first evaluation (when it was not an emergency case) and discussed afterward with the attendant on duty. On average, every student evaluated approximately six patients each night shift.

All tests were performed in the morning before and after a 12-h night shift, from 7:00 pm to 7:00 am. Subjects woke up, with the assistance of an alarm clock, at 6:30–7:00 am. They arrived at the laboratory at approximately 7:30 am for either NS or SR assessments. The evaluations were performed in a quiet, semi-dark room, with an ambient temperature of 23–24°C. Subjects were always addressed by the same researcher (LD), who systematically read the instructions and explained the standardized experimental procedure using a previously published quantitative sensory testing (QST) protocol orientation [26] and adapted to the Brazilian Portuguese language [20].

Data reduction and statistical analysis

We calculated the mean and the standard deviation (SD) of all variables for all subjects. For the QST, we determined the mean threshold values for warm, heat, and electrical pain in each subject and calculated the grand mean and the SD for each group (NS and SR groups).

For the SSR, we measured the amplitude of the two responses elicited by the double stimuli at different ISIs (Fig. 1). The recovery of excitability was calculated as the percentage of the amplitude of the second response compared with the first response. For example, first and second responses of 2 and 1 mV amplitude, respectively, indicate a recovery of 50%. We also averaged nine individual first responses to measure the mean onset latency and the mean amplitude of SSRs. To assess the habituation of electrical stimulus perception, we defined the habituation index of repetitive electrical stimuli (el-HI). This index was calculated as the VAS for perception of the very first electrical stimulus minus the VAS of the last electrical stimulus out of 18 (three double stimuli for each of the three ISIs). Therefore, reduced habituation was indicated by low el-HI values. For the heart rate variability assessment, we calculated the maximum RR interval of expiration minus the minimum RR interval of inspiration (E–I) and divided this value by the minimum RR interval of inspiration (E/I) [25]. The mean E–I and E/I during three successive breathing cycles were calculated. We also measured the longest RR interval at approximately the 30th beat after standing and divided it by the shortest RR interval at approximately the 15th beat after standing (30:15). Figure 2 displays representative EMG recordings of cardiac beats.

We focused our statistical analyses before (NS group) and after night-shift (work-induced SR group). Depending on whether the data were Gaussian or non-Gaussian distributed, paired Student's *t*-tests or Mann–Whitney *U*-tests were used to analyze possible

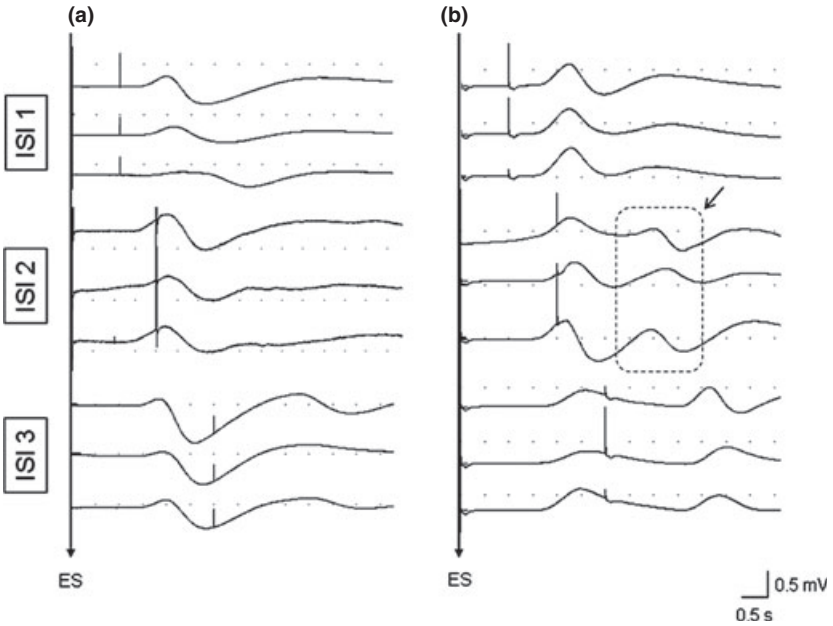

Figure 1 Sympathetic skin responses induced by double electrical stimuli (ES). Note the presence of double responses in an interstimulus interval of 2 s in a subject after (b), but not before (a), sleep restriction.

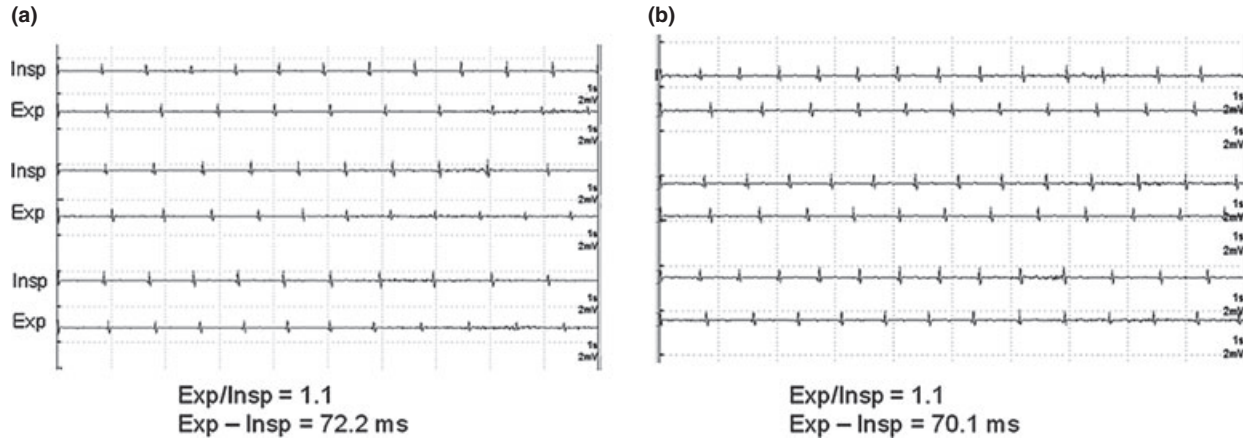

Figure 2 Illustrative RR interval recordings during inspiration/expiration maneuvers in a subject before (a) and after (b) sleep restriction.

differences between pre- and post-SR. We also performed logarithmic transformation of the SSR when necessary. Categorical variables were analyzed using a chi-squared test. Correlation analyses were performed using the Pearson or Spearman tests for correlation of thresholds and autonomic data with clinical characteristics. Finally, multiple linear regression models were performed using heat-pain thresholds as a dependent variable. An interaction term for autonomics versus anxiety was used to assess a possible effect of anxiety on the relationship between autonomic responses and thermal perception. A value of $P < 0.05$ was considered statistically significant.

Results

Initially, a total of 21 subjects were selected for our study. However, after careful evaluation, there were two exclusions: one because the subject was using beta-blocker for physiological tremor and one due to a PSQQI that was higher than 5. Thus, 19 subjects were assessed completely. The demographic and clinical characteristics of the subjects are summarized in Table 1.

Table 1 Clinical and demographic baseline characteristics

Variables	Values
Age (years)	24.3 ± 2.1
Body mass index	25.4 ± 2.0
Hamilton Scale score	3.8 ± 2.6
Pittsburgh Sleep Quality Questionnaire Index	3.6 ± 1.0
STAI score	16.9 ± 3.6
Night-shift sleep duration (min)	120 ± 28

STAI, State-Trait Anxiety Inventory.
Mean ± standard deviation ($n = 19$).

Following SR, heat pain, but neither warm nor electrical threshold, was significantly lower compared with the NS group (Fig. 3). In contrast, STAI scores were higher in SR subjects compared with NS subjects. However, there was no correlation between STAI and sensory thresholds or STAI and autonomic parameters ($P > 0.05$ for all comparisons). Although electrical thresholds were similar between the two conditions, the habituation of electrical perception, measured by VAS, was significantly lower in the SR group compared with the NS group, that is, sleep-restricted subjects did not show as much reduction in perception after repetitive stimuli as NS subjects did.

The amplitude of the SSR was significantly greater in sleep-restricted subjects compared with NS subjects. Similarly, double responses at ISI of 2 s were more prevalent in subjects with SR compared with the NS group. Double responses at ISI of 1 s and ISI of 3 s were similar between groups. Table 2 summarizes the results on psychophysical and electrophysiological analyses. SSR amplitudes correlated significantly with heat-pain thresholds following SR ($r = -0.46$) as seen in Fig. 4. Indeed, subjects with double responses at ISI of 2 s had lower heat-pain thresholds compared with those without double responses at the same ISI (41°C vs. 45°C; Mann–Whitney, $P < 0.001$). There was no correlation between thermal thresholds or autonomic responses and STAI scores ($P > 0.1$ for all correlations). Additionally, following SR, a multivariate linear regression showed a positive correlation between heat-pain perception and autonomic responses, but not with anxiety levels (see Table 3). On the other hand, no differences were found in heart rate variability between the two groups.

Finally, there was no correlation between sensory thresholds or autonomic responses and any of the

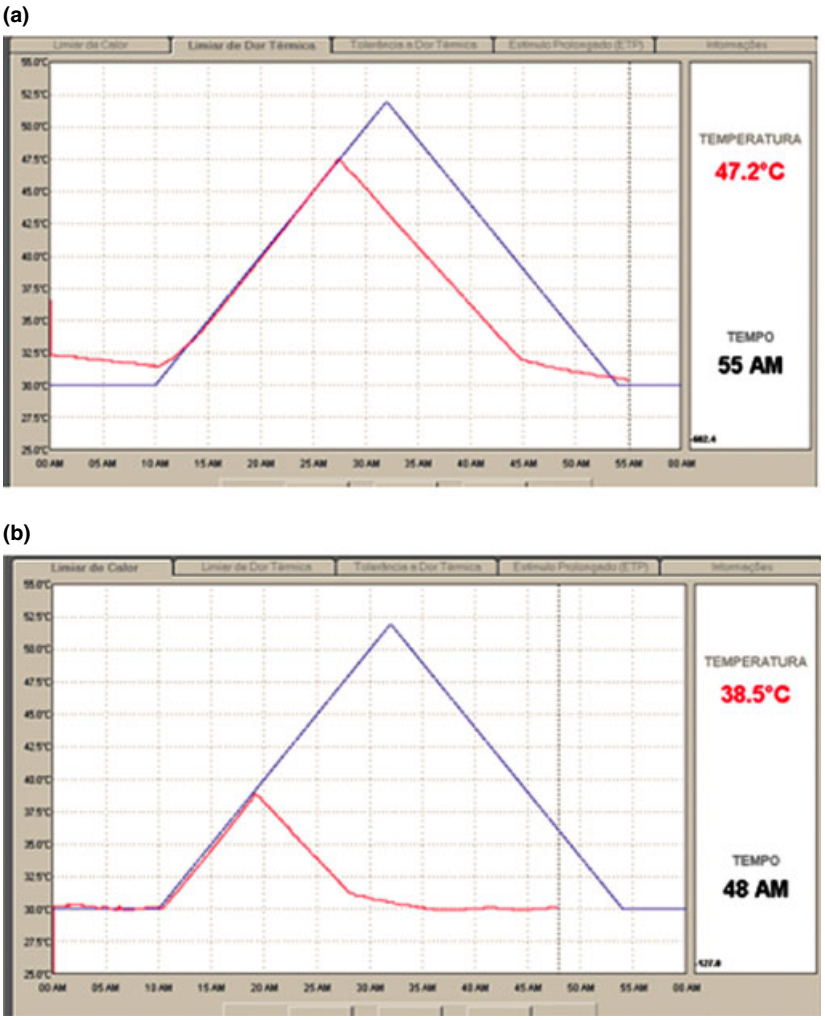

Figure 3 Heat-pain thresholds from the same subject. Note the presence of lower thresholds obtained after sleep restriction (b) in comparison with those obtained before (a) sleep restriction.

clinical or demographic variables apart from STAI scores ($P > 0.05$ for all correlations).

Discussion

Our study has three main findings: (i) heat pain but not warm or electrical thresholds were lowered by work-induced SR, indicating a specificity of the sensory system in reacting to a stressful situation; (ii) skin autonomic responses and anxiety levels were both increased after work-induced SR, suggesting an enhanced arousal reaction in response to acute stress; (iii) work-induced hyperalgesia was associated only with skin autonomic activity and not with anxiety levels, suggesting a role of autonomic function in pain perception that does not depend on the emotional state measured by STAI scores.

Sleep restriction is itself a stressful condition, but to date no studies assessing together sensory thresholds and autonomic function after work-induced SR have been performed. When studied separately, SR usually tends to increase sensory perception [2], whereas stress tends to decrease it [27]. In our study, we aimed to combine stressful work with SR to replicate a common real-life situation. We found significant differences in heat-pain thresholds after only 1 day of SR, which is different from other authors who used more prolonged or elaborate SR paradigms, such as 3 days of SR, selective REM, or stage 4 sleep deprivation [1,7,28–30]. This discrepancy can be explained by the strong potentiating influence of stressful work on the effects of SR on pain perception and autonomic function, as we observed here.

But why does SR in the context of work stress lead to reduce thresholds? One of the reasons underlying such pain predisposition in sleep-deprived subjects may be related to the finding of an impaired endogenous pain-inhibitory function that is associated with disturbances of sleep continuity [5,31]. This phenomenon can be related to dysfunctional serotonergic

Table 2 Psychophysical and neurophysiological data after normal sleep (NS) and sleep restriction (SR)

Variables	NS	SR	*P*
Clinical data			
Sleep duration (min)	436 ± 18	120 ± 28	<0.001*
STAI score	18.9 ± 3.5	22.9 ± 4.3	0.01*
Psychophysical data			
Electrical non-painful (mA)	2.8 ± 0.8	2.6 ± 0.6	0.1
Electrical painful (mA)	27.1 ± 9.7	28.0 ± 15	0.9
Electrical habituation index	4.1 ± 1.1	1.1 ± 0.7	0.02*
Warm (°C)	34.5 ± 2.2	32.1 ± 1.3	0.4
Heat pain (°C)	45.7 ± 1.1	41.1 ± 0.9	0.002*
Autonomic data			
SSR latency (ms)	1.5 ± 0.2	1.4 ± 0.3	0.2
SSR amplitude (mV)	3.8 ± 2.6	5.2 ± 3.8	0.03*
Double electrical stimuli (ISI of 2 s)	2/19	10/19	0.01*
Excitability recovery (%)	56.6 ± 32.2	63.7 ± 46.4	0.3
Expiration–inspiration	1.4 ± 0.2	1.3 ± 0.2	0.1
Expiration – inspiration (ms)	73.8 ± 26.3	70.1 ± 33.5	0.4
Orthostasis – 30:15	1.2 ± 0.2	1.1 ± 0.1	0.5

STAI, State-Trait Anxiety Inventory; SSR, sympathetic skin response; ISI, interstimulus interval.

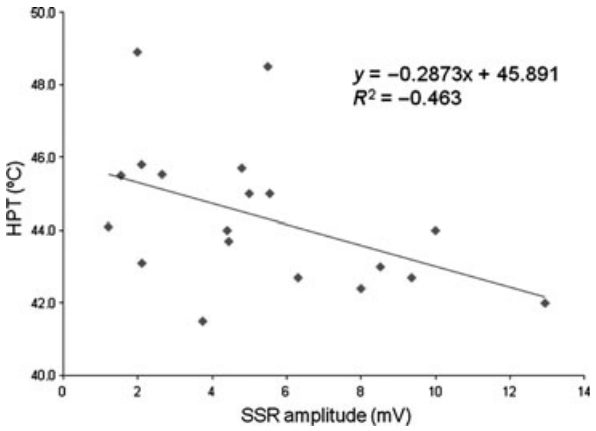

Figure 4 Correlation between heat-pain thresholds (°C) and amplitude of sympathetic skin responses (mV). [Correction added on 14 November 2012, after first online publication: the unit for amplitude of sympathetic skin responses was changed from mA to mV.]

neurons in the brainstem [32], decreased opioid function [33], or cytokine sensitization [34,35]. Habituation to pain is associated with increased activity in the endogenous pain-inhibitory system [36]. After SR, subjects habituated less to repetitive stimuli compared with subjects after normal sleep. Consequently, lower habituation may indicate transient antinociceptive dysfunction following SR, and this might have contributed to the lower thermal thresholds.

Table 3 Multivariate linear regression of the interaction between heat-pain perception, autonomic responses and anxiety after sleep restriction, using 'heat-pain perception' as a dependent variable

	β	*T*	*P*	95% CI
STAI score	−0.03	−0.13	0.89	−0.25 to −0.22
SSR amplitude (mV)	−0.55	−2.60	0.01*	−0.65 to −0.07

STAI, State-Trait Anxiety Inventory; SSR, sympathetic skin response; CI, confidence interval.
Significant correlation between pain perception and autonomic response, but not between pain perception and anxiety.
Dependent variable: heat-pain threshold (°C).

In our subjects, we expected that other forms of sensory stimuli would behave in the same way as the heat-pain sensation (low thresholds) after SR, but no differences were found in responses to warm or electrical stimuli between subjects with normal or restricted sleep (Table 2). This is consistent with the findings of other authors [37] and can be explained by the existence of a top-down regulating system that is specific for pain, but not for other sensory volleys. Indeed, functional neuroimaging studies have shown that the anterior cingulate cortex, the final nucleus of the medial pain system, is activated strongly by noxious heat and noxious cold, but not by innocuous stimuli [38].

We did not observe any changes in heart rate variability following SR. This finding is consistent with those of other authors [16] and indicates a decreased vulnerability of the cardio-autonomic system to acute stress. Indeed, this system is also less affected than skin sympathetic responses in diabetic patients with subclinical dysautonomia [39]. However, recent studies have found significant changes in heart rate variability after SR using power spectrum analysis [40,41]. Therefore, our methods are probably less sensitive than other physiological techniques to detect cardiovascular changes in sleep-restricted subjects.

Similar to Zhong *et al*. [42], we detected higher sympathetic activity in sleep-deprived subjects, which could be explained by the activation of brainstem noradrenergic neurons during stressful conditions [8]. We also found an association between increased autonomic activity and decreased pain thresholds in the palms following SR, which has biological plausibility. Nociceptive- and autonomic-regulatory regions of the central nervous system often respond to the same type of somatic or visceral inputs, receive convergent nociceptive and viscero-sensory information, and contain groups of neurons that initiate autonomic, antinociceptive, and behavioral responses to noxious stimuli [43]. This may explain the significant correlation between pain perception and autonomic responses that we observed in our subjects, detected only after

SR. Recently, we also found decreased thermal thresholds in hyperhidrotic patients after thoracic sympathectomy [44]. In fact, our subjects after SR might have a similar type of nociceptive–autonomic interaction seen in patients with palmar hyperhidrosis (low sensory thresholds and high autonomic activity). Therefore, alpha-adrenoceptors might have been triggered by hyperactive sympathetic post-ganglionic axons and consequently excited primary afferent axons in the 'nociceptive pathway' [9,10]. In conclusion, sleep-deprived subjects might actually have a dysfunction in the autonomic centers of the brainstem that are responsible both for the inhibition of sensory perception and for peripheral autonomic activity. This hypothesis would explain lower thermal thresholds and higher autonomic responses of our sleep-restricted subjects compared with normal subjects after normal sleep.

Sleep restriction cannot be considered a highly stressful condition because it is known that during such conditions, for example a soldier in a battle, higher, not lower, pain thresholds are to be expected [27]. In contrast, our subjects showed lower thresholds. Therefore, SR can be considered a distinctive form of stress. Alternatively, other lines of evidence can explain this finding. First, there may be different phases of pain sensory perception during stress adaptation [43]. It is conceivable that during acute stress stages, pain thresholds elevate and subsequently fall. However, the hypoalgesic effects of stress with respect to sleep may be more gradual, such as occurs in the opioid-mediated analgesia evoked from stimulation of the ventrocaudal periaqueductal gray matter [45]. Secondly, long-lasting stressful situations and anxiety contribute to pain amplification [46]. Thirdly, it is possible that the body changes the individual pain threshold to preserve excitation of the autonomic system and to protect against further pain. Finally, another possibility is that our stress paradigm was not sufficiently stressful to activate the endogenous antinociceptive system and consequently not able to increase pain thresholds. Indeed, salivary cortisol was unaffected by one-night sleep deprivation in healthy subjects [16].

Several methodological issues related to the design of this study must be addressed. First, we did not perform EEGs to characterize the lack of sleep and specifically for stage 4 sleep deprivation and relied only on the subjects' perception about their sleep. However, we preferred to choose an experimental design closer to the patient's reality of an exhaustive work shift together with SR. This paradigm was also chosen to maximize data gathered from such a small sample size. Secondly, it is possible that SR could have increased thermal thresholds secondary to lower reaction time to pressing the QST device's button [47]. However, if this was the case, higher thresholds and lower arousal responses would have been expected.

In conclusion, this study extends previous findings and shows that SR induced by a laborious night of medical work in an emergency room was associated with lower heat-pain thresholds and higher autonomic activity in healthy subjects. These data indicate a specificity of the sensory system in reacting to a stressful situation and to the active role of the autonomic nervous system in pain processing at least in this experimental condition.

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Disclosure of conflict of interest

The authors declare no financial or other conflict of interests.

References

1. Onen SH, Alloui A, Gross A, Eschallier A, Dubray C. The effects of total sleep deprivation, selective sleep interruption and sleep recovery on pain tolerance thresholds in healthy subjects. *J Sleep Res* 2001; **10:** 35–42.
2. Kundermann B, Krieg JC, Schreiber W, Lautenbacher S. The effect of sleep deprivation on pain. *Pain Res Manag* 2004; **9:** 25–32.
3. Haack M, Mullington JM. Sustained sleep restriction reduces emotional and physical well-being. *Pain* 2005; **119:** 56–64.
4. Roehrs T, Hyde M, Blaisdell B, Greenwald M, Roth T. Sleep loss and REM sleep loss are hyperalgesic. *Sleep* 2006; **29:** 145–151.
5. Smith MT, Edwards RR, McCann UD, Haythornthwaite JA. The effects of sleep deprivation on pain inhibition and spontaneous pain in women. *Sleep* 2007; **30:** 494–505.
6. Tiede W, Magerl W, Baumgärtner U, Durrer B, Ehlert U, Treede RD. Sleep restriction attenuates amplitudes and attentional modulation of pain-related evoked potentials, but augments pain ratings in healthy volunteers. *Pain* 2010; **148:** 36–42.
7. Azevedo E, Manzano GM, Silva A, Martins R, Andersen ML, Tufik S. The effects of total and REM sleep deprivation on laser-evoked potential threshold and pain perception. *Pain* 2011; **152:** 2052–2058.

8. Meerlo P, Sgoifo A, Suchecki D. Restricted and disrupted sleep: effects on autonomic function, neuroendocrine stress systems and stress responsivity. *Sleep Med Rev* 2008; **12:** 197–210.
9. Levine JD, Taiwo YO, Collins SD, Tam JK. Noradrenaline hyperalgesia is mediated through interaction with sympathetic postganglionic neurone terminals rather than activation of primary afferent nociceptors. *Nature* 1986; **323:** 158–160.
10. McLachlan EM, Jänig W, Devor M, Michaelis M. Peripheral nerve injury triggers noradrenergic sprouting within dorsal root ganglia. *Nature* 1993; **363:** 543–546.
11. Schlereth T, Brosda N, Birklein F. Spreading of sudomotor axon reflexes in human skin. *Neurology* 2005; **64:** 1417–1421.
12. Benarroch E. Pain-autonomic interactions: a selective review. *Clin Auton Res* 2001; **11:** 343–349.
13. Macefield VG. Developments in autonomic research: a review of the latest literature. *Clin Auton Res* 2009; **19:** 193–196.
14. Benarroch EE. Brainstem respiratory chemosensitivity: new insights and clinical implications. *Neurology* 2007; **68:** 2140–2143.
15. Sayk F, Teckentrup C, Becker C, *et al*. Effects of selective slow-wave sleep deprivation on nocturnal blood pressure dipping and daytime blood pressure regulation. *Am J Physiol Regul Integr Comp Physiol* 2010; **298:** R191–R197.
16. Pagani M, Pizzinelli P, Traon AP, *et al*. Hemodynamic, autonomic and baroreflex changes after one night sleep deprivation in healthy volunteers. *Auton Neurosci* 2009; **145:** 76–80.
17. Vaara J, Kyröläinen H, Koivu M, Tulppo M, Finni T. The effect of 60-h sleep deprivation on cardiovascular regulation and body temperature. *Eur J Appl Physiol* 2009; **105:** 439–444.
18. Kaipper MB, Chachamovich E, Hidalgo MP, Torres IL, Caumo W. Evaluation of the structure of Brazilian State-Trait Anxiety Inventory using a Rasch psychometric approach. *J Psychosom Res* 2010; **68:** 223–233.
19. Buysse DJ, Reynolds CF 3rd, Monk TH, Berman SR, Kupfer DJ. The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. *Psychiatry Res* 1989; **28:** 193–213.
20. Schestatsky P, Stefani LC, Sanches PR, *et al*. Validation of a Brazilian quantitative sensory testing (QST) device for the diagnosis of small fiber neuropathies. *Arq Neuropsiquiatr* 2011; **69:** 943–948.
21. Chong PS, Cros DP. Technology literature review: quantitative sensory testing. *Muscle Nerve* 2004; **29:** 734–747.
22. Schestatsky P, Algaba R, Pérez D, *et al*. Transient decrease of sensory perception after thermoalgesic stimuli for quantitative sensory testing. *Muscle Nerve* 2007; **36:** 466–470.
23. Claus D, Schondorf R. Sympathetic skin response. The International Federation of Clinical Neurophysiology. *Electroencephalogr Clin Neurophysiol Suppl* 1999; **52:** 277–282.
24. Manca D, Valls-Solé J, Callejas MA. Excitability recovery curve of the sympathetic skin response in healthy volunteers and patients with palmar hyperhidrosis. *Clin Neurophysiol* 2000; **111:** 1767–1770.
25. Baron R, Ewing DJ. Heart rate variability. The International Federation of Clinical Neurophysiology. *Electroencephalogr Clin Neurophysiol Suppl* 1999; **52:** 283–286.
26. Rolke R, Baron R, Maier C, *et al*. Quantitative sensory testing in the German Research Network on Neuropathic Pain (DFNS): standardized protocol and reference values. *Pain* 2006; **123:** 231–243.
27. Ford GK, Finn DP. Clinical correlates of stress-induced analgesia: evidence from pharmacological studies. *Pain* 2008; **140:** 3–7.
28. Older SA, Battafarano DF, Danning CL, *et al*. The effects of delta wave sleep interruption on pain thresholds and fibromyalgia-like symptoms in healthy subjects; correlations with insulin-like growth factor I. *J Rheumatol* 1998; **25:** 1180–1186.
29. Lentz MJ, Landis CA, Rothermel J, Shaver JL. Effects of selective slow wave sleep disruption on musculoskeletal pain and fatigue in middle aged women. *J Rheumatol* 1999; **26:** 1586–1592.
30. Arima T, Svensson P, Rasmussen C, Nielsen KD, Drewes AM, Arendt-Nielsen L. The relationship between selective sleep deprivation, nocturnal jaw-muscle activity and pain in healthy men. *J Oral Rehabil* 2001; **28:** 140–148.
31. Lautenbacher S, Kundermann B, Krieg JC. Sleep deprivation and pain perception. *Sleep Med Rev* 2006; **10:** 357–369.
32. Asikainen M, Toppila J, Alanko L, Ward DJ, Stenberg D, Porkka-Heiskanen T. Sleep deprivation increases brain serotonin turnover in the rat. *NeuroReport* 1997; **8:** 1577–1582.
33. Fadda P, Tortorella A, Fratta W. Sleep deprivation decreases mu and delta opioid receptor binding in the rat limbic system. *Neurosci Lett* 1991; **129:** 315–317.
34. Meier-Ewert HK, Ridker PM, Rifai N, *et al*. Effect of sleep loss on C-reactive protein, an inflammatory marker of cardiovascular risk. *J Am Coll Cardiol* 2004; **43:** 678–683.
35. Haack M, Lee E, Cohen DA, Mullington JM. Activation of the prostaglandin system in response to sleep loss in healthy humans: potential mediator of increased spontaneous pain. *Pain* 2009; **145:** 136–141.
36. Bingel U, Herken W, Teutsch S, May A. Habituation to painful stimulation involves the antinociceptive system – a 1-year follow-up of 10 participants. *Pain* 2008; **140:** 393–394.
37. Kundermann B, Spernal J, Huber MT, Krieg JC, Lautenbacher S. Sleep deprivation affects thermal pain thresholds but not somatosensory thresholds in healthy volunteers. *Psychosom Med* 2004; **66:** 932–937.
38. Lenz FA, Rios M, Zirh A, Chau D, Krauss G, Lesser RP. Painful stimuli evoke potentials recorded over the human anterior cingulate gyrus. *J Neurophysiol* 1998; **79:** 2231–2234.
39. Isak B, Oflazoglu B, Tanridag T, Yitmen I, Us O. Evaluation of peripheral and autonomic neuropathy among patients with newly diagnosed impaired glucose tolerance. *Diabetes Metab Res Rev* 2008; **24:** 563–569.
40. Chua EC, Tan WQ, Yeo SC, *et al*. Heart rate variability can be used to estimate sleepiness-related decrements in psychomotor vigilance during total sleep deprivation. *Sleep* 2012; **35:** 325–334.
41. Dettoni JL, Consolim-Colombo FM, Drager LF, *et al*. Cardiovascular effects of partial sleep deprivation in healthy volunteers. *J Appl Physiol* 2012; **113:** 232–236.

42. Zhong X, Hilton HJ, Gates GJ, *et al*. Increased sympathetic and decreased parasympathetic cardiovascular modulation in normal humans with acute sleep deprivation. *J Appl Physiol* 2005; **98:** 2024–2032.
43. Fechir M, Schlereth T, Purat T, *et al*. Patterns of sympathetic responses induced by different stress tasks. *Open Neurol J* 2008; **2:** 25–31.
44. Schestatsky P, Callejas MA, Valls-Solé J. Abnormal modulation of electrodermal activity by thermoalgesic stimuli in patients with primary palmar hyperhidrosis. *J Neurol Neurosurg Psychiatry* 2011; **82:** 92–96.
45. Lovick TA. Integrated activity of cardiovascular and pain regulatory systems: role in adaptive behavioural responses. *Prog Neurobiol* 1993; **40:** 631–644.
46. Rhudy JL, Meagher MW. Fear and anxiety: divergent effects on human pain thresholds. *Pain* 2000; **84:** 65–75.
47. Bougard C, Espié S, Larnaudie B, Moussay S, Davenne D. Effects of time of day and sleep deprivation on motorcycle-driving performance. *PLoS One* 2012; **7:** e39735.