

## Heart rate activation during spontaneous arousals from sleep: effect of sleep deprivation

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Accepted 4 June 2004

Available online 22 July 2004

### Abstract

**Objective:** Arousal (AR) from sleep is associated with an autonomic reflex activation raising blood pressure and heart rate (HR). Recent studies indicate that sleep deprivation may affect the autonomic system, contributing to high vascular risk. Since in sleep disorders a sleep fragmentation and a partial sleep deprivation occurs, it could be suggested that the cardiovascular effects observed at AR from sleep might be physiologically affected when associated with sleep deprivation. The aim of the study was to examine the effect of sleep deprivation on cardiac arousal response in healthy subjects.

**Methods:** Seven healthy male subjects participated in a 64 h sleep deprivation protocol. Arousals were classified into four groups, i.e.  $>3 < 6$  s,  $>6 < 10$  s,  $>10 < 15$  s and  $>15$  s, according to their duration. Pre-AR HR values were measured during 10 beats preceding the AR onset, and the event-related HR fluctuations were calculated during the 20 beats following AR onset. As an index of cardiac activation, the ratio of highest HR in the post-AR period over the lowest recorded before AR (HR ratio) was calculated.

**Results:** For AR lasting less than 10 s, the occurrence of AR induces typical HR oscillations in a bimodal pattern, tachycardia followed by bradycardia. For AR lasting more than 10 s, i.e. awakenings, the pattern was unimodal with a more marked and sustained HR rise. The HR response was consistently similar across nights, during NREM and REM sleep, without difference between conditions.

**Conclusions:** Overall, total sleep deprivation appeared to have no substantial effect on cardiac response to spontaneous arousals and awakenings from sleep in healthy subjects. Further studies are needed to clarify the role of chronic sleep deprivation on cardiovascular risk in patients with sleep disorders.

**Significance:** In healthy subjects acute prolonged sleep deprivation does not affect the cardiac response to arousal.

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**Keywords:** Arousal; Awakening; Sleep deprivation; Heart rate; Sleep state; Autonomic system

### 1. Introduction

Several studies estimating autonomic activity in the different states of the wake–sleep cycle have produced findings suggesting reduced sympathetic activity and increased vagal tone during sleep (Bonnet and Arand, 1997; Mancina, 1993; Somers et al., 1993; Umali et al., 2000). These changes induce a nocturnal decline in heart rate (HR) and blood pressure (Snyder et al., 1964; Van de Borne

et al., 1994) following homeostatic and circadian influences (Burgess et al., 1997, 2001; Umali et al., 2000). Therefore, extended hours of wakefulness may have direct negative effects on the cardiovascular system contributing to increased cardiovascular risk (Tofler et al., 1990).

The interaction between extended waking and variations in autonomic and hormonal systems has been the subject of a number of studies (Chen, 1991; Vgontzas et al., 1999), but the cardiovascular changes occurring during both extended wakefulness and subsequent recovery sleep has not been fully documented. Previous investigations examining the daytime effects have shown controversial results, sleep

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deprivation inducing either an increase (Lusardi et al., 1999; Ogawa et al., 2003), a decrease (Fiorica et al., 1968) or no change (Kato et al., 2000) in blood pressure, HR and sympathetic drive. Only a few studies have examined the effect of partial or total sleep deprivation on the nocturnal cardiovascular system. Delamont and coworkers (Delamont et al., 1998) using a beat-to-beat index of cardiac parasympathetic activity, found a rise in cardiac vagal tone in the first 90 min of recovery sleep related to the amount of sleep pressure, as indicated by electroencephalographic (EEG) slow wave activity. The rise in vagal tonus was greater when sleep deprivation was prolonged by 30 h and associated with a fall in sympathetic drive (Holmes et al., 2002). These changes in autonomic activity during both wakefulness and subsequent sleep could provide protective and recuperative values (Holmes et al., 2002) limiting, therefore, the negative effects of prolonged wakefulness.

It is well documented that arousals from sleep, either spontaneous (Sforza et al., 2000), associated with the cyclic alternating pattern (Ferri et al., 2000) or induced by environmental noise exposure (Carter et al., 2002; Davies et al., 1993), are associated with a substantial cardiovascular activation, similar to that described during obstructive sleep apnoea (Shepard, 1989) and periodic leg movement (Sforza et al., 1999, 2002). Since in such conditions a partial sleep deprivation also occurs, it could be suggested that the cardiovascular effects of arousal from sleep might be affected by the associated partial sleep deprivation. The effect of sleep deprivation on cardiac autonomic response to arousal from sleep has not been previously investigated. Therefore, the aim of the present study was to investigate to what extent sleep deprivation influences the cardiac response occurring during spontaneous arousal from sleep. If a down regulation of cardiac autonomic activity is present during extended hours of wakefulness and subsequent sleep, sleep deprivation might directly dampen the arousal response itself, resulting in changes in HR during and after arousal with a pattern consistent with an increased vagal tonus and a reduced sympathetic drive. Otherwise, if cardiac activity is reduced only during recovery sleep, the differences between wakefulness and sleep would be greater and, therefore, the magnitude of the cardiac response to arousals stronger. To identify such HR arousal-related variations, 7 normal subjects who were totally sleep-deprived and for whom two consecutive recovery nights were available, were examined.

## 2. Methods

### 2.1. Subjects

Seven healthy male subjects, mean age  $28.3 \pm 1.2$  yr (range 19–44) were analyzed. Prior to study, all subjects were screened for any current or past medical, neurological,

cardiac, psychiatric and sleep history and were drug-free at the time of the study. None of them was obese, a smoker or had hypertension or cardiac disease. The laboratory procedures were approved by the Human Ethics Committee of the Defence and Civil Institute of Environmental Medicine, Ontario (Canada), and all subjects gave written consent before their participation.

### 2.2. Experimental design

As previously described (Pigeau et al., 1995), the study was undertaken to examine the effects on mood, performance and body temperature of modafinil and *d*-amphetamine compared to placebo. The subjects participated in a 6-day study, with two baseline days and nights, followed by a 64 h sleep deprivation ending with two recovery nights. Polysomnographic recordings were taken during baseline and recovery nights and during the daytime period to assess the sleep deprivation condition. During the 64 h of sustained wakefulness, the subjects performed several 105 min sessions of cognitive tasks. When not involved in testing sessions, they were allowed to carry out usual activities such as reading, writing, listening to music, and watching TV under the supervision of at least one experimenter. They were not allowed to consume alcohol or caffeine; lying down and vigorous physical activity were not permitted. During night-time, the subjects were allowed to sleep a maximum of 13 h during the first recovery night, whereas time in bed was scheduled between 2200 and 0600 h during the two consecutive baseline recordings and the second recovery night.

### 2.3. Nocturnal sleep recording

The subjects were fitted with electrophysiological recording equipment (Oxford Medilog 9000 II recording system) to measure four EEG leads (C3, C4, P3, P4 referred to linked ears), an electrooculogram (outer canthus), and an electromyogram of chin muscle. An electrocardiogram (ECG) was recorded from a standard D2 lead and higher sampling rate (256 Hz) was achieved after in-laboratory modification. During the first night, monitoring of breathing and leg movements allowed exclusion of sleep-related breathing disorders and periodic leg movement syndrome. Sleep data of the second baseline night and the two recovery nights without alerting substances (placebo condition) were used in the analysis.

Sleep stages were visually scored according to the criteria of Rechtschaffen and Kales (1968) modified for epoch scoring of 20 s, and standard sleep parameters were computed for total sleep.

Recordings were analysed with the PRANA software package (Phitools<sup>®</sup>, Strasbourg, France) for polysomnography and quantitative analyses.

## 2.4. Data analyses

### 2.4.1. Visual EEG scoring

Arousal (AR) was defined according to ASDA criteria (ASDA, 1992) as an abrupt shift in EEG frequency, irrespective of chin EMG changes during non-rapid-eye movement (NREM) sleep but associated with a concurrent EMG increase in rapid-eye movement (REM) sleep. Awakening (AWA) was defined as a transition in fast activities lasting more than 10 s. According to previously described criteria (Trinder et al., 2003), AR and AWA were classified into 4 types on the basis of their duration, i.e. more than 3 s and less than 6 s ( $AR > 3 < 6$ ), more than 6 s and less than 10 s ( $AR > 6 < 10$ ), more than 10 s and less than 15 s ( $AWA > 10 < 15$ ) and  $> 15 < 30$  s ( $AWA > 15$ ). The point-of-onset of each AR and AWA was defined as the first occurrence of alpha or fast EEG activity or K-complex, and the termination as the onset of theta activity persisting for at least 10 s indicating return to sleep. For the baseline night and the two consecutive recovery nights, an AR and AWA index was calculated by dividing the number of AR and AWA by total sleep time. Their number and duration were also calculated for NREM and REM sleep. All arousals and awakenings were scored by two examiners (ES and SL) blinded to experimental conditions.

### 2.4.2. Arousal-related heart rate analysis

To assess the HR changes during arousals and awakenings, up to 10 heartbeats before and 20 heartbeats after the event onset were analysed, with the number of heartbeats included in the post-arousal period independent of when the arousal or the awakening terminated. Only AR and AWA separated for a minimum of 30 s were analysed according to previous criteria (Blasi et al., 2003; Sforza et al., 2000, 2002) and events with obvious HR artefacts or body movements interfering with analysis of the HR signal were visually identified and discarded. After selection of arousals separated by at least 30 s and rejection of the events showing artefacts, the study was made in 70% of the scored events throughout the 3 sleep studies.

Heartbeats were analysed by computer using algorithms developed within the laboratory with visual verification on a beat-to-beat basis. QRS peaks were firstly detected, and then the HR was calculated directly from the R–R interval. Two parameters of HR response were computed for each night. The first variable was the *HR pattern*, calculated from examining the HR fluctuations during AR and AWA. Heartbeats were numbered backwards and forwards from the point of event onset and averaged over AR and AWA within each numbered position. Measurements of HR were normalized by subtracting from each HR value before and after the onset of the AR and AWA, the mean value obtained over the ten beats immediately preceding the event onset. Values were averaged within subjects for each position with respect to the AR and AWA and then averaged over subjects. As a second variable we calculated

the ratio of the highest HR of the twenty beats following the onset of event over the lowest one recorded before onset (*HR ratio*), used as a marker of the magnitude of cardiac activation.

Since REM sleep is associated with a marked state-specific increase in sympathetic drive (Berald et al., 1993; Hornyak et al., 1991; Somers et al., 1993) to assess if sleep stage effect may affect the HR response to arousal, the analysis was conducted separately for NREM and REM sleep. To check whether time of night might affect the cardiac response to AR and/or AWA from sleep, the same analysis was also performed, considering for the 3 nights a similar sleep duration of about 7 h.

## 2.5. Statistical analysis

For overall AR and AWA index, as well as for the number and the duration of the different types of events occurring in NREM and REM sleep, a two-way repeated ANOVA with ‘night’ (baseline, first recovery and second recovery) and ‘sleep stage’ as factors was used. Comparison of the mean values of HR ratio and HR fluctuations across the 4 types of events and between nights was carried out by use of analysis of variance (ANOVA) for repeated measures with night and ‘time’ with respect to AR and AWA type as the factors. The probability values presented are based on Greenhouse-Geisser corrected degree of freedom. Whenever significant main factors or interaction effects were present, post-hoc Tukey’s HSD test was used to assess significant differences. Statistical significance was determined as  $P < 0.05$ . All statistical analyses were performed with the SPSS statistical software package (SPSS for Windows, 10, SPSS Inc, Chicago). Data are reported as means  $\pm$  SEM.

## 3. Results

### 3.1. Polygraphic data and EEG arousal scoring

Details of sleep parameters and visual AR and AWA scoring are reported in Table 1. At baseline, all subjects slept well with high sleep efficiency and a normal sleep stage distribution. As expected, the first recovery night was characterised by important modifications of sleep structure. The major changes were the increase in total sleep time ( $P = 0.001$ ), the rise in slow wave sleep (SWS) ( $P = 0.001$ ) and REM sleep ( $P = 0.001$ ), and a significant reduction in stage 2 ( $P = 0.001$ ). Comparison of the second recovery night versus baseline showed decreases in total sleep time and in stage 2, and an increase in SWS. The differences, however, reached significance only for total sleep time ( $P = 0.01$ ).

Of the total number of events,  $AR > 3 < 6$  and  $AR > 6 < 10$  represented, respectively, 36.2 and 38.4% of the total events in the baseline night. Another 15.1% of

Table 1  
Sleep measures and polygraphic characteristics of arousals and awakenings during the three considered nights (mean(SEM))

	Baseline	First recovery	Second recovery
Total sleep time (min)	446.1 (1.4)	718.4 (2.6) <sup>a,b</sup>	379.2 (2.9) <sup>a</sup>
Stage 1 (%)	8.0 (0.6)	6.3 (0.7)	9.2 (0.8)
Stage 2 (%)	51.2 (0.9)	46.1 (0.8) <sup>a</sup>	48.6 (0.8)
Stages 3–4 (%)	17.0 (1.0)	21.7 (0.8) <sup>a,b</sup>	19.1 (0.9)
Stage REM (%)	23.8 (0.6)	26.0 (0.7) <sup>a,b</sup>	23.2 (0.9)
Sleep efficiency (%)	95.3 (0.5)	96.6 (0.6)	91.9 (1.4)
AR > 3 < 6 s index (n/h)	7.8 (0.8)	4.1 (1.2) <sup>a,b</sup>	7.7 (1.0)
AR > 6 < 10 s index (n/h)	8.2 (0.8)	4.9 (0.6) <sup>a,b</sup>	6.8 (0.7)
AWA > 10 < 15 s index (n/h)	3.3 (0.6)	2.6 (0.4)	3.0 (0.5)
AWA > 15 s index (n/h)	2.2 (0.3)	2.1 (0.3)	2.4 (0.4)
AR > 3 < 6 s duration (s)	4.8 (0.2)	4.8 (0.2)	4.7 (0.2)
AR > 6 < 10 s duration (s)	7.6 (0.2)	7.7 (0.2)	7.6 (0.1)
AWA > 10 < 15 s duration (s)	12.4 (0.2)	12.4 (0.2)	12.2 (0.2)
AWA > 15 s duration (s)	19.7 (0.6)	17.9 (0.6)	18.3 (0.5)

AR, arousal; AWA, awakening.

<sup>a</sup> Significant difference from baseline.

<sup>b</sup> Significant difference from second recovery night.

events were defined as AWA > 10 < 15 and 10.3% as AWA > 15. During the first recovery night, a significant fall in AR lasting less than 10 s was found ( $P = 0.01$ ) with no significant difference across conditions for events lasting more than 10 s.

### 3.2. Cardiac response analysis

Tables 2 and 3 report the number and the duration of the 4 types of events considered for HR analysis throughout the 3 considered nights and during NREM and REM sleep. Their duration was not different compared to overall values and the number was not significantly different between nights.

Table 2  
Heart rate values of analysed arousals and awakenings in the baseline and recovery nights during NREM sleep

	Baseline	First recovery	Second recovery
AR > 3 < 6 s (n)	200	163	175
AR > 3 < 6 s duration (s)	4.82 (0.07)	4.95 (0.06)	4.77 (0.09)
AR > 3 < 6 s HR ratio	1.33 (0.01)	1.25 (0.03)	1.24 (0.03)
AR > 6 < 10 s (n)	218	209	145
AR > 6 < 10 duration (s)	7.76 (0.1)	7.63 (0.1)	7.80 (0.1)
AR > 6 < 10 s HR ratio	1.28 (0.04)	1.27 (0.02)	1.28 (0.03)
AWA > 10 < 15 s (n)	95	110	76
AWA > 10 < 15 duration (s)	12.2 (0.2)	12.3 (0.4)	12.0 (0.3)
AWA > 10 < 15 s HR ratio	1.43 (0.06)	1.36 (0.04)	1.36 (0.03)
AWA > 15 s (n)	76	85	50
AWA > 15 duration (s)	19.0 (0.9)	18.2 (0.6)	18.3 (0.7)
AWA > 15 s HR ratio	1.60 (0.09)	1.54 (0.05)	1.50 (0.05)

AR, arousal; AWA, awakening, HR ratio, ratio of the highest HR of the 20 beats following the onset of event over the lowest one recorded before arousal and awakening onset.

Table 3  
Heart rate values of analysed arousals and awakenings in the baseline and recovery nights during REM sleep

	Baseline	First recovery	Second recovery
AR > 3 < 6 s (n)	73	111	109
AR > 3 < 6 s duration (s)	4.88 (0.13)	4.86 (0.12)	4.99 (0.22)
AR > 3 < 6 s HR ratio	1.25 (0.03)	1.21 (0.03)	1.23 (0.04)
AR > 6 < 10 s (n)	84	111	100
AR > 6 < 10 duration (s)	7.69 (0.1)	7.72 (0.2)	7.12 (0.1)
AR > 6 < 10 s HR ratio	1.28 (0.03)	1.28 (0.03)	1.23 (0.04)
AWA > 10 < 15 s (n)	35	42	40
AWA > 10 < 15 duration (s)	12.8 (0.5)	12.6 (0.4)	11.9 (0.3)
AWA > 10 < 15 s HR ratio	1.41 (0.08)	1.37 (0.04)	1.28 (0.05)
AWA > 15 s (n)	24	34	20
AWA > 15 duration (s)	19.5 (0.9)	18.3 (0.6)	17.6 (0.4)
AWA > 15 s HR ratio	1.73 (0.1)	1.46 (0.07)	1.49 (0.07)

AR, arousal; AWA, awakening, HR ratio, ratio of the highest HR of the twenty beats following the onset of event over the lowest one recorded before arousal and awakening onset.

Occurrence of AR or AWA induced a typical tachycardia significantly greater for AWA > 10 < 15 and AWA > 15. As reported in Table 2, during NREM sleep the HR ratio rose from an average value of  $1.33 \pm 0.07$  and  $1.28 \pm 0.04$ , respectively, for AR > 3 < 6 and AR > 6 < 10 to a value of  $1.43 \pm 0.06$  and  $1.6 \pm 0.09$  during AWA > 10 < 15 and AWA > 15 ( $P = 0.03$ ), with the significantly highest rise during AWA > 15. A similar pattern was found in REM sleep (Table 3).

In baseline night, both in NREM and REM sleep, the analysis of temporal HR fluctuations during arousal showed differences between arousals during the post-arousal period. As shown in Fig. 1, in the baseline night during NREM sleep, ANOVA for repeated measures, with time and event type as the factors, revealed a significant time effect ( $F = 33.8$ ,  $df: 29$ ,  $P < 0.0001$ ) and a significant interaction ( $F = 17.1$ ,  $df: 87$ ,  $P < 0.0001$ ), indicating that the magnitude and duration of HR activation were somewhat affected by the event type with a tendency to larger and sustained HR activation for AWA. Pre-arousal level of HR did not differ between event types, all characterised by a small and insignificant HR rise during the second and first beats preceding the arousal onset. The pattern of HR response was similar for the AR > 3 < 6 ( $F = 19.3$ ,  $P < 0.0001$ ) and the AR > 6 < 10 ( $F = 9.8$ ,  $P < 0.0001$ ) with a significant rise at the first beat after the arousal onset ( $P < 0.0001$ ) and persisting until the 4th beat ( $P < 0.001$ ). Then, HR progressively fell below baseline level reaching significance after the 7th beat ( $P < 0.0001$ ) for AR > 3 < 6 and after the 9th beat for AR > 6 < 10 ( $P < 0.0001$ ). AWA > 10 < 15 ( $F = 31.1$ ,  $P < 0.0001$ ) and AWA > 15 ( $F = 19.3$ ,  $P < 0.0001$ ) were characterised by a different pattern, consisting of a greater increase in HR starting two beats before the visual onset, reaching a significant peak between the second and 9th beats ( $P < 0.0001$ ), and then

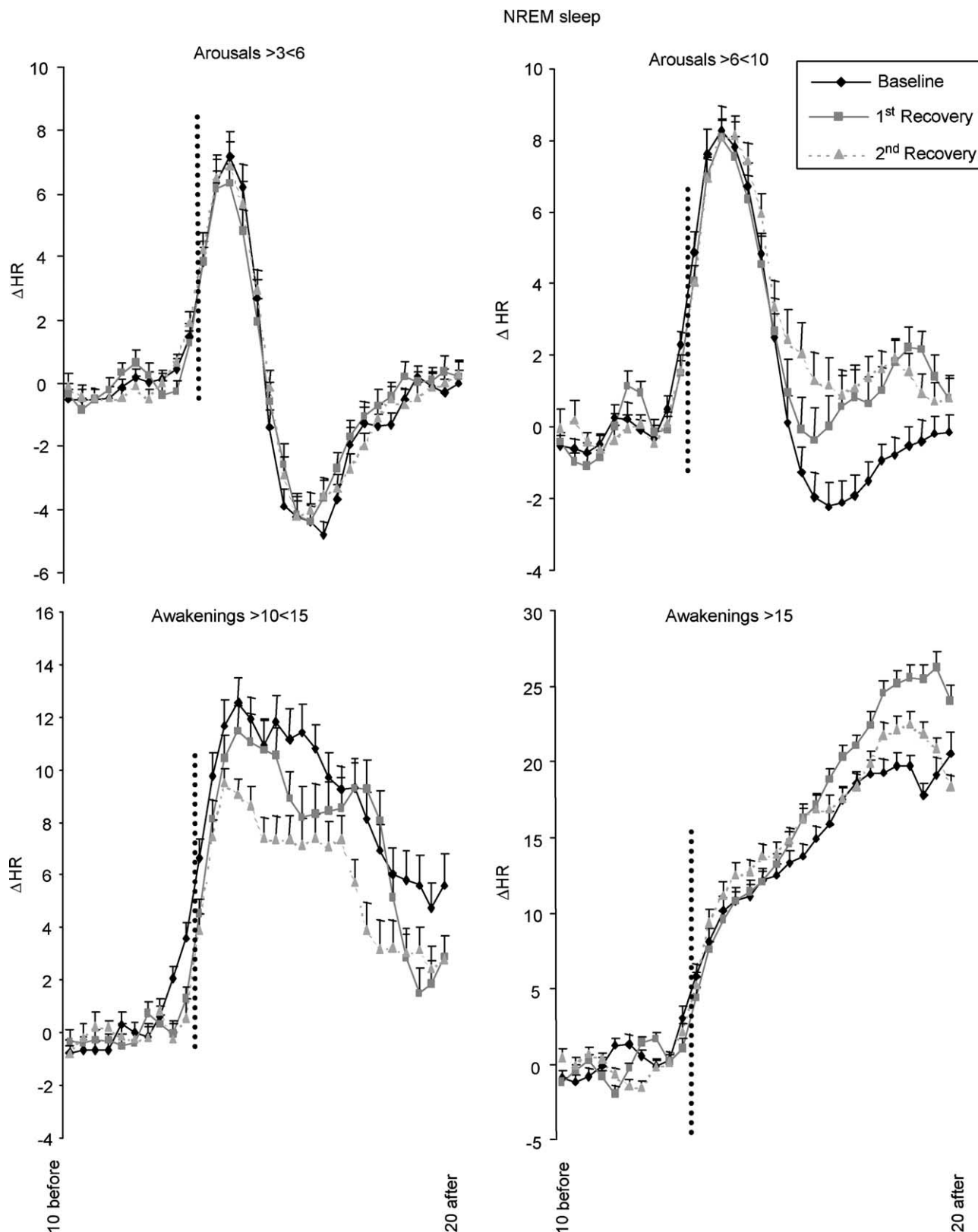


Fig. 1. Temporal heart rate changes averaged across subjects for the 10 beats preceding (before) and the 20 beats following (after) the arousal onset during NREM sleep. Data represent the HR fluctuations for each type of arousal and awakening during NREM sleep. The dashed line indicates the onset of the arousal and awakening for each type. Two different patterns of HR response were found related to arousal duration consisting of a unimodal progressive and sustained rise in HR for arousal lasting  $> 10$  s and a bimodal pattern, i.e. tachycardia followed by bradycardia, for arousal lasting  $< 10$  s. The temporal evolution of HR fluctuations throughout the three nights (black solid line: baseline; solid grey line: first recovery night; dashed grey line: second recovery night) was consistently similar across conditions.

persisting significantly higher ( $P < 0.0001$ ) compared to pre-arousal value.

To take into consideration the possible effect of sleep stage on the dependent variables, we also analysed the data of the 4 event types in REM sleep (Table 3). In REM sleep (Fig. 2), ANOVA revealed a similar pattern of HR variation as in NREM with a significant time effect ( $F = 25.7$ ,  $df: 29$ ,  $P < 0.0001$ ) and a significant interaction ( $F = 12.8$ ,

$P < 0.0001$ ). No differences in HR pattern for all event types and for both pre-arousal and post-arousal periods were seen between NREM and REM sleep ( $P = 0.34$ ).

Comparison of HR response to arousal response in baseline and recovery nights showed that during all event types the cardiac response to arousal was consistently similar over conditions (Table 4). Fig. 1 reveals for each night a significant main effect of time for all event types, but

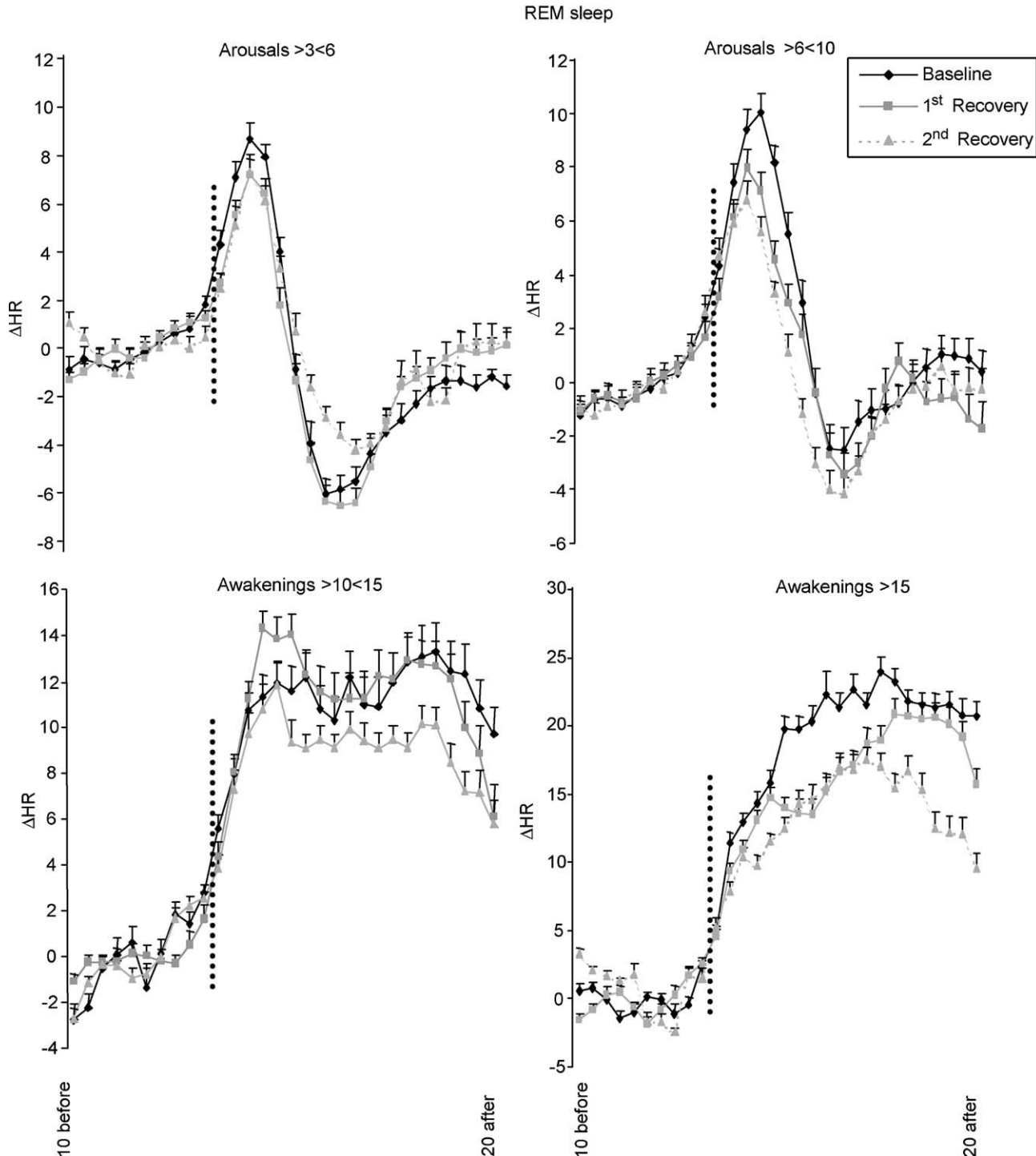


Fig. 2. Temporal evolution of heart rate response to arousal during REM sleep. The unimodal and bimodal patterns of HR response according to arousal duration paralleled the cardiac changes found in NREM sleep without any significant difference between nights.

Table 4  
Two-way rANOVA with the factors Time and Night for arousal types during NREM and REM sleep

Variable	Time			Night			Time × night		
	$F_{58,5}$ -value	<i>P</i> -value	df	$F_{58,5}$ -value	<i>P</i> -value	df	$F_{58,5}$ -value	<i>P</i> -value	df
<i>NREM sleep</i>									
AR > 3 < 6	48.3	<0.0001	29	0.15	ns	2	0.35	ns	58
AR > 6 < 10	32.5	<0.0001	29	0.75	ns	2	1.10	ns	58
AWA > 10 < 15	19.4	<0.0001	29	0.75	ns	2	0.56	ns	58
AWA > 15	163.6	<0.0001	29	0.50	ns	2	1.81	0.004	58
<i>REM sleep</i>									
AR > 3 < 6	48.6	<0.0001	29	0.35	ns	2	1.31	ns	58
AR > 6 < 10	24.4	<0.0001	29	0.89	ns	2	0.84	ns	58
AWA > 10 < 15	19.2	<0.0001	29	0.91	ns	2	1.87	ns	58
AWA > 15	56.0	<0.0001	29	0.47	ns	2	1.47	ns	58

no interaction for events lasting less than 15 s, the pattern of HR response being consistently similar in baseline and recovery nights during NREM sleep. A tendency to reduced lowering of HR values in the post-arousal period was noted in the two recovery nights for the AR > 6 < 10, HR values not falling below reference values. This difference, however, did not reach significance ( $P = 0.07$ ). For AWA > 15 we noted a tendency to a greater rise in HR in the post-arousal period in the recovery nights, with difference reaching significance for the first recovery night ( $P = 0.004$ ). As shown in Fig. 2, the experimental conditions did not induce any significant difference in the degree of arousal-related HR response also in REM sleep, the overall HR over-time fluctuations being similar between nights.

To check whether the lack of changes in HR response to arousal was related to longer sleep time duration in the first recovery night, the same ANOVA design was performed, considering for the 3 nights a similar sleep duration of about 7 h. As depicted in Fig. 3, the results of this ANOVA completely paralleled those previously described. All event types showed a significant time effect but no interaction (AR < 3 < 6: time effect:  $F = 42.6$ ,  $P < 0.0001$ , interaction:  $F = 0.41$ ,  $P = 0.99$ ; AR > 6 < 10: time effect:  $F = 30.0$ ,  $P < 0.0001$ , interaction:  $F = 1.29$ ,  $P = 0.07$ ; AWA > 10 < 15: time effect:  $F = 16.8$ ,  $P < 0.0001$ , interaction  $F = 0.51$ ,  $P = 0.99$ ; AWA > 15: time effect:  $F = 60.0$ ,  $P < 0.0001$ , interaction:  $F = 0.78$ ,  $P = 0.87$ ). Again for AR > 6 < 10, HR values did not fall below reference values compared to baseline night, the difference, however, not reaching significance ( $P = 0.08$ ). A trend to stronger HR rise after arousal onset was present for AWA > 15 in the first recovery night without reaching statistical significance ( $P = 0.07$ ).

#### 4. Discussion

The present study examined the cardiac activation during arousal from sleep in the recovery nights in order to assess

whether sleep deprivation might affect the cardiac response to arousals from sleep. The first important finding of this study was that the time-course of HR arousal-related response was substantially similar between conditions, both the absolute values and the amplitude of the HR response not differing significantly in the sleep deprivation conditions. Second, the HR ratio, an indirect marker of magnitude of cardiac activation, was not associated with significant difference compared to baseline evaluation. Therefore, despite the fact that an activation of parasympathetic tonus and an inhibition of sympathetic tonus after extended wakefulness has been proposed (Delamont et al., 1998; Holmes et al., 2002), our results do not suggest that acute sleep deprivation causes a significant interference on the arousal-related cardiac activation.

In previous studies (Horner, 1996; Sforza et al., 2000; Trinder et al., 2003) the HR variability around arousal has been introduced as a non-invasive tool for the assessment of autonomic control during arousal from sleep. In line with the above studies, we found that arousal from sleep induces a typical time course of HR response, related to duration (Trinder et al., 2003) and intensity of the event (Sforza et al., 2000), and similar in NREM and REM sleep, suggesting that the 'arousal state' is a waking reflex condition, neurophysiologically different from sustained wakefulness (Horner et al., 1997; Trinder et al., 2001, 2003) and inducing a different cardiac response. The increase in cardiac activity during shorter arousal is mainly a reflex activation response (Horner, 1996, 2000; Hornyak et al., 1991; Trinder et al., 2001) implying a withdrawal of vagal tonus and a sympathetic activation during the arousal period (Baust and Bonhert 1969; Horner et al., 1995; Hornyak et al., 1991; Morgan et al., 1996) and a rise in vagal tonus in the post-arousal period related to return to sleep. On the contrary, when arousal lasts longer, a wakefulness state is established, inducing a persistent sympathetic hypertonus and, therefore, a sustained and unimodal HR rise.

Assuming that sleep deprivation may affect the sympathovagal balance, our initial expectation was that the changes in HR during arousal and awakenings would

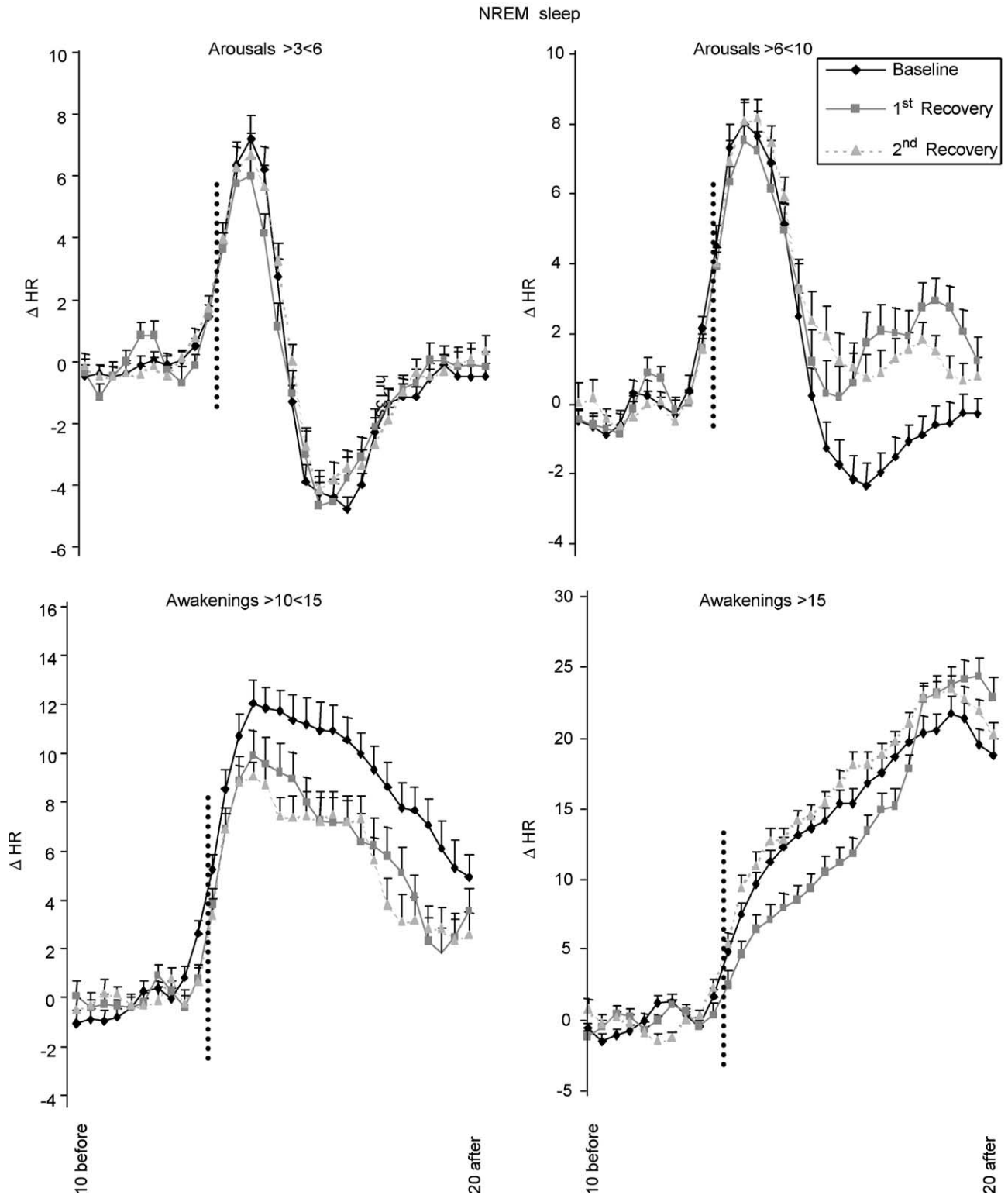


Fig. 3. Time course of HR response to arousal during NREM sleep considering the first 7 h of sleep in the baseline, first recovery and second recovery nights. A similar pattern of HR response during each arousal type without any difference across nights is apparent.

closely parallel the nocturnal changes in vagal and sympathetic activity, a potentiation or a dampening of the HR arousal response occurring during arousals and awakenings. However, contrary to this prediction, we

found that the time course of HR response and the HR ratio did not differ between conditions and between sleep stages, the pattern and the magnitude of cardiac response being substantially similar in baseline and recovery nights.



To clarify whether circadian influences and time asleep (Burgess et al., 1997) may affect our results, we further performed a separate analysis (Fig. 3) considering only the first 7 h of sleep in baseline and recovery nights. Again, the analysis confirms the same pattern of HR variations for all considered arousal types without differences between conditions, suggesting that circadian influences are unlikely to explain the lack of effect of sleep deprivation. Overall, these data indicate that sleep deprivation has little impact on cardiac response to arousal, confirming previous data (Delamont et al., 1998; Kato et al., 2000; Ogawa et al., 2003) that sleep deprivation may induce a resetting of the cardiovascular system caused by peripheral mechanisms such as activation of the renin-angiotensin system (Kato et al., 2000) or by resetting of baroreflex sensitivity (Ogawa et al., 2003).

In our subjects we found a trend to reduced bradycardia for AR shorter than 10 s and greater tachycardia for awakening >15 s in the first recovery night. Although speculative, in the absence of direct measures of sympathetic and vagal drive, we can hypothesise that the lower fall in HR in the post-arousal period may reflect a 'ceiling effect' of prolonged wakefulness on vagal drive not allowing any further rise in vagal tonus when a transitory sympathetic activation takes place (Hornyak et al., 1991). In contrast, the greater HR rise occurring in the post-arousal period for arousal lasting more than 10 s may be explained as a consequence of the return to a wakefulness state in which, due to the 'stressful' effects of sleep deprivation (Bohus and Koolhaas, 1993; Holmes et al., 2002), a hypersympathetic activity may take place (Lusardi et al., 1999).

The lack of any significant difference in the arousal-related HR response after sleep deprivation may be partially explained by some methodological limitations. Firstly, the limited number of subjects studied and their age and health state may contribute to the failure to reach statistical significance in some analyses. However, the close similarity of the transient activation responses obtained in our subjects with those previously described (Trinder et al., 2003) and the analysis conducted in the second recovery night give some confidence that these results are not influenced only by these factors. Further studies considering a larger sample and including older subjects and patients with sleep disorders are needed to replicate our results. Finally, one could question whether the use of quantitative spectral analysis of HR variability would be a better technique to assess more specifically the sympathetic and parasympathetic influences on cardiac activity during sleep. Conventional spectral analysis assumes stationarity in the signal being analysed and requires that a preset amount of data be collected over a fixed time, thus making it unsuitable for processes such as arousals in which there is a significant transient activity. New techniques such as wavelet analysis (Pichot et al., 1999), time-varying spectral analysis (Blasi et al., 2003) and fractal dimension (Togo and Yamamoto,

2000) have been introduced in sleep research to examine HR variability during sleep. These methods, although elegant, are based on a HR variability window length of 5–15 min, thus not sensitive enough to detect autonomic variation when shorter EEG changes occur. New methods applying shorter time windows (Yang et al., 2002) may circumvent this limitation, providing new insight on sympathetic-vagal balance during arousals.

In summary, this is the first study examining the effect of acute sleep deprivation on cardiac response to arousals from sleep. The results indicate that, although arousal from sleep is associated with a typical pattern of cardiac activation influenced by intensity and duration of the arousal, a prolonged sleep deprivation protocol seems to have no significant effect on the pattern and degree of cardiac response to arousal. Whether these findings have an impact on patients with sleep disorders will require future extensive investigation because chronic sleep fragmentation and sleep deprivation may have different and disparate effects on physiological systems.

#### Acknowledgements

We are grateful to Andrea Hawton, Angela Ricci, Alain Roux, John Bowen, Tim Pointing and Patty Odell for valuable technical assistance. We are indebted to Captain Kirk Evoy who coordinated and arranged the selection of Canadian Forces volunteers, and to Dr David Salisbury for providing medical assistance.

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