Clonidine Has a Paradoxical Effect on Cyclic Arousal and Sleep Bruxism during NREM Sleep

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Study Objective: Clonidine disrupts the NREM/REM sleep cycle and reduces the incidence of rhythmic masticatory muscle activity (RMMA) characteristic of sleep bruxism (SB). RMMA/SB is associated with brief and transient sleep arousals. This study investigates the effect of clonidine on the cyclic alternating pattern (CAP) in order to explore the role of cyclic arousal fluctuation in RMMA/SB.

Design: Polysomnographic recordings from a pharmacological study.

Setting: University sleep research laboratory.

Participants and Interventions: Sixteen SB subjects received a single dose of clonidine or placebo at bedtime in a crossover design.

Measurements and Results: Sleep variables and RMMA/SB index were evaluated. CAP was scored to assess arousal instability between sleep-maintaining processes (phase A1) and stronger arousal processes (phases A2 and A3). Paired t-tests, ANOVAs, and cross-correlations were performed. Under clonidine, CAP time, and particularly the number of A3 phases, increased (P ≤ 0.01). RMMA/SB onset was time correlated with phases A2 and A3 for both placebo and clonidine nights (P ≤ 0.004). However, under clonidine, this positive correlation began up to 40 min before the RMMA/SB episode.

Conclusions: CAP phase A3 frequency increased under clonidine, but paradoxically, RMMA/SB decreased. RMMA/SB was associated with and facilitated in CAP phase A2 and A3 rhythms. However, SB generation could be influenced by other factors besides sleep arousal pressure. NREM/REM ultradian cyclic arousal fluctuations may be required for RMMA/SB onset.

Keywords: Clonidine, sleep bruxism, rhythmic masticatory muscle activity, cyclic alternating pattern

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Table 1—CAP variables during placebo and clonidine nights

<table>
<thead>
<tr>
<th></th>
<th>PLACEBO NIGHT</th>
<th>CLONIDINE NIGHT</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CAP rate (%)</td>
<td>27.8 ± 1.2</td>
<td>27.3 ± 1.4</td>
<td>0.65</td>
</tr>
<tr>
<td>CAP time (min)</td>
<td>99.8 ± 4.6</td>
<td>127.7 ± 6.1</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>CAP sequence (n)</td>
<td>36.5 ± 1.3</td>
<td>44.4 ± 1.8</td>
<td>0.001</td>
</tr>
<tr>
<td>A1 (n)</td>
<td>99.4 ± 9.1</td>
<td>84.5 ± 8.2</td>
<td>0.19</td>
</tr>
<tr>
<td>A2 (n)</td>
<td>40.1 ± 5.2</td>
<td>48.8 ± 6.6</td>
<td>0.12</td>
</tr>
<tr>
<td>A3 (n)</td>
<td>59.2 ± 6.2</td>
<td>96 ± 13.9</td>
<td>0.01</td>
</tr>
<tr>
<td>A1 (%)</td>
<td>49.2 ± 2.7</td>
<td>37.9 ± 3.6</td>
<td>0.01</td>
</tr>
<tr>
<td>A2 (%)</td>
<td>19.8 ± 1.7</td>
<td>21.2 ± 2.5</td>
<td>0.47</td>
</tr>
<tr>
<td>A3 (%)</td>
<td>31 ± 3.5</td>
<td>40.9 ± 4.5</td>
<td>0.03</td>
</tr>
<tr>
<td>Phase A duration (s)</td>
<td>8.9 ± 0.4</td>
<td>9.1 ± 0.4</td>
<td>0.58</td>
</tr>
<tr>
<td>Phase B duration (s)</td>
<td>21.6 ± 0.5</td>
<td>24.6 ± 0.6</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>CAP cycle duration (s)</td>
<td>30.5 ± 0.7</td>
<td>33.7 ± 0.6</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>A1 duration (s)</td>
<td>5.6 ± 0.2</td>
<td>5.5 ± 0.2</td>
<td>0.5</td>
</tr>
<tr>
<td>A2 duration (s)</td>
<td>7.9 ± 0.3</td>
<td>7.7 ± 0.4</td>
<td>0.58</td>
</tr>
<tr>
<td>A3 duration (s)</td>
<td>15.2 ± 0.6</td>
<td>13.5 ± 0.6</td>
<td>0.03</td>
</tr>
</tbody>
</table>

Data are presented as mean ± SEM. CAP refers to cyclic alternating pattern.

METHODS

Subjects

We analyzed polysomnographic data of 16 SB subjects (6 males and 10 females; mean age 24.5; range 21 to 31) recorded in our laboratory between 2000 and 2004. Subjects were selected based on a history of tooth-grinding (> 3 nights/week) and clinical signs and symptoms of SB (tooth wear, masseter muscle hypertrophy, morning jaw muscle fatigue or tenderness). SB diagnosis was confirmed if subjects met the standard polysomnographic research criteria (> 4 episodes of RMMA/hour of sleep) in the second night of sleep recording. The experimental protocol was approved by the research ethics board of the Hôpital du Sacré-Coeur, Montreal.

Study Design

All subjects underwent 4 nights of audio-video polysomnographic recordings. The first night was for habituation and the second for SB diagnosis. The experimental protocol was conducted during the third and fourth nights. Subjects received a single dose of either placebo or clonidine (0.3 mg by mouth) 1 hour before bedtime in a randomized double-blind crossover design. A 1-week washout interval between the 2 experimental nights was provided. According to its pharmacokinetic profile, clonidine reaches a plasmatic peak within approximately 2 h following oral administration. The experimental protocol was approved by the research ethics board of the Hôpital du Sacré-Coeur, Montreal.

Sleep Data Collection and Scoring

The following polysomnographic variables were recorded and analyzed blind to medication or placebo allocation: EEG (C, A), electrooculogram (EOG), electrocardiogram (EKG), chest respiratory movements, and electromyographic (EMG) activity in the suprahyoid, masseter, temporalis and anterior tibialis muscles. All signals were digitalized at 128 Hz using commercial software (Harmonie, Stellate, Montreal, Canada), as described elsewhere. Sleep stages, sleep arousals, and RMMA/SB episodes were scored according to standard criteria. Each CAP phase A was visually detected during NREM sleep on the C, A derivation by the use of Somnologica (Embla, Germany), then classified into subtypes A1, A2, or A3. The following CAP parameters were evaluated: total CAP time, CAP rate, number and duration of CAP sequences, number and duration of A phases, number and duration of B phases, and number and percent of subtypes A1, A2, and A3.

Data Analyses and Statistics

Duration of the first 4 NREM/REM sleep cycles was normalized within each subject by dividing NREM periods into 20 intervals and REM periods into 5 intervals, for 100 intervals over the entire night. Each sleep cycle was then averaged into 4 NREM sections and 1 REM section (each comprising 5 intervals). Both placebo and clonidine nights were normalized. However, for clonidine nights, REM sections were time estimated due to the strong reduction in REM sleep duration. Frequency distributions of RMMA/SB episodes and CAP phases A1, A2, and A3 per hour of sleep were calculated over sleep cycles.

Statistical analyses were performed using SPSS (SPSS Inc., Chicago, IL, USA). The paired t-test was applied for within-group comparison (placebo vs. clonidine) and repeated measures ANOVAs (treatment × cycle × section) were applied to CAP variables and RMMA/SB episode distribution over sleep cycles. Placebo and clonidine nights were compared for NREM sleep only, as CAP cannot be scored during REM. Repeated measures ANOVAs were also performed for each NREM cycle (treatment × section) due to the clonidine pharmacokinetics profile. The Huynh-Feldt correction for sphericity was applied to all ANOVA calculations. P-values were considered significant at ≤ 0.05.

RMMA/SB episodes were cross-correlated with CAP phases A1, A2, and A3. Cross-correlation plots are used to identify temporal associations between 2 series of events at individual time lags. Each time lag corresponds to 4 minutes. A significant correlation at lag 0 indicates that the 2 events occur at the same time. Correlations at positive lags relate values in the first series to subsequent values in the second series. Similarly, correlations at negative lags relate values in the first series to preceding values in the second series.

RESULTS

CAP Variables and Sleep Bruxism

CAP time and CAP sequence increased significantly (≤ 0.001) with clonidine compared with the placebo night. Because clonidine increased the duration of both total CAP time and NREM sleep, the CAP rate did not change. The number of CAP A3 phases increased significantly with clonidine compared with the placebo night (P = 0.01) (Table 1). As previously reported, both RMMA/SB and sleep variables changed significantly under the influence of clonidine. RMMA/SB index decreased by 61% (P = 0.02), sleep stage 2 increased, and REM sleep was markedly reduced (both P < 0.001). The number of sleep arousals per hour of sleep did not change under clonidine.
RMMASB and CAP Phase A Distribution over Sleep Cycles

Clonidine blunted the linear crescendo pattern of RMMASB occurrence across the NREM/REM transition period compared to placebo (overall treatment effect P = 0.002). This effect was stronger in the third and fourth sleep cycles (ANOVA for cycles 3 and 4, P = 0.001). Under clonidine, phase A1 frequency (number/hour of sleep) was slightly reduced (overall treatment effect, P = 0.02), but no difference was observed between placebo and clonidine nights for the phase A1 ultradian NREM/REM fluctuation pattern. In contrast, phase A2 and A3 distributions across the sleep cycle changed markedly under clonidine (overall treatment x section interaction, P = 0.0005 and 0.0009, respectively). Within each cycle, the linear crescendo mode of phase A2 and A3 frequencies disappeared from cycles 2 to 4 (ANOVA for each cycle, placebo linear contrast P < 0.002; clonidine linear contrast P > 0.2) (Figure 1).

Cross-Correlation Analysis between RMMASB Episodes and CAP A Phases

For both placebo and clonidine nights, an equipotent positive correlation was found between RMMASB episodes and CAP phases A2 and A3 (Figure 2). During the placebo night the time correlation for phase A2 was significant at lag 0 (RMMASB onset), and lasted for the next 8 min (lag 0 coefficient = 0.287, P = 0.004). For phase A3, a significant time correlation began in the 4 min preceding RMMASB episodes and lasted for the next 8 min (lag 0 coefficient = 0.637, P < 0.0001). Under clonidine, a significant positive time correlation at RMMASB onset was still present for A2 (lag 0 coefficient = 0.304, P = 0.002) and A3 (lag 0 coefficient = 0.569, P < 0.0001). However, the time correlations in clonidine nights were significantly higher in the 40 min preceding RMMASB episodes for both A2 and A3 phases.

DISCUSSION

This experimental pharmacological study evaluated the relationship between RMMASB and CAP phase A. Polysomnographic recordings showed that both CAP phase A patterns and RMMASB occurrence change under clonidine. Specifically, the cyclic rise in phase A2 and A3 frequency and RMMASB activity in the pre-REM periods were blunted under clonidine. Paradoxically, the number of RMMASB decreased and the number of phase A3 rose. In addition, in both placebo and clonidine nights, RMMASB episodes were temporally correlated with CAP phases A2 and A3 and their endogenous fluctuation across ultradian NREM/REM sleep cycles. Previous studies showed the association between CAP phases A2 and A3 with RMMASB, but no time correlation.9

The present study found that clonidine increases CAP time, CAP sequence, and phase A3, indicating increased arousal instability and sleep perturbation,16,17 probably related to clonidine action on the brainstem neuronal systems that regulate the switch from NREM to REM sleep.29 However, whether the effect of clonidine on sleep arousal instability is linked to the observed reduction in RMMASB episodes remains to be demonstrated.30

CAP as the Permissive Window for RMMASB Occurrence

Previous studies have described RMMASB as a motor activity secondary to sleep arousal,8-10,13 and have associated it with CAP phase A.9 Our study provides new insights into the role of phase A and the differences between phases A1, A2, and A3. Phase A1 corresponds to the slow EEG oscillations, and is associated with the build-up and maintenance of NREM sleep.21,31,32 In contrast, arousal subtypes A2 and A3 are lower in NREM deep sleep and they increase linearly before REM sleep onset. The RMMASB occurrence pattern reflects the ultradian fluctuation in CAP phases A2 and A3. Phase A1 is also

**Figure 1**—Distribution over NREM/REM sleep cycles of CAP phases A1, A2, and A3 (number/h), and RMMASB activity (episodes/h) for placebo (black circles) and clonidine (white circles) nights. Vertical dotted lines delimit each NREM/REM sleep cycle. Mean values (SEM) are shown. C1 = first sleep cycle; C2 = second sleep cycle; C3 = third sleep cycle; C4 = fourth sleep cycle; RMMASB = rhythmic masticatory muscle activity/ sleep bruxism.
Although the quantitative time-correlation analysis does not reveal a causal relationship between CAP phase A and SB, it suggests that phases A2 and A3 constitute the permissive physiological window for RMMA/SB generation during sleep.

Figure 2—Cross-correlation plots between RMMA/SB episodes and CAP phases A1, A2, and A3 for placebo (A) and clonidine (B) nights. Horizontal upper and lower lines denote significant P value at 0.05. The vertical line denotes lag 0, or RMMA/SB onset. Each lag lasts 4 minutes (minus 40 min before SB onset and plus 40 min after SB onset).

negatively correlated with RMMA/SB episodes, while phases A2 and A3 are strongly time-correlated with RMMA/SB onset. In fact, the time correlation between RMMA/SB activity and phases A2 and A3 is preserved even in clonidine nights.
A recent study evaluating the time relationship between RMMA/SB episodes and autonomic nervous system activity showed a change in sympathetic/parasympathetic modulation starting approximately 8 to 4 minutes before RMMA/SB onset.12,13 Our analysis goes further to show that the balance between subtypes A1 to A3 shifts within the same time range as the previously described shift in sympathetic cardiac activity. This suggests that phase A changes are the EEG counterpart of the sympathetic/parasympathetic balance in sleep arousal.16

Other studies15-18 support the facilitatory role of CAP phase A in the occurrence of motor events during sleep. For example, periodic limb movements in sleep (PLMS) have been reported to occur frequently within CAP, with 96% of PLMS episodes observed in phases A2 and A3. However, the CAP itself does not generate these movements during sleep; instead, it is the temporal window in which arousal and motor events are grouped and in which their rhythmic occurrence is facilitated.

Putative Effects of Clonidine on SB and CAP

Clonidine is an α2-adrenergic agonist that acts on multiple sites in the central and peripheral nervous systems.38 It has both a direct and indirect influence on the autonomic nervous system,41-43 sleep cycles,44 motor control,42-43 and most probably arousal instability.

Clonidine exerts a strong sympathetic inhibitory effect by altering the balance from sympathetic to parasympathetic tone,4,39 due to activation of the α2 adrenergic autoreceptors and postsynaptic receptors in the brainstem and the imidazoline-I1 receptors in the rostral ventrolateral medulla.46 By preventing sympathetic rise, clonidine may blunt the cascade of arousal-related neurovegetative responses that precede RMMA/SB episodes, thereby inhibiting SB.

Clonidine also has marked effects on sleep macrostructure.4 Significant increases in stage 2 as well as reduced deep and REM sleep duration have been reported.13 REM sleep onset appears to be strongly influenced by the adrenergic and noradrenergic systems.45,46 The neurotransmitters in these systems are implicated in inhibitory mechanisms that mainly involve postsynaptic α2-adrenoceptors in the locus coeruleus area of the brainstem.47 However, clonidine may also activate non-adrenergic neurons in the pontine reticular formation and thereby exert an indirect influence on the GABA-ergic and cholinergic neurons that modulate REM sleep onset.48-50 In the present study, clonidine strongly reduced REM sleep duration, blunting the cyclic rise of CAP phases A2 and A3 in the NREM/REM transition period. Thus, clonidine perturbation of the NREM/REM ultradian cycle may by itself have prevented RMMA/SB onset. This fluctuation in ultradian arousal may be required for RMMA/SB occurrence. Experimental protocols using pharmacological or physical REM alteration (REM sleep deprivation or enhancing methods) may help determine the nature of this influence.51-53

Clonidine may also affect motor control pathways. RMMA/SB are spontaneous motor events that occur during sleep. So far, the exact mechanism responsible for the generation of RMMA/SB remains unknown. Possible contributing factors include autonomic sympathetic cardiac activity,13,54 the hypothalamic-adrenal axis,55-57 genetics,58,59 and complex neurochemical influences involving catecholamines, serotonin, histamine, acetylcholine, or orexin.60 Because clonidine affects the noradrenergic pathways, it may indirectly influence dopamine release from the nucleus striatum.58,61,62 However, there is only weak evidence for the role of catecholamines in generating RMMA/SB, with conflicting results and a lack of randomized controlled trials.60,63,64

CONCLUSIONS

RMMA/SB is associated with and facilitated by CAP phase A2 and A3 rhythms within a sleep arousal. However, under clonidine, increased CAP phase A3 frequency (arousal pressure) is observed with a paradoxical reduction in RMMA/SB activity. Although CAP phases A2 and A3 could reflect permissive physiological windows, RMMA/SB generation could be influenced by other factors besides arousal pressure. Notably, fluctuations in the NREM/REM ultradian cyclic arousal may be required for RMMA/SB to occur.

REFERENCES


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DISCLOSURE STATEMENT

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