Effects of Acoustic Stimulation on Cardiovascular Regulation During Sleep

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Abstract: The interaction of wake-sleep states and acoustic stimulation on cardiovascular regulation was studied on rats implanted with electroencephalogram and electromyogram electrodes and an arterial catheter. Mild acoustic stimuli (1000 Hz, 90 dB, 50-ms beeps) were administered during Wakefulness (W), non-rapid eye movement (NREM) sleep and REM sleep and the changes induced in heart period (HP, ms) and mean arterial pressure (MAP, mmHg) were analyzed. Two 30-s sequences of beat-to-beat HP and MAP values were considered before (I) and after (II) acoustic stimulation, respectively. By the effect of stimulation, state-dependent stimulus-locked HP and MAP oscillations were observed, HP oscillations being grossly parallel to the MAP ones but delayed with respect to MAP in the ascending part only; HP and MAP spontaneous fluctuations (HP and MAP variability) increased in NREM and REM sleep (but not in W); HP vs MAP correlation coefficient increased in an algebraic sense. These results show that 1) acoustic stimulation primarily affects the peripheral resistance, and secondarily, through the baroreceptor reflex, HP, thereby increasing the impact of peripheral versus centrally driven autonomic influences on the heart; 2) in NREM sleep, heart excitability is higher than requested by the baroreflex function; 3) cardiac variability is increased by acoustic stimulation during sleep (but not in W); this, in addition to the effects of point 2, may favor cardiac arrhythmias in NREM sleep. Thus, mild acoustic stimuli not perturbing cardiovascular regulation during W may create a specific risk factor during sleep in pathophysiologic conditions.

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INTRODUCTION

THE NEURAL REGULATION OF HEART RATE IS BASED ON A DIRECT EFFECT OF AUTONOMIC OUTFLOW ON THE HEART and an indirect effect through the arterial pressure—heart rate reflex. On the other hand, heart rate and arterial pressure interact with each other through both the arterial pressure—heart rate baroreflex and the direct effect of cardiac output on arterial pressure. Cardiovascular regulation undergoes wide changes in the different states of the wake-sleep cycle (W, wakefulness; NREM, non-rapid eye movement sleep; REM, rapid eye movement sleep).[1] Arterial blood pressure and heart rate, in particular, change in mean value and variability around the mean.[2] Moreover, the balance between centrally driven and baroreflexly mediated influences on the heart and vessels is deeply modified by the wake-sleep state.[3] In addition, cardiovascular regulation during sleep is also influenced by acoustic stimuli.[4-7]

The present work is aimed at studying how acoustic stimulation affects arterial pressure and heart rate and how these effects change in the different states of the wake-sleep cycle.

The experiments were performed on rats, whose high heart rate makes it possible to monitor the vegetative effects of acoustic stimulation with a time resolution as short as 170 ms (length of the heart period, HP, in rat). Acoustic stimulation was performed through a short stimulus of mild intensity. In previous reports,[4-7] longer lasting acoustic stimuli were used, which elicited cardiovascular responses showing up during the stimulus administration. Instead, we utilized short acoustic stimuli to evaluate HP and arterial pressure responses to acoustic stimulation once the stimulus is over. These responses include 1) stimulus phase-locked oscillations, i.e., evoked responses; 2) changes in the size of HP and arterial pressure spontaneous fluctuations; and 3) changes in HP vs arterial pressure statistical dependence. All these parameters, measured in the absence of an enduring acoustic stimulus, reflect the dynamic properties of the cardiovascular regulatory system; studying these oscillations is of importance to understand the mechanisms of cardiovascular instability, i.e., the tendency of arterial pressure and HP to oscillate. Such instability may be revealed by sudden events, like a short acoustic stimulus, increasing the amplitude of the normally present HP and arterial pressure oscillations. We analyzed sequences of beat-to-beat HP and arterial pressure values beginning 30 s before and ending 30 s after the acoustic stimulus, during W, NREM, and REM sleep. Mean HP and arterial pressure evoked responses were evaluated, as well as HP and arterial pressure variability and statistical dependence, in order to understand the mechanisms underlying the acoustic effects on HP and arterial pressure regulation.

We found that 1) in all wake-sleep states, the primary cardiovascular effects of acoustic stimulation consist of fluctuations in peripheral resistance and arterial pressure, determining, via the baroreceptor heart-rate reflex, parallel fluctuations in HP—the relative weight of baroreflex vs centrally driven influences on HP is also increased; 2) the evoked responses to acoustic stimulation depend on the wake-sleep state; and 3) in NREM and REM sleep, the overall HP and arterial pressure variability is increased by the effect of acoustic stimulation, while in W it is decreased.

METHODS

The following protocol was approved by the Bologna University ethics committee on animal experimentation. The experiments were carried out on 6 male Sprague-Dawley rats (Charles River, 250-300 g).

Surgery

Under general anesthesia (1% halothane, 30% O_2, balance N_2O), electrodes were chronically implanted for standard electroencephalographic (EEG) and electromyographic (EMG) recordings. In particular, four miniature stainless steel screws were soldered to copper insulated wire

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and implanted into the skull (1.0 mm anterior and 2.0 mm lateral to bregma, and 0.0 mm anterior and 2.0 mm lateral to lambda). In addition, two teflon-coated platinum stranded wire electrodes were inserted bilaterally into the dorsal nuchal muscle to record EMG activity. A Silastic catheter (0.30 mm i.d., 0.64 mm o.d.) was positioned in the abdominal aorta via the right femoral artery for arterial pressure and HP measurements and withdrawal of blood samples. Calcium heparin (~15 IU/100g/day) was continuously administered through an osmotic pump (ALZA, Palo Alto, CA, Model ALZET 2002) implanted subcutaneously in the interscapular region.

Recording Procedure

After 1 week's recovery, each animal was habituated for 1 day to the recording apparatus in a thermoregulated box (22° C). Food and water were available ad libitum. On the day of the experiment, the arterial catheter was connected to a transducer (Statham, Hato Rey, Puerto Rico, Model P23) for pressure signal recording, and blood gases were measured (Instrumentation Laboratory, Milano, Italy, Model 1304). Acoustic stimuli consisted of short beeps (1000 Hz, 50 ms, 90 dB) delivered through a computerized system during stable W, NREM, and REM sleep states (in REM sleep, the acoustic stimulus was delivered 30 s after the beginning of the episode). Stimuli were manually administered by the operator once both visual observation and automatic state identification (see below) coincided.

A personal computer equipped with software-operated Sound Blaster audio card and analog-to-digital converter was used for acoustic stimulation, data collection, on-line analysis, screen display, and data storage on hard disk.

On-Line Data Analysis

HP and arterial pressure (systolic, diastolic and mean arterial pressure, MAP) values were calculated within each cardiac cycle. EEG spectral power was calculated by Fast Fourier Transform (FFT) on 8-s epochs, a new epoch including the last 6 s of the preceding epoch plus the following 2 s. EMG root mean square (rms) value was calculated over 2-s epochs. EEG spectral components and EMG rms value were retained for sleep-state identification.[3] All the calculated values, as well as the sampled values of arterial pressure, EEG, and EMG, were displayed on the screen. Timing of acoustic stimulation was recorded, and the exact occurrence of the acoustic stimulus displayed on the screen by a marker superimposed on the different tracks (arterial pressure, EEG, EMG). The wake-sleep state was automatically identified by the software utilized. The mean value of EMG rms over the last 2-s epoch, as well as EEG spectral components over the last 8-s epoch, were considered. High EMG and high theta/delta ratio indicated W, low EMG and low theta/delta ratio indicated NREM sleep, low EMG and high theta/delta ratio indicated REM sleep. Reference values for sleep-state identification were set for each rat during a preliminary session and readjusted at the end of each recording session. Awakening from NREM or REM sleep as a consequence of acoustic stimulation was identified to exclude the episode from the following analysis. There is no consensus regarding the classification of wake-sleep states in laboratory animals[8] and even less in terminology and quantitative criteria for sleep-wake transition (awakening) in research subjects (rat,[9,10] human[6]). Awakening identification was based on changes in EEG spectral power and EMG rms. In particular, we recognized an awakening from NREM sleep when a decrease in sigma/theta ratio greater than 22% and an increase in EMG rms greater than 120% were jointly observed following stimulation. Likewise, we recognized an awakening from REM sleep when an increase in sigma/theta ratio greater than 83% and an increase in EMG rms greater than 162% jointly occurred following stimulation. Wake-sleep state and awakening identification was also performed by the operator on the basis of behavior and physiologic criteria. Both classifications were stored and only periods with concurrent classifications were taken into account in the following off-line analysis. The experimental sessions lasted from 10:00 to 17:00 for 5 to 7 days.

Off-Line Data Analysis

Artifact-free epochs starting 30 s before and ending 30 s after the acoustic stimulus were considered, the time origin being placed at the occurrence of acoustic stimulation. The different variables were studied during time intervals I (from –30 s to 0 s) and II (from 0 s to +30 s). Overall interval lengths were chosen as a compromise between the length available for the analysis and the absence in the same sequence of movement artifacts in W and spontaneous awakenings in NREM and REM sleep. With the selected parameters of acoustic stimulation (see above), awakening occurred in about 25% of cases. MAP instead of systolic or diastolic pressure was utilized in the following analysis because MAP is more reliable than both systolic and diastolic pressures, especially in long-term recordings.[11]

In order to describe the HP and MAP evoked responses to acoustic stimulation, i.e., HP and MAP oscillations phase locked to the stimulus, HP and MAP differences with respect to their control value were averaged over different stimulation episodes. To obtain the control values for HP and MAP, the best fit lines of HP and MAP vs time in interval I (i.e., from -30 to 0 s) were calculated, and the predicted values at 0 time were chosen as control values for HP and MAP, respectively.

Spectral analysis on HP and MAP sequential values was performed in intervals I and II according to Zoccoli et al[3] (Appendix A) and the LF_{HP}/HF_{HP} sympathovagal index, i.e. the ratio of low frequency (LF) to high frequency (HF) components of HP power spectrum was evaluated. LF and HF components were defined as the overall spectral power within the 0.033 to 0.8 Hz and 0.81 to 2.5 Hz bands, respectively. Total spectral power was taken as an index of overall HP and MAP variability within intervals I and II. The selected length of 30 s for intervals I and II restricted the power spectrum of HP and MAP variability to frequencies > 0.033 Hz, thus neglecting the peak frequency (0.025 Hz) of the open loop gain determined for the baroreceptor-blood pressure reflex in rat by Dworkin et al[12] but, nonetheless, encompassing most of the frequency range they considered.

The correlation coefficient of HP vs MAP fluctuations (r_{HM}) was taken as an index of statistical dependence of HP vs MAP and was calculated in intervals I and II, according to the procedure used by Zoccoli et al[3] (Appendix B) to calculate the regression coefficient of HP vs MAP fluctuations. r_{HM} provides an index of the balance of central vs peripheral influences to the heart. Cardiovascular autonomic outflow includes central + baroreflex commands to the vessels (sympathetically mediated) and heart (sympathetically + vagally mediated). If an increase occurs in central commands to the heart and vessels, an increase in peripheral resistance and blood pressure will result at the vascular level. At the cardiac level, opposite effects may be observed: a) if the increase in centrally driven commands to the heart prevails over the increase in baroreflex influence from vessels, an increase in heart rate (a decrease in HP) will be observed (negative r_{HM}); b) if the increase in baroreflex influence from vessels prevails, a decrease in heart rate (an increase in HP) will be observed (positive r_{HM}).

At the cardiac level, pure centrally driven effects may be seen in sinoaortic denervated rats: electrical stimulation of the mesencephalic tectum decreased HP in the presence of an increased MAP (negative r_{HM}) at all stimulus intensities.[13] It is worth remembering that prevailing centrally driven influences to the heart (negative r_{HM}) are observed in different physiologic conditions (e.g., at the onset of voluntary exercise[15]). The sign of the balance between central and peripheral influences to the heart may depend on stimulus intensity: in intact rats, whereas moderate electrical stimulation of the mesencephalic tectum increased both MAP and HP, a stronger stimulation increased MAP and decreased HP.[13]

Statistical Analysis

The statistical significance of interstate differences was evaluated by two-way ANOVA, with factors being the animal (6 levels) and the wake-sleep state (3 levels—W, NREM, and REM sleep), together with modified t-test and Bonferroni's method.[16] The statistical significance of differences between intervals I and II was evaluated by paired t-test. To check for habituation, HP and MAP evoked responses were separately evaluated on the first and last 2 days of the experimental period. No significant differences were found, and data from all the experimental days were then pooled for the following analysis. The overall number of HP and MAP –30-s to +30-s sequences considered was 97 in W, 138 in NREM sleep, and 118 in REM sleep.

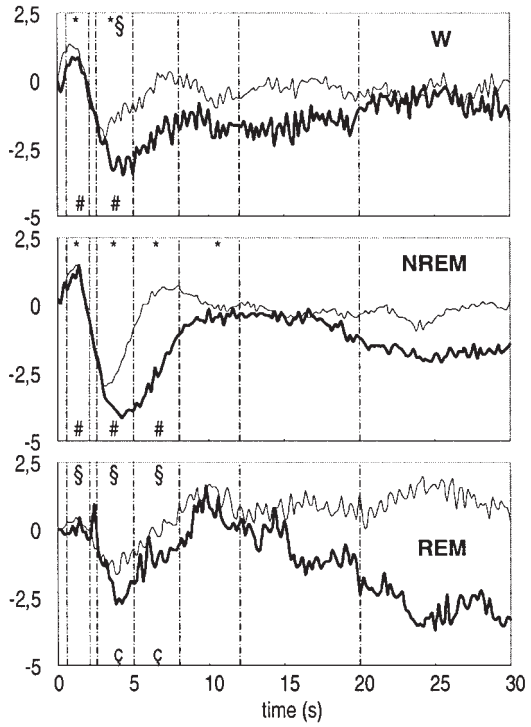

Figure 1—Changes in heart period (HP, ms, bold line) and mean arterial pressure (MAP, mmHg, thin line) in response to acoustic stimulation (averaged values). In the different states, significant differences ($p<0.05$) with respect to 0 are indicated by # and * for HP and MAP, respectively. Both in wakefulness (W) and rapid eye movement (REM) sleep, significant differences ($p<0.05$) with respect to non-rapid eye movement (NREM) sleep are indicated by ç and § for HP and MAP, respectively.

Table 1—Heart period, mean arterial pressure and the sympathovagal index before (I) and after (II) acoustic stimulation in the different wake-sleep states.

	W		NREM		REM
HP (I)	158.69 ± 1.01 §	***	170.10 ± 0.82 §§	*	167.56 ± 0.98 §§§
HP (II)	160.41 ± 1.06	***	169.18 ± 0.85	**	165.54 ± 0.96
MAP (I)	106.12 ± 0.80 NS	NS	105.56 ± 0.83 NS	*	108.10 ± 1.20 §§§
MAP (II)	106.48 ± 0.81	NS	105.71 ± 0.85	**	110.40 ± 1.32
LF_{HP}/HF_{HP} (I)	5.99 ± 0.72 §	***	2,59 ± 0.20 §§§	***	8.12 ± 1.12 NS
LF_{HP}/HF_{HP} (II)	4.19 ± 0.51	NS	4.88 ± 0.41	***	10.11 ± 1.10

HP, heart period, ms; MAP, mean arterial pressure, mmHg; LF_{HP}/HF_{HP}, sympathovagal index, i.e. the ratio between low- and high-frequency components of HP spectral power, adimensional unit; W, NREM, and REM: wakefulness, non-rapid eye movement sleep, and rapid eye movement sleep, respectively; I and II, time intervals -30 s to 0 s and 0 s to +30 s, respectively, time 0 being the time of acoustic stimulation. Values are expressed as mean±SEM. Significant differences with respect to NREM (modified t-test and Bonferroni's method; *, **, *** and NS for $p<0.05$, $p<0.01$, $p<0.001$ and not significant, respectively), as well as between intervals I and II (paired t-test; §, §§, §§§ and NS for $p<0.05$, $p<0.01$, $p<0.001$ and not significant, respectively), are indicated.

RESULTS

Mean values of HP, MAP, and the sympathovagal index LF_{HP}/HF_{HP} in intervals I and II are presented in Table 1 for W, NREM, and REM sleep (Mean ± SEM). HP, MAP and LF_{HP}/HF_{HP} mean values change with the wake-sleep state in agreement with the literature data (cf [2]). The average HP and MAP evoked responses to acoustic stimulation in the different wake-sleep states are presented in Figure 1. For both HP and MAP plots, significant responses (i.e., mean values significantly different from zero) are indicated.

These responses are better represented in NREM sleep and consist of a short-lasting early increase in MAP (and in HP), followed by a longer lasting and deeper decrease in MAP (and HP), followed in turn by return to the control value. The time course of a return from negative peak to control value is significantly different in HP with respect to MAP: when MAP starts increasing from its negative peak, HP continues dropping and returns to its control value with a delay with respect to MAP (Fig. 1). This delay is significantly different from 0 in NREM sleep (1.95±0.53 s, mean±SEM, $p<0.001$) and is also different among states ($p<0.001$). The pattern of HP and MAP evoked responses in NREM sleep is generally similar to the responses obtained in REM sleep and W. However, important differences may be seen among states; significant differences in W and REM sleep with respect to NREM sleep are marked. In particular, the early increase in MAP, clearly seen in NREM sleep and W, is absent in REM sleep.

The index of HP vs MAP statistical dependence, r_{HM}, is presented in Table 2 for intervals I and II. Significant differences in W and REM sleep with respect to NREM sleep and between intervals I and II are indicated. This index was highest in NREM sleep and progressively decreased from NREM sleep to W to REM sleep (cf [3]); it significantly increased from interval I to II, the effects of acoustic stimulation enhancing r_{HM} positively in W and NREM sleep and decreasing its negativity in REM sleep.

The overall HP and MAP variability, expressed by total spectral power, is reported in Table 2. By the effect of acoustic stimulation, HP and MAP total spectral power significantly increased in NREM and REM sleep. In W, MAP total spectral power significantly decreased, while HP total spectral power decreased, though not significantly. As a second index of overall variability, HP and MAP variance was calculated during the last 10-s period of intervals I and II, in order to exclude the contribution of evoked responses (data not reported in Table 2). By the effect of stimulation, HP variance significantly increased ($p<0.001$) in NREM sleep, while MAP variance significantly increased in both NREM and REM sleep ($p<0.001$). In W, MAP variance significantly decreased ($p<0.05$), while HP variance decreased not significantly ($p<0.1$).

DISCUSSION

Sequences of HP and MAP beat-to-beat values 30 s before and 30 s after acoustic stimulation were selected during W, NREM, and REM sleep, and the cardiovascular responses to stimulation were considered. Acoustic stimulation was performed through mild intensity short stimuli: mild intensity stimuli (90 dB), to have a low percentage of awakening from NREM and REM sleep (about 25%); short stimuli (50 ms) to have an impulse stimulus eliciting HP and MAP responses occurring once the stimulus is over. Acoustic stimulation affected the balance of peripheral vs centrally driven autonomic influences to the heart and vessels, as expressed by the HP vs MAP correlation coefficient (r_{HM}), the overall HP and MAP variability, and the evoked responses, i.e. HP and MAP oscillations phase locked to the stimulus (Fig.1). All these parameters reflect the intrinsic dynamic properties (time constants, time lags, HP vs MAP statistical dependence) of cardiovascular regulation. This study is important to better understand the cardiovascular regulatory mechanisms that could give rise to regulatory instability if impaired in pathophysiologic conditions.

Effects of Acoustic Stimulation on the Balance Between Central and Peripheral Influences on the Heart

The significant increase in r_{HM} determined in all states by acoustic stimulation (Table 2) can be interpreted according to the scheme presented above in off-line analysis. This index displays the balance between centrally driven autonomic influences to the heart and influences originating in MAP fluctuations, which are relayed to the heart via the arterial pressure-heart rate reflex. The present results indicate that this balance not only changes between wake-sleep states,[3] but is also modified by the effect of acoustic stimulation. Thus, on the one hand, centrally driven autonomic influences on the heart prevail in REM sleep and peripherally driven influences prevail in NREM sleep, with W lying at mid-way. On the other hand, in all states, acoustic stimulation increases the relative weight of peripheral vs central influences on the heart.

Effects of Acoustic Stimulation on HP and MAP Overall Variability

Spontaneous fluctuations in HP and MAP occur in all wake-sleep states (cf [3]). They also occur in the absence of muscle activity in paralyzed animals.[17] The overall HP and MAP variability, expressed by their total spectral power, consists of spontaneous fluctuations alone in interval I or superimposed on evoked responses in the first part of interval II. During NREM and REM sleep, HP and MAP total spectral power significantly increased from interval I to interval II. The mechanisms involved may include a direct effect of acoustic stimulation on the fluctuations of autonomic outflow to the heart and vessels, as well as an increase in respiratory frequency and tidal volume,[6,18] affecting HP and MAP variability. This increase was not only due to the presence of evoked HP and MAP fluctuations, since HP and MAP variance in the last 10 seconds of interval II also increased during sleep. This shows that cardiovascular regulation is affected by acoustic stimulation well beyond the duration of the acoustic stimulus and the effects of stimulation consist in a larger variability, i.e., a greater instability, in HP and MAP regulation. The importance of this effect can be appreciated by considering that, in conditions of poor regulatory ability, such an increase in variability could result in a frank disruption of heart rate control. On the other hand, the effects of acoustic stimulation during sleep, leading to a wider variability and greater instability in HP and MAP regulation, lasted well beyond the duration of the acoustic stimulus. Thus, short acoustic stimuli may exert long-lasting effects on cardiovascular regulation. Finally, mild acoustic stimuli not increasing and even reducing cardiovascular instability during W may, on the contrary, significantly enhance cardiovascular variability during sleep. This should be taken into account when evaluating the quality of a sleeping as compared with a working environment.

Table 2—Overall variability of mean arterial pressure and heart period, as well as correlation coefficient of heart period vs mean arterial pressure before (I) and after (II) acoustic stimulation in the different states of the wake-sleep cycle.

	W		NREM		REM
TSP_{HP} (I)	2.20 ± 0.046	***	1.52 ± 0.025	***	1.97 ± 0.034
	NS		§§§		§§§
TSP_{HP} (II)	2.10 ± 0.050	***	1.80 ± 0.039	***	2.24 ± 0.068
TSP_{MAP} (I)	1.95 ± 0.047	***	1.46± 0.026	***	1.80 ± 0.037
	§		§§		§§§
TSP_{MAP} (II)	1.83 ± 0.048	*	1.55 ± 0.028	***	2.10 ± 0.059
r_{HM} (I)	0.078 ± 0.034	***	0.253 ± 0.020	***	-0.160 ± 0.031
	§§§		§§§		§§
r_{HM} (II)	0.247 ± 0.039	***	0.412 ± 0.022	***	-0.0253 ± 0.040

TSP_{HP} and TSP_{MAP}, total spectral power of heart period (HP) and mean arterial pressure (MAP), respectively, taken as indexes of overall HP and MAP variability, expressed in arbitrary units; r_{HM}, correlation coefficient of HP vs MAP; W, NREM and REM: wakefulness, non-rapid eye movement sleep, and rapid eye movement sleep, respectively. Values are expressed as Mean±SEM. Significant differences with respect to NREM (modified t-test and Bonferroni's method; *, **, *** for $p<0.05$, $p<0.01$ and $p<0.001$, respectively), as well as between intervals I and II (paired t-test; §, §§, §§§ and NS for $p<0.05$, $p<0.01$, $p<0.001$, and not significant respectively), are indicated.

Evoked Response

In every wake-sleep state, in particular in REM sleep, HP and MAP evoked responses were characterized by a wide variability, at least in part due to HP and MAP spontaneous fluctuations (see above) with magnitude comparable to that of evoked responses. Therefore, averaging was required to obtain a better signal-to-noise ratio. HP and MAP evoked responses are better represented in NREM sleep, possibly due to the steady conditions characterizing this state (see for instance the lower HP and MAP overall variability in NREM sleep with respect to W and REM sleep, Table 2), so that NREM sleep appears to be a state of choice for revealing specific features of the autonomic control of the cardiovascular system.

Different cardiovascular responses to acoustic stimulation have been found among species and studies. In humans, acoustic stimulation induced peripheral vasoconstriction,[4] and blood pressure increase, which was higher in NREM than in REM sleep and not correlated with EEG arousal grade.[5] Consequences of abrupt change in sleep state following acoustic stimulation[6,18] included increased blood pressure and heart rate, decreased stroke volume and cardiac output, increased ventilation (mostly due to increased tidal volume) and increased sympathetic outflow to skeletal muscle. Stimuli not altering EEG pattern produced smaller but consistent pressor responses. On the basis of the above data, the authors concluded that "the arousal induced pressor response appears to be caused by increased peripheral vascular resistance rather than by increased cardiac output."[6] In infants, on the contrary, acoustic stimulation produced a biphasic response with an initial increase, followed by a decrease, in blood pressure and heart rate.[19] In rats, nonsignal stimuli induced a biphasic pressor-depressor response, associated with predominantly deceleratory (at low stimulus intensity) or acceleratory (high stimulus intensity) heart rate responses.[20] Different cardiovascular responses to an acoustic startle stimulus were found in different rat strains,[21] consisting of a rise in blood pressure and minimal variations in heart rate, with the exception of F344 rats, in which heart rate decreased.

As far as the differences in cardiovascular responses are concerned, some points may be considered: a) the biphasic pressor response, found in infants and rats, indicates a tendency of the pressure regulatory system to oscillate under the effects of acoustic stimulation. This tendency, depending on regulatory system parameters (loop gain, time delay, etc), may change with species and age, as well as behavioral conditions; b) the cardiovascular response pattern also depends on stimulus parameters: in rats, a consistent rise in blood pressure was observed by Baudrie et al[21] in response to an acoustic startle stimulus, whereas Quigley et al[20] found responses similar to ours; and c) the relationship between blood pressure and heart rate changes depends on the balance between centrally and peripherally driven influences on the heart—this balance, among others, is affected by stimulus intensity (see off-line analysis).[13]

In our experimental conditions, the changes observed in HP and MAP grossly paralleled each other; this is consistent with the hypothesis that acoustic stimulation induced primary fluctuations in MAP through changes in peripheral resistance; then, by the effect of the baroreceptor-heart rate reflex, secondary parallel fluctuations ensued in HP. This hypothesis is in agreement with the data of Di Nisi et al[4] and the conclusions of Morgan et al.[6]

Return to control value was significantly delayed in HP, with respect to MAP, in NREM sleep and W. This deserves some consideration. First of all, by the effect of this delay, the negative oscillation of HP lasts longer than that of MAP. In other words, HP negative oscillations are slower than MAP oscillations. This is in agreement with the findings of Dworkin et al,[12] showing that the amplitude of the heart rate response to carotid sinus mechanical stimulation decreases with increasing frequency of sinus stimulation more rapidly than the arterial pressure response. Second, according to our hypothesis, HP changes are determined by a

baroreflex response to MAP changes. Based on this hypothesis, the arterial pressure-heart rate reflex appears to work in a non-linear fashion. When MAP drops to its negative peak, a concomitant parallel drop occurs in HP. On the contrary, when MAP starts increasing toward its control value, HP continues decreasing and begins increasing toward the control value after a delay, with the delay changing with the state. Such a delay can also be seen in data from Quigley et al,[20] cf. Figure 1. This delay denotes a condition in which HP is lower (and heart rate higher) than requested by MAP regulatory needs. In other words, heart excitability is higher than requested by the baroreflex function, and this is particularly evident in NREM sleep. It is worth noting that some types of arrhythmias are more frequent in NREM sleep.[22-24]

Interstate differences in evoked responses may be due to different signal-to-noise ratios, i.e., the ratio between evoked responses and spontaneous fluctuations, in the different states, with this ratio being higher in NREM sleep and lower in W and REM sleep. Control of vascular resistance may also differ among states. The lack of an early MAP increase in REM sleep is in agreement with the results of Fewell et al,[25] who induced baroreceptor activation through a reduction in blood volume. In REM sleep, they found a higher increase in heart rate and a lower increase in MAP compared with NREM sleep and W. This indicates that in REM sleep vascular responses are less effective than in NREM sleep and W. Finally, it is worth considering that different auditory signal processing in the different states,[26] and different working points of the autonomic nervous system,[27] may be implicated.

CLINICAL IMPLICATIONS

HP and MAP Overall Variability

It is well established that changes in cardiac autonomic control occur in different pathophysiologic conditions: myocardial infarction[28] and ventricular tachycardia.[29] In these conditions, a reduced HP variability has been reported (cf also [30]). To the extent that a reduced HP variability may be considered an adaptive protection mechanism in cardiovascular disease, any condition that increase HP variability will represent a risk factor. In our experimental setting, the acoustic stimulation during sleep determined an increased MAP and HP variability. Thus, even mild acoustic stimuli not perturbing cardiovascular regulation during wakefulness, if occurring during sleep (both NREM and REM sleep), may create a risk factor for paroxysmal rhythm disturbances in the above mentioned pathophysiologic conditions.

Evoked Response to the Stimulus

Following acoustic stimulation, HP returned to control values later than did MAP. The time delay of an effector response is relevant to the stability of a physiologic control system (cf [27]). This delay also reveals a heart rate higher than that required by the baroreflex function, i.e., a condition of increased heart excitability. An increased cardiac excitability underlies many pathophysiologic conditions. The arrhythmogenic properties of sleep are the topic of long-standing research and remain controversial. Cardiac arrhythmias occur both in REM and NREM sleep,[22-24,31-33] albeit with different underlying mechanisms: a) "The increase in sympathetic nerve activity that occurs at the onset of REM sleep provides a potent stimulus for ventricular tachyarrhythmias because of the arrhythmogenic influence of neurally released catecholamines. Sympathetic nerve activation…can increase cardiac vulnerability."[33] and b) in our experimental conditions, the delay between HP and MAP, i.e., the duration of increased excitability, was longest during NREM sleep, suggesting a mechanism responsible for arrhythmias occurring during this sleep state.

REFERENCES

1. Franzini C. Cardiovascular physiology: the peripheral circulation. In: Kryger MH, Roth T, Dement WC, eds. Principles and practice of sleep medicine, 3rd ed. Philadelphia: WB Saunders Company, 2000:193-203.
2. Lenzi P, Zoccoli G, Franzini C. Regulation of the cerebral and extracerebral circulation during the wake-sleep cycle. In: Lugaresi E, Parmeggiani PL, eds. Somatic and autonomic regulation in sleep. Physiological and clinical aspects. Milan: Springer-Verlag, 1997:25-41.
3. Zoccoli G, Andreoli E, Bojic T, Cianci T, Franzini C, Predieri S, Lenzi P. Central and baroreflex control of heart rate during the wake-sleep cycle in rat. Sleep 2001;24:753-758.
4. Di Nisi J, Muzet A, Ehrhart J, Libert JP. Comparison of cardiovascular responses to noise during waking and sleeping in humans. Sleep 1990;13:108-120.
5. Davies RJ, Belt PJ, Robert SJ, Ali NJ, Stradling JR. Arterial blood pressure responses to graded transient arousal from sleep in normal humans. J Appl Physiol 1993;74:1123-1130.
6. Morgan BJ, Crabtree DC, Puleo DS, Badr MS, Toiber F, Skatrud JB. Neurocirculatory consequences of abrupt change in sleep state in humans. J Appl Physiol 1996;80:1627-1636.
7. Bao G, Metreveli N, Fletcher EC. Acute and chronic blood pressure response to recurrent acoustic arousal in rats. Am J Hypertens 1999;12:504-510.
8. Witting W, van der Werf D, Mirmiran M. An on-line automated sleep-wake classification system for laboratory animals. J Neurosci Meth 1996;66:109-112.
9. Trachsel L, Tobler I, Borbely AA. Electroencephalogram analysis of non-rapid eye movement sleep in rats. Am J Physiol 1988;255:R27-R37.
10. Neckelmann D, Ursin R. Sleep stages and EEG power spectrum in relation to acoustical stimulus arousal thereshold in the rat. Sleep 1993;16:467-477.
11. Oosting J, Struijker-Boudier HAJ, Janssen BJA. Validation of a continuous baroreflex sensitivity index calculated from spontaneous fluctuations of blood pressure and pulse intervals in rats. J Hypertens 1997;15:391-399.
12. Dworkin BR, Tang X, Snyder AJ, Dworkin S. Carotid and aortic baroreflexes of the rat: II. Open-loop frequency response and the blood pressure spectrum. Am J Physiol 2000;279:R1922-R1933.
13. Schenberg LC, Vasquez EC, da Costa MB. Cardiac baroreflex dynamics during the defence reaction in freely moving rats. Brain Res 1993;621:50-58.
14. Mancia G, Marks AL. Arterial baroreflexes in humans. In: Shepherd JT, Abboud FM, eds. Handbook of physiology, section 2, the cardiovascular system, Vol. 3, part 1. Bethesda, American Physiological Society, 1983;755-794.
15. Rowell LB. Human cardiovascular control. Oxford: Oxford University Press, 1993;500.
16. Wallenstein S, Zucker CL, Fleiss JL. Some statistical methods useful in circulation research. Circ Res 1980;47:1-9.
17. Dworkin BR, Dworkin S, Tang X. Carotid and aortic baroreflexes of the rat: I. Open-loop steady-state properties and blood pressure variability. Am J Physiol 2000;279:R1910-R1921.
18. Badr MS, Morgan BJ, Finn L, et al. Ventilatory response to induced auditory arousals during NREM sleep. Sleep 1997;20:707-714.
19. Harrington C, Kirjavainen T, Teng A, Sullivan CE. Cardiovascular responses to three simple, provocative tests of autonomic activity in sleeping infants. J Appl Physiol 2001;91:561-568.
20. Quigley KS, Berntson GG. Autonomic origins of cardiac responses to nonsignal stimuli in the rat. Behav Neurosci 1990;104:751-762.
21. Baudrie V, Laude D, Chaouloff F, Elghozi JL. Genetic influences on cardiovascular responses to an acoustic startle stimulus in rats. Clin Exp Pharmacol Physiol 2001;28:1096-1099.
22. Skinner JE, Mohr DN, Kellaway P. Sleep-stage regulation of ventricular arrhythmias in the unanesthetized pig. Circ Res 1975;37:342-349.
23. Otsuka K, Yanaga T, Ichimaru Y, Seto K. Sleep and night-type arrhythmias. Jpn Heart J 1982;23:479-486.
24. Carter NL, Hunyor SN, Crawford G, Kelly D, Smith AJ. Environmental noise and sleep—A study of arousal, cardiac arrhythmia and urinary catecholamines. Sleep 1994;17:298-307.
25. Fewell JE, Williams BJ, Hill DE. Behavioral state influences the cardiovascular response to hemorrhage in lambs. J Dev Physiol 1984;6:339-348.
26. Velluti RA. Interactions between sleep and sensory physiology. J Sleep Res 1997;6:61-77.
27. Keyl C, Schneider A, Dambacher M, Bernardi L. Time delay of vagally mediated cardiac baroreflex response varies with autonomic cardiovascular control. J Appl Physiol 2001;91:283-289.
28. La Rovere MT, Bigger Jr JT, Marcus FI, Mortara A, Schwartz PJ. Baroreflex sensitivity and heart-rate variability in prediction of total cardiac mortality after myocardial infarction. Lancet 1998;351:478-484.
29. Shusterman V, Aysin B, Weiss RR, et al. Dynamics of low-frequency R-R interval oscillations preceding spontaneous ventricular tachycardia. Am Heart J 2000;139:126-133.
30. Schwartz PJ. The neural control of heart rate and risk stratification after myocardial infarction. Eur Heart J 1999;1:H33-H43.
31. Orr WC, Stahl ML, Whitsett T, Langevin E. Physiological sleep patterns and cardiac arrhythmias. Am Heart J 1979;97:128-129.
32. Skinner JE, Douglas FM, Harper HM. Higher cerebral regulation of cardiovascular and respiratory function. In: Kryger MH, Roth C, Dement WC, eds. Principles and practice of sleep medicine. Philadelphia: WB Saunders Company, 1989;276-292.
33. Verrier RL, Harper RM, Hobson JA. Cardiovascular Physiology: Central and autonomic regulation. In: Kryger MH, Roth T, Dement WC, eds. Principles and practice of sleep medicine, 3rd ed. Philadelphia: WB Saunders Company, 2000;179-191.