Disturbed Sleep Patterns in Elders with Mild Cognitive Impairment: The Role of Memory Decline and ApoE ε4 Genotype

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Abstract: Sleep disturbances are prevalent in patients with Alzheimer’s disease (AD), being one of the most troubling symptoms during the progression of disease. However, little research has been made to determine if impaired sleep patterns appear years before AD diagnosis. This study tries to shed light on this issue by performing polysomnographic recordings in healthy elders and patients with mild cognitive impairment (MCI). We further investigated whether changes in sleep patterns parallel memory decline as well as its relationship with the Apolipoprotein E (ApoE) ε4 genotype. Results showed a significant shortening of rapid eye movement (REM) sleep together with increased fragmentations of slow-wave sleep in MCI patients relative to healthy elders. Interestingly, we further showed that reduction of REM sleep in MCI patients with ApoE ε4 was more noticeable than in ε4 non-carriers. Contrary to our initial hypothesis, changes in sleep patterns were not correlated with memory performance in MCI patients. Instead, increased REM sleep accompanied enhanced immediate recall in MCI ε4 non-carriers. Taken together, these results suggest that sleep disruptions are evident years before diagnosis of AD, which may have implications for early detection of dementia and/or therapeutic management of sleep complaints in MCI patients.

Keywords: Aging, Alzheimer disease, ApoE, Memory loss, Mild cognitive impairment, Polysomnography, REM, Sleep fragmentation.

INTRODUCTION

Aging is associated with progressive impairment of the organism’s functional capacity, from cells to systems. With advancing age, a progressive loss of coordination among circadian systems and external inputs results in an advanced phase of the circadian cycle [1], which leads to earlier sleep onset and earlier morning waking in elderly subjects [2]. In line with these findings, previous studies have reported age-related volume reductions of the ventrolateral preoptic (VLPO) nucleus [3], an anterior hypothalamic structure critically involved in sleep onset and maintenance [4]. The aforementioned results might contribute to increased sleep fragmentation [5] and slow-wave sleep (SWS) deficits found in older adults [6]. Therefore, evidence suggests that neural systems governing regulation and maintenance of sleep-wake cycle are particularly vulnerable to senescence [7], and might be caused by age-related neural impairments [8].

Accumulating evidence suggest a high prevalence of sleep disorders in age-related neurodegenerative conditions as Parkinson [9], dementia with Lewy bodies [10], frontotemporal dementia [11], and Alzheimer’s disease (AD) [12-16]. AD patients, depending on disease severity, have shown impaired sleep patterns ranging from sleep fragmentation to decreased time spent in SWS and/or rapid eye movement (REM) sleep [17]. However, whether these altered sleep patterns precede AD diagnosis remains controversial to date.

Elders with mild cognitive impairment (MCI) show a significant decline in memory over time beyond what is expected from normal aging. In fact, 19% to 50% of individuals with MCI progress to dementia (typically AD) over a period of 3 years [18]. The belief that MCI represents the prodromal stage of AD has received strong support from neuropathological [19, 20], biochemical [21], neuroimaging [22, 23], and neuropsychological studies [24, 25]. To the best of our knowledge, only one study has explored nocturnal sleep patterns in MCI patients by performing polysomnographic (PSG) recordings [26]. Authors found no significant changes in sleep structure of MCI patients when compared to age-matched controls, which might be due in part to the small and heterogeneous sample employed in that study. In contrast, a recent meta-analysis study has revealed that 17-59% of MCI patients reported sleep problems [27]. This controversy highlights the need of determining whether elders with MCI also exhibit objective alterations in sleep patterns, which may have implications for early diagnosis of AD and preliminary therapeutic management of sleep complaints in these patients.

The presence of the allele ε4 in the apolipoprotein (ApoE) gene is the major genetic risk factor for AD [28], and it predicts the rate of cognitive decline in elderly subjects [29]. Previous studies have reported significant changes of melatonin levels in AD patients with different ApoE polymorphisms [30], suggesting a potential relationship between hormonal regulation of the sleep-wake cycle and ApoE car-
rier status in AD. Recent studies, based on either actigraph recordings [31] or self-reports [32], have tried to relate the ApoE e4 genotype to sleep disturbances in AD patients, but results appeared contradictory. Consequently, further PSG studies in MCI patients are clearly needed to verify the combined value of ApoE e4 genotype and sleep patterns as an early marker of AD.

The objective of the present study is threefold. First of all, to determine whether sleep patterns differ between healthy old persons and elders with MCI. Secondly, to evaluate the potential impact of ApoE e4 genotype on sleep structure of MCI patients. And thirdly, to examine whether there is a relationship between memory performance and sleep/wake parameters in healthy elders and MCI patients.

**MATERIALS AND METHODS**

**Subjects**

Twenty-five MCI patients (7 females, mean age: 70.5 ± 6.8 yr) and 25 healthy old (HO) volunteers (13 females, mean age: 67.1 ± 5.3 yr) were recruited from the Dementia Unit of the University Hospital Virgen del Rocio and the local community, respectively. HO participants and MCI patients were included in the study on a first-come, first-served basis. Demographic profiles of the two groups are shown in Table 1. All subjects provided written informed consent prior to their participation in the study. Experimental protocols were previously approved by the Ethical Committee for Clinical Investigations at the University Hospital Virgen del Rocio, and the Ethical Committee for Human Research at the University Pablo de Olavide. Research procedures were conducted in accordance with the Helsinki Declaration.

The diagnosis of amnestic MCI was based on consensus criteria [33]: (i) subjective memory complaints corroborated by the informant, (ii) objective memory decline evidenced by neuropsychological tests (scorings 1.5 standard deviations below the age-appropriate mean). Immediate and delayed recall were assessed with the Spanish version of the Logical Memory subtest extracted from the Wechsler Memory Scale-Third Edition (WMS-III), (iii) global score of 0.5 (questionable dementia) in the clinical dementia rating (CDR), (iv) normal independence function, judged both clinically and by means of the interview for deterioration in daily living activities (IDDD) validated in the Spanish population [34], and (v) not meeting DSM-IV criteria for dementia. Global cognitive status was assessed with the Mini Mental State Examination (MMSE). Depression was excluded by the Geriatric Depression Scale (GDS) (shorter version) [35]. The GDS cut off was ≤ 5. MCI patients were not taking cholinesterase inhibitors at the time of recruiting. Inclusion criteria for HO subjects were (i) absence of objective memory decline as revealed by the same neuropsychological tests used with MCI patients, (ii) CDR global score of 0 (no dementia), and (iii) normal independent function judged clinically and by means of the IDDD. Depression was excluded by the GDS using the same criteria as for MCI patients.

All participants underwent a neurological exploration to rule out obvious neurological diseases. Furthermore, cerebral magnetic resonance imaging (MRI) was performed in both HO subjects and MCI patients to determine structural lesions such as territorial cerebral infarction, brain tumor, hippocampal sclerosis, and vascular malformation. Those with periventricular or deep white matter ischemia, as revealed by scorings above 1 on the Fazekas ischemic scale [36], were excluded from the study. Participants reporting a history of neurological, psychiatric disorders, and/or major medical illness (chronic renal, hepatic, pulmonary or endocrine) were also excluded. The use of medication known to affect the sleep-wake cycle (benzodiazepines, tricyclic, or serotonin reuptake inhibitor antidepressants) was also considered cause for exclusion, both in HO and MCI patients. The absence of secondary causes of cognitive deficits was assessed by laboratory tests including complete blood count, blood chemistry, vitamin B12/folate, and thyroid function tests. Participants reported no complaints of sleep-disordered breathing, movement disorders during sleep or unusual sleep schedules (i.e.,

**Table 1. Demographic and Cognitive Profile of HO Subjects and MCI Patients**

<table>
<thead>
<tr>
<th></th>
<th>HO (n = 25)</th>
<th>MCI (n = 25)</th>
<th>P &lt;</th>
<th>MCI e4 - (n = 14)</th>
<th>MCI e4 + (n = 11)</th>
<th>p &lt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, yr</td>
<td>67.1 ± 5.3</td>
<td>70.5 ± 6.8</td>
<td>0.06</td>
<td>67.4 ± 6.9</td>
<td>74.4 ± 4.5</td>
<td>0.01</td>
</tr>
<tr>
<td>Gender (F / M)</td>
<td>13 / 12</td>
<td>7 / 18</td>
<td>0.08</td>
<td>3 / 11</td>
<td>4 / 7</td>
<td>0.4</td>
</tr>
<tr>
<td>Education, yr</td>
<td>8.5 ± 2.6</td>
<td>9.5 ± 5.3</td>
<td>0.4</td>
<td>8.0 ± 5.1</td>
<td>11.4 ± 5.1</td>
<td>0.1</td>
</tr>
<tr>
<td>CDR (sum of boxes)</td>
<td>0</td>
<td>0.5</td>
<td>N/A</td>
<td>0.5</td>
<td>0.5</td>
<td>N/A</td>
</tr>
<tr>
<td>MMSE</td>
<td>28.1 ± 1.3</td>
<td>26.7 ± 2.4</td>
<td>0.02</td>
<td>27.3 ± 2.4</td>
<td>26.0 ± 2.5</td>
<td>0.2</td>
</tr>
<tr>
<td>ESS</td>
<td>5 ± 2.4</td>
<td>5.4 ± 3.5</td>
<td>0.6</td>
<td>5.1 ± 2.6</td>
<td>5.8 ± 4.6</td>
<td>0.6</td>
</tr>
<tr>
<td>Immediate recall</td>
<td>13.9 ± 2.9</td>
<td>9.0 ± 2.9</td>
<td>10**</td>
<td>10.3 ± 2.9</td>
<td>7.4 ± 1.9</td>
<td>0.008*</td>
</tr>
<tr>
<td>Delayed recall</td>
<td>13.0 ± 2.9</td>
<td>6.1 ± 3.9</td>
<td>10**</td>
<td>8.1 ± 3.6</td>
<td>3.6 ± 2.6</td>
<td>0.002*</td>
</tr>
</tbody>
</table>

HO (healthy old); MCI (mild cognitive impairment); e4 (ApoE e4 non-carriers); e4+ (ApoE e4 carriers). Results are expressed as mean ± SD (standard deviation). F / M (female / male). CDR (Clinical Dementia Rating). CDR = 0 no dementia, CDR = 0.5 questionable or very mild dementia. ESS (Epworth Sleepiness Scale) scores ranged from 0 to 24, the cut off to suspect pathological daytime sleepiness was set in 10. Gender differences were assessed by the chi-square test. *p-value obtained after applying the Sidak-Bonferroni correction. N/A (not applicable).
Statistical analyses included the following dependent variable: sleep stage by the time (in hours) spent in that sleep stage. This index resulted from dividing the number of arousals in a sleep stage by the time (in hours) spent in that sleep stage. The Epworth Sleepiness Scale (ESS) was administered to HO subjects and MCI patients to establish the level of subjective daytime sleepiness [37].

**Apolipoprotein E Genotyping**

Genomic DNA was extracted from peripheral blood using short proteinase K digestion. ApoE genotype was determined by conventional PCR (polymerase chain reaction) methods, using protocols previously described [38].

**Polysomnographic Recordings**

Participants were admitted to the laboratory at 17:00h for electrode application. Time of lights-out was based on the individual’s habitual bedtime, and confirmed by bed partners-caregivers. All subjects were instructed to follow their normal daily routine, take their usual meals, and to refrain from the intake of alcohol for 24 h prior to the study. The day of the experiment, subjects were not allowed to nap.

The PSG protocol included simultaneous recordings of electroencephalography (EEG), vertical and horizontal electrooculography (EOG), and electromyography (EMG) of submental muscles. All these physiological measures are required for sleep staging purposes. Respiratory measures were not collected because all participants reported no complaints of sleep-disordered breathing, corroborated by their bed partners, and ESS scorings below the cut-off for suspecting sleep disorders associated with excessive daytime sleepiness. EEG was recorded from scalp locations referred to linked-mastoids. Electrode-scapal impedance was kept below 5 KΩ. Vertical and horizontal EOG recordings were performed with pairs of electrodes placed above and below the left eye, and 1 cm apart from the outer canthus of each eye, respectively. EOG was recorded from a pair of electrodes placed on chin muscles (3 cm apart). All recordings were performed with gold cup, 10 mm diameter electrodes (Grass, Inc.) filled with electrolytic cream, and attached with either surgical tape (face placements) or collodion (scalp placements). PSG recordings were performed in a sound-attenuated bedroom under infrared video-controlled supervision.

All electrophysiological measures were amplified (BrainAmp MR, Brain Vision®), filtered (0.1-100 Hz band-pass), digitized (250 Hz, 16-bit resolution), and stored in digital format for off-line analysis. Scoring of sleep stages (20-seconds EEG epochs) was conducted blindly by a trained technician according to standard guidelines [39], based on the following montage: Fp1, Fp2, F3, F4, Fz, C3, C4, Cz, P3, P4, Pz, O1, O2, EMG and EOG. This electrode montage allowed us to reliably determine EEG features intrinsic to each sleep stage as well as EEG arousals. Scoring criteria for EEG arousals were taken from the American Sleep Disorders Association report [40], and the level of sleep disruption was determined with the arousal index (AI). This index resulted from dividing the number of arousals in a sleep stage by the time (in hours) spent in that sleep stage. Statistical analyses included the following dependent variables: Total sleep time (TST, time spent in sleep stages), percent of SWS, REM, wake after sleep onset (WASO, wake from the sleep onset to the last sleep epoch before the wake-up time), and the AI for SWS and REM sleep.

**Statistical Analysis**

A logarithmic transformation was applied to non-normally distributed dependent variables to approach normal distribution. Gender differences between groups (HO versus MCI; MCI ε4 non-carriers versus MCI ε4 carriers) were evaluated by applying the chi-square test, whereas the remaining demographic and cognitive variables were assessed with the Student T-test (Table 1). The Sidak-Bonferroni procedure was applied to correct for multiple comparisons.

A multivariate analysis of covariance (MANCOVA) was computed to study group differences in sleep/wake parameters introducing age as the only covariate, since years of education and levels of daytime sleepiness (ESS scorings) did not reach statistical significance. The effect on each dependent variable was evaluated with post-hoc analyses derived from analyses of covariance (ANCOVAs).

Multivariate regression (MVAR) analyses were used to determine whether selected sleep/wake variables predicted memory performance (immediate and/or delayed recall) in any of the groups (HO, MCI, MCI ε4 carriers, and MCI ε4 non-carriers). Next, we performed a multiple regression analysis with each single sleep/wake variable by eliminating the amount of the variation explained by age to establish if that sleep/wake variable significantly predicted memory performance. If the sleep/wake predictor reached significance in at least one of the two groups (HO and/or MCI; MCI ε4-carriers and/or MCI ε4 non-carriers), then we tested if the relationship between memory performance and the sleep/wake parameter differed between the two groups (as revealed by differences between regression slopes). Statistical significance was defined as \(p<0.05\). All statistical analyses were performed with SPSS v. 15 (SPSS Inc., Chicago, IL).

**RESULTS**

**Demographic Variables**

HO subjects and MCI patients showed a similar demographic profile (Table 1). Differences between MCI ε4 carriers and non-carriers were only evident for age \((p<0.01)\). The presence of ApoE ε4 has been associated with a major risk of AD in MCI patients [28,41]. In our study, 11 MCI patients were ApoE ε4 heterozygote \((ε3/ε4, n = 9; ε2/ε4, n = 2)\) and the remaining 14 MCI patients were ε4 non-carriers \((ε3/ε3, n = 12; ε2/ε3, n = 2)\). Overall, the prevalence of the ApoE ε4 allele was 3.6-fold greater in MCI patients than in HO, and it was present in 44% of our MCI sample in contrast to the 12% observed in HO subjects. Distribution of the ApoE ε4 genotype in our MCI sample resulted from chance; the presence of ApoE ε4 allele was not employed as an inclusion criterion during the recruiting process.

**Memory Function and Sleep/Wake Variables**

MCI patients showed a significant memory decline in immediate \((p<10^{-6})\) and delayed recall \((p<10^{-8})\) when compared to HO (Table 1). Memory loss was further aggravated in MCI ε4 carriers when compared to non-carriers for the two memory tests (immediate, \(p<0.008\); delayed, \(p<0.002\)).
The same pattern of results (HO>MCI-ε4>MCI-ε4+) was corroborated when memory performance in the two tests was compared among the three groups (Pillai's trace, F_{1,42}=10.6, p<10^{-5}, post hoc analyses, p<0.03). Although differences in memory performance between MCI ε4 carriers and non-carriers were significantly reduced after accounting for age effects, they still remained significant (Pillai's trace for group F_{4,92}=9.2, P<10^{-5}; Pillai's trace for age F_{2,45}=7.4, p<0.002).

Sleep patterns of our HO sample are in agreement with those previously reported [42, 43]. Table 2 summarizes sleep/wake measures in HO and MCI patients, and in MCI ε4 carriers and non-carriers. Age was introduced as a covariate due to significant correlations with sleep/wake parameters (Pillai's trace, F_{6,42}=3.2, p<0.03). No significant relationship was found between ESS scorings and sleep/wake parameters, indirectly supporting the absence of sleep-related breathing disorders associated with excessive daytime sleepiness in our study population.

The MANCOVA yielded significant group differences in sleep/wake parameters (Pillai's trace, F_{6,42}=9.8, p<0.003). We further found that REM sleep was shortened by approximately 5% in MCI when compared to HO (F_{1,47}=9.8, p<0.007). Although REM reduction was even more prominent in MCI ε4 carriers than in non-carriers (F_{1,23}=6.2, p<0.02), this effect didn't reach significance. Fig. (1) shows differences in sleep-wake parameters between HO and MCI, and between MCI ε4 carriers and non-carriers.

Relationships between Sleep/Wake Parameters and Cognition

Neither SWS arousals nor REM percentage predicted cognitive decline in HO and/or MCI patients. As REM sleep was significantly shortened in MCI ε4 carriers when compared to non-carriers (p<0.02, uncorrected for multiple comparisons), we evaluated whether this reduction predicted memory performance in both subgroups of MCI patients. This analysis confirmed a positive relationship between REM sleep and memory impairment for immediate recall, but only for MCI ε4 non-carriers (F_{2,11} = 6.3, p<0.01, adjusted R squared = 0.45, beta = 0.63, p<0.01). Comparison of regression slopes between MCI ε4 carriers and non-carriers confirmed significant differences between the two groups (F_{4,20} = 7.1, p<0.001, adjusted R squared = 0.5, beta for the interaction term = 1.3, p<0.006). Fig. (2) illustrates the regression slopes for MCI ε4 carriers and non-carriers after accounting for age.

DISCUSSION

The present study provides the first evidence of impaired sleep patterns in elders with MCI. MCI patients showed reduced amounts of REM sleep and increased fragmentation of SWS. REM sleep deficits in MCI patients were aggravated by the presence of the ApoE ε4 genotype, but the relationship between REM shortening and memory performance was only evident in MCI ε4 non-carriers. Overall, these findings suggest that sleep patterns are affected years before AD diagnosis, which may have implications for early detection of dementia and/or therapeutic management of sleep disturbances in MCI patients.

Pioneering evidence has shown that wake duration correlates positively with accumulation of amyloid-beta (Abeta) and with increased levels of orexin [44], a neuropeptide synthesized in neurons of the lateral hypothalamic area which projects to and inhibits the VLPO nucleus, critical for the maintenance of wakefulness [45]. We hypothesize that sleep loss (increased wake duration) caused by SWS fragmentation in MCI patients might contribute to facilitate molecular pathways involved in the production of toxic amyloid oligomers for years in a silent but irreversible manner. Two lines of evidence support this hypothesis. Firstly, sleep loss caused by excessive sleep fragmentation has been previously reported in mild-to-moderate AD patients [15, 16], and it is aggravated as a function of dementia severity [14]. And secondly, recent studies suggest that aging.

Table 2.  Sleep/Wake Patterns in HO Subjects and MCI Patients

<table>
<thead>
<tr>
<th></th>
<th>HO (n=25)</th>
<th>MCI (n=25)</th>
<th>p &lt;</th>
<th>MCI ε4 (n=14)</th>
<th>MCI ε4+ (n=11)</th>
<th>p &lt;</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Sleep Parameters</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TST (min.)</td>
<td>397.9 ± 30.1</td>
<td>369.8 ± 53.8</td>
<td>0.2</td>
<td>375.8 ± 34.2</td>
<td>362.2 ± 72.9</td>
<td>0.6</td>
</tr>
<tr>
<td>SWS (%)</td>
<td>24.0 ± 6.9</td>
<td>22.0 ± 11.7</td>
<td>0.7</td>
<td>21.4 ± 7.1</td>
<td>22.8 ± 16.2</td>
<td>0.8</td>
</tr>
<tr>
<td>REM (%)</td>
<td>14.7 ± 3.7</td>
<td>10.1 ± 5.4</td>
<td>0.007*</td>
<td>12.3 ± 4.3</td>
<td>7.4 ± 5.5</td>
<td>0.02*</td>
</tr>
<tr>
<td><strong>Wake Parameters</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>WASO (%)</td>
<td>12.8 ± 5.7</td>
<td>18.0 ± 11.4</td>
<td>0.2</td>
<td>16.4 ± 6.6</td>
<td>19.9 ± 15.7</td>
<td>0.6</td>
</tr>
<tr>
<td>AI SWS</td>
<td>0.09 ± 0.11</td>
<td>0.19 ± 0.10</td>
<td>0.003*</td>
<td>0.20 ± 0.11</td>
<td>0.17 ± 0.08</td>
<td>0.5</td>
</tr>
<tr>
<td>AI REM</td>
<td>0.17 ± 0.13</td>
<td>0.16 ± 0.14</td>
<td>0.9</td>
<td>0.17 ± 0.16</td>
<td>0.14 ± 0.12</td>
<td>0.3</td>
</tr>
</tbody>
</table>

HO (healthy old); MCI (mild cognitive impairment); ε4 (ApoE ε4 non-carriers); ε4+ (ApoE ε4 carriers). Results are expressed as mean ± SD (standard deviation). TST (total sleep time); WASO (wakeup after sleep onset); SWS (sleep-wake sleep); REM (rapid eye movement sleep); AI (arousal index); *p < 0.05 (MANCOVA with age as covariate). *p-value after applying the Sidak-Bonferroni correction.
Significant differences in sleep patterns between HO subjects and MCI patients, and between MCI ε4 carriers and non-carriers for the percentage of REM sleep and the amount of arousals during SWS. Sleep arousals were determined with the arousal index (AI). SWS (slow-wave sleep); REM (rapid eye movement sleep).

Fig. (1). Significant differences in sleep patterns between HO subjects and MCI patients, and between MCI ε4 carriers and non-carriers for the percentage of REM sleep and the amount of arousals during SWS. Sleep arousals were determined with the arousal index (AI). SWS (slow-wave sleep); REM (rapid eye movement sleep).

Fig. (2). Scatter plot of partial correlation (after eliminating variations explained by age) displaying a significant positive correlation between REM percentage and immediate memory performance in MCI ε4 carriers (triangles) and no significant correlation between the same variables in MCI ε4 non-carriers (circles).
MCI ε4-carriers being more affected than non-carriers, which could be used in combination with biological and neuroimaging markers of early neurodegeneration to better determine boundaries between healthy aging and the prodromal stage of AD.

Previous studies have explored potential links between changes in sleep patterns and cognitive decline in AD patients. For instance, Moe and collaborators [16] reported significant correlations between cognitive and PSG measures (WASO, SWS, and REM) supporting the predictive value of this relationship in AD patients with different levels of disease severity. Sleep propensity has also shown to be inversely related to lower scores in global cognitive status, psychomotor, and memory function in mild to moderate AD patients [57]. In addition, increased REM sleep was found to correlate positively with enhanced cognitive function in patients with mild to moderate AD after donepezil administration [68]. Relationships between sleep patterns and cognitive decline in MCI patients had not been explored to date. Our study showed significant associations between immediate recall and REM sleep in MCI ε4 non-carriers, supporting the combined role of REM sleep together with memory performance as a potential early marker of AD. However, no significant relationship was found between delayed recall and REM sleep in MCI patients with or without the ApoE ε4 genotype. By using actigraph recordings, Yesavage and collaborators [31] also found significant relationships between deterioration of sleep parameters and lower MMSE scorings in AD ε4 non-carriers. However, it is unclear why impaired cognitive function and sleep disturbances are more frequently related to the lack of the ε4 allele in the ApoE gene. Previous evidence suggests that the most rapid cognitive decline occurs in AD patients with early onset ε4 non-carriers [69]. It may happen that our MCI sample included patients who will develop AD with both early and late onset as well as persons who will not progress to AD. This heterogeneity might indeed influence the impact of the ApoE ε4 genotype on cognitive decline and sleep disturbances. Further longitudinal studies are needed to better understand the combined role that ApoE ε4 and disturbed sleep patterns play in prodromal stages of AD.

Evidence suggests that prevalence of sleep disturbances in MCI is elevated [27,70, 71], although most of the studies have been based on interviews with the patients and/or the bed partner and on administration of different neuropsychiatric scales. To our knowledge, only one study has explored the presence of sleep disorders in MCI patients by using PSG recordings [12]. Authors concluded that sleep-related breathing disorders and sleep movement disorders are more prevalent in MCI patients. Unfortunately, our study cannot exclude the presence of sleep apnea and/or periodic limb movements on the basis of PSG recordings, which might partially confound our results. In our favour is the fact that all participants and their bed partners reported no related symptoms associated with these disorders during the neurologic exploration. Furthermore, subjective levels of daytime sleepiness were similar in both groups, individual ESS scorings were in all cases below the cut off for suspecting sleep disorders associated with excessive daytime sleepiness, and they were not significantly correlated with either cognitive or sleep/wake variables considered in our study. Nevertheless, further PSG studies including polygraphic respiratory and movement measures are needed to definitively confirm whether altered sleep patterns observed in MCI patients have a neurodegenerative basis or, in contrast, are due to sleep disorders exacerbated in healthy elders and MCI patients.

CONCLUSIONS
This study reveals altered sleep patterns in MCI patients, selectively aggravated for REM sleep in MCI ε4 carriers. These results in combination with biological and neuroimaging markers of early neurodegeneration might contribute to establish the boundaries between healthy and pathological aging in humans. Important questions for future research include how sleep impairments evolve along the continuum of healthy aging to severe AD, and how sleep markers could be of help for improving early diagnosis of AD and for managing sleep disorders during progression to AD. Further longitudinal studies are also required to validate the complex relationship among sleep, ApoE ε4 genotype, and early neurodegeneration.

ACKNOWLEDGMENTS
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CONFLICT OF INTEREST
None declared.

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