Apolipoprotein E and Alpha Brain Rhythms in Mild Cognitive Impairment: A Multicentric Electroencephalogram Study

Claudio Babiloni, PhD,[1,2] Luisa Benussi, MD,[2] Giuliano Binetti, MD,[2] Emanuele Cassetta, MD,[2] Gloria Dal Forno, MD[4] Claudio Del Percio, PhD,[1,2] Florinda Ferreri, MD,[3,4] Raffaele Ferri, MD,[5] Giovanni Frisoni, MD,[2,3] Roberta Ghidoni, PhD,[2] Carlo Miniussi, PhD,[2] Guido Rodriguez, MD,[6] Gian Luca Romani, PhD,[7,8] Rosanna Squitti, PhD,[3] Maria Carla Ventriglia, PhD,[3] and Paolo M. Rossini MD[2–4]

***Objective*: Relationships between the apolipoprotein E ε4 allele and electroencephalographic (EEG) rhythmicity have been demonstrated in Alzheimer's disease (AD) patients but not in the preclinical stage prodromic to it, namely, mild cognitive impairment (MCI). The present multicentric EEG study tested the hypothesis that presence of ε4 affects sources of resting EEG rhythms in both MCI and AD subjects. *Methods*: We enrolled 89 MCI subjects (34.8% with ε4) and 103 AD patients (50.4% with ε4). Resting eyes-closed EEG data were recorded for all subjects. EEG rhythms of interest were delta (2—4Hz), theta (4–8Hz), alpha 1 (8–10.5Hz), alpha 2 (10.5–13Hz), beta 1 (13–20Hz), and beta 2 (20–30Hz). EEG cortical sources were estimated by low-resolution brain electromagnetic tomography. *Results*: Results showed that amplitude of alpha 1 and 2 sources in occipital, temporal, and limbic areas was lower in subjects carrying the ε4 allele than in those not carrying the ε4 allele ($p < 0.01$). This was true for both MCI and AD. For the first time to our knowledge, a relationship was shown between ApoE genotype and global neurophysiological phenotype (ie, cortical alpha rhythmicity) in a preclinical AD condition, MCI, in addition to clinically manifest AD. *Interpretation*: Such a demonstration motivates future genotype–EEG phenotype studies for the early prediction of AD conversion in individual MCI subjects.**

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Mild cognitive impairment (MCI) is characterized by selective memory impairment insufficient to meet criteria for a diagnosis of dementia.[1–3] This condition is considered as a prodromic stage of Alzheimer's disease (AD),[4–6] because a high rate of progression to AD has been clearly shown.[3,7,8] Annual conversion rate to AD is 0.2 to 3.9% in normal aging (with no MCI symptoms) and 6 to 25% in MCI subjects.[3,9] At the end of 6 years of observation, approximately 80% of MCI subjects develop AD.[10] Taken together, these data suggest the hypothesis that in most (yet not all) of cases, MCI is a transition state on a linear progression toward AD. According to such a hypothesis, early identification of MCI patients might be clinically crucial.[11,12]

In mild AD, electroencephalographic (EEG) rhythms differ from normal elderly (Nold) and vascular dementia subjects, AD patients being characterized by higher delta (0–3Hz) and lower parietooccipital alpha (8–12Hz).[13–17] Similarly, MCI subjects have shown increase of theta (4–7Hz) power[18–20] as well as decrease of alpha power,[15,18–22] when compared with Nold subjects. These EEG parameters have presented an intermediate magnitude in MCI with respect to Nold and dementia patients.[15,23,24]

Despite the converging evidence of abnormal cortical rhythms in MCI and AD, EEG analysis alone is unable to predict conversion of MCI to dementia. It is reasonable that additional biological parameters are needed for this purpose. In this regard, several studies have shown a strict relationship between apolipopro-

From the [1]Dipartimento di Fisiologia Umana e Farmacologia, Universitá "La Sapienza" Rome, Italy; [2]Associazione-IRCCS "San Giovanni di Dio-Futebenefratelli", Brescia, Italy; [3]Associazione Dipartimentodi Neuroscience, Ospedele Fatebenefratelli; Isola Tiberina, Rome, Italy; [4]Clinica Neuroligica Universitá "Campus Biomedico" Rome, Italy; [5]Department of Neurology, Oasi Institute for Research on Mental Retardation and Brain Aging (IRCCS), Troina, Italy; [6]Division of Clinical Neurophysiology, University of Genova, Italy; [7]Dipartimento di Scienze Cliniche e Bioimmagini and [8]Istitutodi di Tecnologie Avanzate Biiomediche (ITAB), Fondazione "Università Gabriele D'Annunzio," Chieti, Italy.

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Address correspondence to Dr Babiloni, Dipartimento di Fisiologia Umana e Farmacologia, Università degli Studi di Roma "La Sapienza," P.le Aldo Moro 5, 00185 Rome, Italy.
E-mail: claudio.babiloni@uniroma1.it

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tein E (ApoE) genotype and late-onset AD. The human ApoE gene has three alleles (ε2, ε3, ε4), ε3 accounting for the majority of the ApoE gene pool (approximately 70–80%), whereas ε4 and ε2 account for 10 to 15% and 5 to 10%, respectively.[25] Previous epidemiological and genetic studies on ApoE have shown that (1) ApoE ε4 significantly increases the risk of developing AD[25–34]; (2) ApoE ε2 and ApoE ε3 are associated with a reduced Alzheimer's disease risk[32–36]; (3) association between ApoE ε4 and AD risk is stronger in men than in women[33,37]; (4) ApoE ε4 is the only genetic risk factor associated with MCI[38]; and (5) ApoE ε4 predisposes to cognitive decline.[39,40]

ApoE ε4 is combined with different types of functional brain imaging abnormalities in preclinical as well as in clinically evident cognitive decline of dementia type.[41–43] Compared with patients with ApoE ε2 or ε3, AD carriers of ε4 allele had a reduction in neural metabolism and activity both at rest and after activation procedures in temporal, parietal, limbic, and prefrontal areas.[44–48] Similarly, ε4 has been found to affect EEG rhythms in AD.[49–52] AD patients with ε4 had higher theta (4.1–7.3Hz) and lower beta (14.2–20Hz) power than AD patients with ε2 or ε3.[51] Furthermore, compared with AD patients with ε2 or ε3, AD patients with ε4 showed: (1) higher theta and lower beta power at baseline recording[52]; and (2) higher delta (1.5–3.9Hz) and lower alpha (7.6–13.9Hz) power at the follow-up (3 years later).[52] Finally, ApoE ε4 has been associated with selective decrement of corticocortical functional connectivity in AD, as shown by a reduction of right and left temporoparietal, right temporofrontal, and left occipitoparietal EEG coherence at the alpha band (8–13Hz).[49]

To our knowledge, the ApoE ε4 effects on EEG rhythmicity in the preclinical stage of AD, namely MCI, have not been previously investigated. The present EEG study tested the hypothesis that the ApoE ε4 allele affects cortical sources of resting EEG rhythms in MCI.

Materials and Methods

Part of the procedures (EEG recordings and low-resolution brain electromagnetic tomography [LORETA] analysis) pertinent to this study as well as a description of the potential meaning of cortical rhythms in aging have been extensively described in two recent papers.[17,53] However, it should be stressed that the aims of the previous and current studies are totally different. The previous studies aimed at analyzing (1) the distributed EEG sources specific to mild AD as compared with vascular dementia (VaD) or normal aging[17] and (2) the distributed EEG sources across physiological aging.[53] In contrast, this study focused on the effects of the ApoE ε4 allele on EEG rhythmicity in the MCI subjects. We apologize for some repetitions in the description of the present procedures with respect to those reported in the previous articles using similar EEG methodological approach. This was done to stress the fact that already validated methodological procedures were used.

Subjects

Patients population included 89 MCI subjects and 103 AD patients, whereas the control group included 86 cognitively Nold individuals. Local institutional ethics committees approved the study. All experiments were performed with the informed and overt consent of each participant or caregiver, in line with the Code of Ethics of the World Medical Association (Declaration of Helsinki) and the standards established by the authors' institutional review board.

Diagnostic Criteria

Probable AD was diagnosed according to National Institute of Neurological and Communication Disorders Alzheimer's Disease and Related Disorders Association[54] and Diagnostic Statistical Manual IV criteria. All recruited AD patients underwent general medical, neurological, and psychiatric assessments. Patients were also rated with several standardized screening, diagnostic, and severity instruments that included Mini-Mental State Examination (MMSE),[55] Clinical Dementia Rating Scale (CDR),[56] Geriatric Depression Scale (GDS),[57] Hachinski Ischemic Scale (HIS),[58] and the Instrumental Activities of Daily Living scale (IADL).[59] Neuroimaging diagnostic procedures (computed tomography or magnetic resonance imaging) and complete blood chemistries tests were conducted to exclude other causes of progressive or reversible dementias, to have a homogenous AD patient sample. Exclusion criteria included the presence of symptoms suggestive of (1) frontotemporal dementia, (2) vascular dementia, (3) extrapyramidal syndromes, (4) reversible dementias, (5) fluctuations in cognitive performance as well as of movement disorders and visual hallucinations suggestive of a possible Lewy body dementia. Of note, antidepressant and/or antihypertensive were suspended for 24 to 48 hours before EEG recordings.

Inclusion and exclusion criteria for MCI diagnosis aimed at selecting elderly persons with objective cognitive deficits, especially in the memory domain, who did not meet criteria for dementia or AD.[1,3,60–64] Inclusion criteria for MCI were represented by (1) objective memory impairment on neuropsychological evaluation, as defined by performances 1.5 or higher standard deviation below age and education-matched controls; (2) full autonomy in the activities of daily living as documented by dedicated tests and by history and evidence of independent living; and (3) a CDR of 0.5. Exclusion criteria for MCI were (1) AD, as diagnosed by the procedures described above; (2) evidence of concomitant dementia such as frontotemporal, vascular dementia, reversible dementias, fluctuations in cognitive performance, and/or features of mixed dementias; (3) evidence of concomitant extrapyramidal symptoms; (4) clinical and indirect evidence of depression as shown by GDS scores greater than 14; (5) other psychiatric diseases, epilepsy, drug addiction, alcohol dependence, and use of psychoactive drugs or drugs interfering with brain cognitive functions including acetylcholinesterase inhibitors; and (6) current or previous systemic diseases (including diabetes mellitus) or traumatic brain injuries. Subjects' blood examination for the evaluation of ApoE was per-

Table 1. Demographic and Neuropsychological Data of Participants

Characteristic	Nold	MCI	AD
N	86	89	103
Age (yr)	70.1 (±0.6 SE)	70.8 (±0.9 SE)	75.1 (±0.8 SE)
Sex (F/M)	53/33	53/36	84/19
MMSE	28.1 (±0.1 SE)	25.7 (±0.3 SE)	20.6 (±0.3 SE)
Education (yr)	9.6 (±0.4 SE)	7.6 (±0.4 SE)	6.3 (±0.4 SE)

Nold = normal elderly; MCI = mild cognitive impairment; AD = Alzheimer's disease; SE = standard error; MMSE = Mini-Mental State Examination.

formed on MCI subjects as well. On the basis of such genotyping, the AD group was subdivided in two genetic subgroups: AD carriers of the ApoE ε4 allele (52 subjects) and AD noncarriers of the ApoE ε4 allele (51 subjects). As the AD group, the MCI group was subdivided in two genetic subgroups: MCI carriers of the ApoE ε4 allele (31 subjects) and MCI noncarriers of the ApoE ε4 allele (58 subjects). Besides such differences in ApoE genotyping, these subgroups did not significantly differ in demographic or in clinical parameters.

To preliminarily ascertain that the selected AD patients and MCI subjects indeed presented the pattern of EEG rhythmic activity that typically is combined with the disease and cognitive impairment, we also recruited a control group of healthy elderly subjects (Nold), mostly among nonconsanguineous patients' relatives. All Nold subjects underwent physical and neurological examinations as well as cognitive screening. Subjects affected by chronic systemic illnesses, subjects receiving psychoactive drugs, and subjects with a history of present or previous neurological or psychiatric disease were excluded. All Nold subjects had a GDS score less than 14.

Table 1 summarizes the relevant demographic and clinical data of participants. As expected, women were overrepresented in the AD group. Because there is no previous evidence of sex-specific effects on EEG rhythms, it was felt that this would not interfere with the results. Table 2 reports the relevant demographic and clinical parameters of participants subdivided in two genetic subgroups: ApoE ε4 allele MCI/AD carriers and noncarriers, respectively. Age and education were used as covariates in the statistical evaluation of the cortical sources of EEG rhythms, to remove possible confounding effects.

Apolipoprotein E Genotyping

Genomic DNA was extracted from whole-blood samples of AD patients and MCI subjects. ApoE genotype was determined using standard methods.[65]

Electroencephalogram Recordings

EEG was recorded in waking-rest conditions (eyes-closed, 0.3–70Hz bandpass) from 19 scalp electrodes positioned according to the International 10-20 System (ie, Fp1, Fp2, F7, F3, Fz, F4, F8, T3, C3, Cz, C4, T4, T5, P3, Pz, P4, T6, O1, O2). A specific reference electrode was not imposed, because preliminary data analysis and LORETA source analysis were conducted after EEG data were re-referenced to a common average reference. To monitor eye movements, we also collected the electrooculogram (0.3–70Hz bandpass). All data were digitized in continuous recording mode (128–256Hz sampling rate). In all subjects, EEG recordings were performed at about lunch time. State of vigilance was controlled by on-line visual inspection of EEG traces during recording session and subjects' drowsiness was avoided by verbal warnings. No patient received medications that could influence EEG rhythms such as antidepressant or benzodiazepines. Of note, EEG recordings lasting 5 minutes allowed the comparison of the results with several previous AD studies using either EEG recording periods shorter than 5 minutes[17,66–71] or shorter than 1 minute.[13,14] Longer resting EEG recordings in AD patients would have reduced data variability but would have increased the possibility of EEG rhythmic oscillations slowing because of reduced vigilance and arousal.

EEG data were analyzed and fragmented off-line in consecutive epochs of 2 seconds. For standardization purposes, preliminary data analysis was centralized in one research unit. The EEG epochs with ocular, muscular, and other types of artifact were preliminary identified by a computerized automatic procedure. EEG epochs with ocular artifacts (<15% of the total) were corrected by an autoregressive method.[72] Two independent experimenters manually confirmed the EEG segments accepted for further analysis.

Spectral Analysis of the Electroencephalogram Data

A digital Fast Fourier Transform (FFT)-based power spectrum analysis (Welch technique, Hanning windowing function, no phase shift) was conducted by computing power density of EEG rhythms with 0.5Hz frequency resolution. The following frequency bands were studied: delta (2–4Hz), theta (4–8Hz), alpha 1 (8–10.5Hz), alpha 2 (10.5–13Hz), beta 1 (13–20Hz), and beta 2 (20–30Hz) in line with previous EEG studies on dementia[17,19,71–77] Of note, sharing of a frequency bin by two contiguous bands is a widely accepted procedure.[19,73,–91]

Choice of fixed EEG bands did not account for individual alpha frequency (IAF) peak, defined as the frequency associated with the strongest EEG power at the extended alpha range. This choice should not affect the results, because most of the subjects had IAF peaks within the alpha 1 band (8–10.5Hz). In particular, mean IAF peak was 9.3Hz (±0.1 standard error [SE]) in Nold subjects, 8.9Hz (±0.1 SE) in MCI subjects, and 8.3Hz (±0.1 SE) in AD patients. In the two MCI subgroups, the mean IAF peak was 9.2Hz (±0.2 SE) in MCI subjects not carrying the ε4 allele (MCI−) and 8.6Hz (±0.2 SE) in MCI carriers of the ε4 allele (MCI+).

Table 2. Demographic and Neuropsychological Data of the MCI and AD Subjects, Each Clustered in Two Genetic Subgroups: MCI/AD Not Carrying ApoE ε4 allele (ApoE ε4 noncarriers) and MCI/AD carrying the ε4 allele (ApoE ε4 carriers)

	MCI		AD	
Characteristic	ApoE ε4 Noncarriers	ApoE ε4 Carriers	ApoE ε4 Noncarriers	ApoE ε4 Carriers
N	58	31	51	52
Age (yr)	70.9 (±1.2 SE)	70.7 (±1.2 SE)	74.9 (±1.3 SE)	75.2 (±1.2 SE)
Sex (F/M)	35/23	18/13	44/7	40/12
MMSE	25.8 (±0.3 SE)	25.4 (±0.4 SE)	21.2 (±0.4 SE)	20 (±0.5 SE)
Education (yr)	7.3 (±0.5 SE)	8 (±0.8 SE)	6.7 (±0.2 SE)	5.8 (±0.4 SE)

MCI = mild cognitive impairment; AD = Alzheimer's disease; ApoE = apolipoprotein E; SE = standard error; MMSE = Mini-Mental State Examination.

In the two AD subgroups, the mean IAF peak was 8.4Hz (±0.2 standard error, SE) in AD− and 8.2Hz (±0.2 SE) in AD+. To control the effect of IAF on the EEG comparisons between these two subgroups, we used IAF peak as a covariate (together with age and education) for further statistics.

Cortical Source Analysis of the Electroencephalographic Rhythms by Low-Resolution Brain Electromagnetic Tomography

LORETA technique was used for EEG source analysis,[14,17,92–96] because its validity was tested with invasive depth EEG recordings,[97] positron emission tomography,[98] and simultaneous functional magnetic resonance imaging scans.[99,100] LORETA computes three-dimensional linear solutions (LORETA solutions) for EEG inverse problem within a three-shell spherical head model including scalp, skull, and brain compartments. Brain compartment is restricted to the cortical gray matter/hippocampus of a head model coregistered to Talairach probability brain atlas and digitized at the Brain Imaging Center of the Montreal Neurologic Institute.[101] This compartment includes 2,394 voxels (7mm resolution), each voxel containing an equivalent current dipole.

LORETA source analysis is reference-free, in that one obtains the same LORETA source distribution for EEG data referenced to any reference electrode including common average. Furthermore, LORETA can be used from data collected by low spatial sampling of the 10-20 system (19 electrodes) when cortical sources are estimated from resting EEG rhythms. Several previous studies have shown that these rhythms are generated by largely distributed cortical sources that can be accurately investigated by standard 10-20 system and LORETA.[17,102–112]

LORETA solutions consisted of voxel current density values able to predict EEG spectral power density at scalp electrodes. The LORETA data for each subject were normalized to unit global power (over all discrete frequencies and voxels), to reduce variance and thus enhance tomographic results. The general procedure fitted the LORETA solutions in a Gaussian distribution and reduced intersubject variability.[73,113] Of note, other methods of normalization using the principal component analysis are effective for estimating the subjective global factor scale of the EEG data.[114] These methods are not yet available in the LORETA package, so they were not used here.

Solutions of the EEG inverse problem are underdetermined and ill-conditioned when the number of spatial samples (electrodes) is lower than that of the unknown samples (current density at each voxel). To account for that, the cortical LORETA solutions predicting scalp EEG spectral power density were regularized to estimate distributed rather than discrete EEG source patterns.[92–94] Such spatial smoothing of the LORETA solutions (resolution in centimeters) can reliably take into account the slight change in the cortical volume (resolution in millimeters) present in AD subjects. In line with the low spatial resolution of the LORETA technique, we collapsed LORETA solutions at frontal, central, temporal, parietal, occipital, and limbic regions of the brain model coded into Talairach space. The Brodmann areas listed in Table 3 formed each of these regions of interest (ROIs).

Statistical Analysis of the Low-Resolution Brain Electromagnetic Tomography Solutions

Regional normalized LORETA solutions were used by analysis of variance (ANOVA) analysis, using subjects' age and education as covariates. Mauchly's test evaluated the sphericity assumption. Correction of the degrees of freedom was made with the Greenhouse–Geisser procedure. Duncan test was used for post hoc comparisons ($p < 0.05$). In particular, two ANOVA designs were addressed in this study. (1) A first control ANOVA analysis verified the sensitivity of the present methodological approach, namely, to estimate EEG source differences among Nold, MCI, and AD groups. This ANOVA analysis used Group (Nold, MCI, AD), Band (delta, theta, alpha 1, alpha 2, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic). The LORETA solutions of interest were those showing progressive changes in mean magnitude across Nold, MCI, and AD subjects (Nold→MCI→AD and Nold←MCI←AD). Furthermore, these solutions had to show linear correlations with MMSE score in all subjects as a single group (Bonferroni-corrected Pearson test, $p < 0.05$). (2) The second ANOVA evaluated the working hypothesis, namely, the existence of EEG source differences between MCI+ and AD+ versus MCI− and AD− groups. This ANOVA design used Group (MCI, AD), Genotype (presence or absence of ε4), Band (delta, theta, alpha 1, alpha 2, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic) as factors. The planned Duncan post hoc testing evaluated the

Table 3. LORETA Brodmann Areas into the Regions of Interest

Region	Area
Frontal	8, 9, 10, 11, 44, 45, 46, 47
Central	1, 2, 3, 4, 6
Parietal	5, 7, 30, 39, 40, 43
Temporal	20, 21, 22, 37, 38, 41, 42
Occipital	17, 18, 19
Limbic	31, 32, 33, 34, 35, 36

LORETA solutions were collapsed in frontal, central, parietal, temporal, occipital, and limbic ROIs.
LORETA = low-resolution brain electromagnetic tomography.

prediction of the working hypothesis. That prediction would be confirmed by the following LORETA patterns: Δ4 noncarriers ≠ ε4 carriers in both MCI and AD groups.

Results

Control Analysis

Grand average of LORETA solutions (ie, relative current density at cortical voxels) modeling the distributed EEG sources for delta, theta, alpha 1, alpha 2, beta 1, and beta 2 bands presented specific spatial features in Nold, MCI, and AD groups. Compared with the Nold group, AD patients showed an increase of widespread delta sources, along with a drastic reduction of parietooccipital alpha 1 sources. With respect to the Nold and AD groups, MCI subjects showed intermediate magnitude of alpha 1 sources and greater magnitude of alpha 2 sources.

These data were used for an ANOVA control analysis to test the hypothesis that MCI and AD subjects had typical EEG features as described by the literature. LORETA solutions (distributed EEG sources) showed a statistical ANOVA interaction ($F[50,6975] = 2.6$; $MSe = 0.6$; $p < 0.00001$) among the factors Group (Nold, MCI, AD), Band (delta, theta, alpha 1, alpha 2, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic). It was shown that parietal, occipital, temporal, and limbic alpha 1 sources had stronger amplitudes in Nold versus MCI ($p < 0.00001$) and in MCI versus AD ($p < 0.01$). Furthermore, the amplitude of the parietal delta source was stronger in AD than Nold ($p < 0.005$). Finally, parietal alpha 2 sources showed stronger amplitude in MCI than Nold ($p < 0.0001$) and AD ($p < 0.0001$). MMSE score correlated positively with parietal ($r = 0.21$, $p = 0.0003$; Bonferroni threshold to $p < 0.0125$), occipital ($r = 0.24$, $p = 0.00004$), temporal ($r = 0.21$, $p = 0.0003$), and limbic ($r = 0.23$, $p = 0.00006$) alpha 1 source intensities. These results demonstrated the existence of significant EEG source intensity differences among Nold, MCI, and AD groups.

Topography of the Electroencephalographic Cortical Sources Estimated by Low-Resolution Brain Electromagnetic Tomography in Apolipoprotein E ε4 Carriers and Noncarriers

For illustrative purposes, Figure 1 maps the grand average of the LORETA solutions (ie, relative current density at cortical voxels) modeling the distributed EEG sources for delta, theta, alpha 1, alpha 2, beta 1, and beta 2 bands in MCI and AD subjects not carrying the ε4 allele (MCI−, AD−) and in MCI and AD carriers of the ε4 allele (MCI+, AD+). Both MCI− and AD− groups presented alpha 1 sources with maximal values of the relative current density distributed in the parietooccipital regions. Delta, theta and alpha 2 sources had moderate relative current density values when compared with alpha 1 sources. Finally, beta 1 and beta 2 sources were characterized by the lowest relative current density values. In comparison, MCI+ and AD+ ε4 carriers showed a reduction of alpha 1 and alpha 2 sources intensities.

Statistical Analysis of the Electroencephalographic Cortical Sources Estimated by Low-Resolution Brain Electromagnetic Tomography Characterizing Apolipoprotein E ε4 Noncarriers with respect to the ApoE ε4 Carriers

ANOVA analysis for the evaluation of the working hypothesis showed a statistical interaction ($F[25,4,700] = 2.8$; $MSe = 0.5$; $p < 0.004$) among the factors Genotype (presence or absence of ApoE ε4 allele), Band (delta, theta, alpha 1, alpha 2, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic). Figure 2 shows the mean regional LORETA solutions relative to this statistical ANOVA interaction. In line with the working hypothesis, alpha 1 and alpha 2 sources in occipital, temporal, and limbic areas showed stronger amplitude in noncarriers compared to carriers of the ApoE ε4 ($p < 0.01$). This was true for both MCI and AD subjects, because there was no interaction with the factor Genotype (presence or absence of the ε4 allele). As a statistical trend, temporal theta source showed stronger amplitude in carriers compared with noncarriers of the ApoE ε4 ($p < 0.07$).

The ANOVA analysis also showed a statistical interaction among the factors Group (MCI, AD), Band (delta, theta, alpha 1, alpha 2, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic). Figure 3 shows mean regional LORETA solutions relative to this statistical ANOVA interaction. Alpha 1 (central, parietal, occipital, temporal and limbic areas) and alpha 2 (parietal and occipital areas) sources were stronger in MCI than AD ($p < 0.03$). Furthermore, delta (occipital and limbic areas) and theta (occipital areas) sources were stronger in AD compared with MCI ($p < 0.03$).

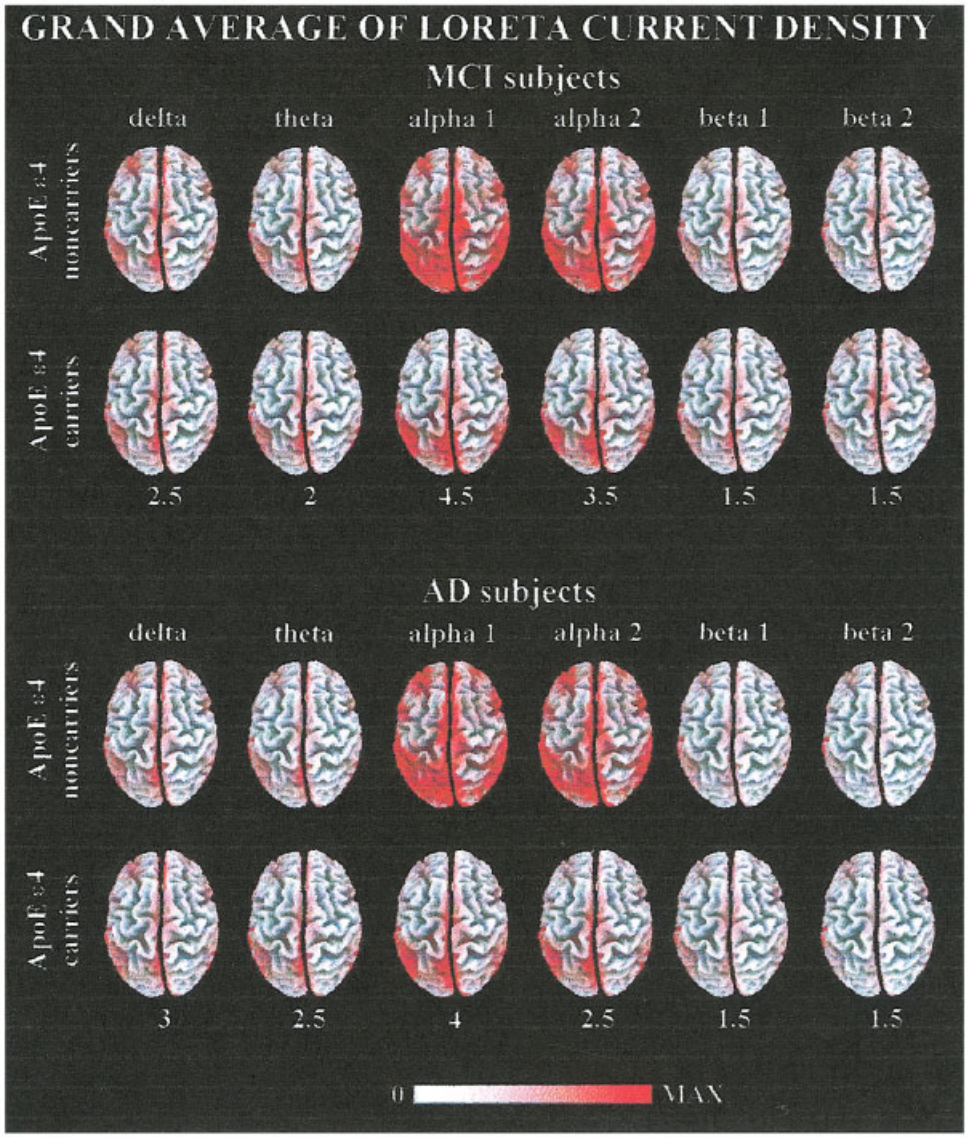

Fig 1. Grand average of low-resolution brain electromagnetic tomography (LORETA) solutions (ie, normalized relative current density at the cortical voxels) modeling the distributed EEG sources for delta (2–4Hz), theta (4–8Hz), alpha 1 (8–10.5Hz), alpha 2 (10.5–13Hz), beta 1 (13–20Hz), and beta 2 (20–30Hz) bands in mild cognitive impairment (MCI) and Alzheimer's disease (AD) groups, both subdivided in two genetic subgroups: MCI−/AD− not carrying the apolipoprotein E (ApoE) ε4 and MCI+/AD+ carrying the ε4 allele. The left side of the maps (top view) corresponds to the left hemisphere. Color scale: all power estimates were scaled based on the averaged maximum value (ie, alpha 1 power value of occipital region in MCI not carrying the ApoE ε4 allele). The maximal power value is reported under each column.

Discussion

Electroencephalographic Characteristics in Mild Cognitive Impairment and Alzheimer's Disease Subjects with ApoE ε4

Alpha sources in occipital, temporal, and limbic areas showed lower amplitude in MCI and AD subjects with ApoE ε4 compared with those not carrying ε4 ($p < 0.01$). Furthermore, temporal theta source showed a trend for stronger amplitude in ApoE ε4 carriers compared with noncarriers ($p < 0.07$). These results extend in the cortical spatial domain previous scalp EEG evidence in AD carrying ε4.[49–52] They complement previous evidence of reduced regional cerebral blood flow and/or glucose metabolism in temporal, parietal, limbic, and prefrontal areas of AD carrying ε4 when compared with AD noncarriers.[44–48] As a novelty, they demonstrate a similar trend for the preclinical stage of AD, namely, MCI. These results warrant further EEG investigations in normal subjects who carry the ε4 allele. Indeed, previous evidence has

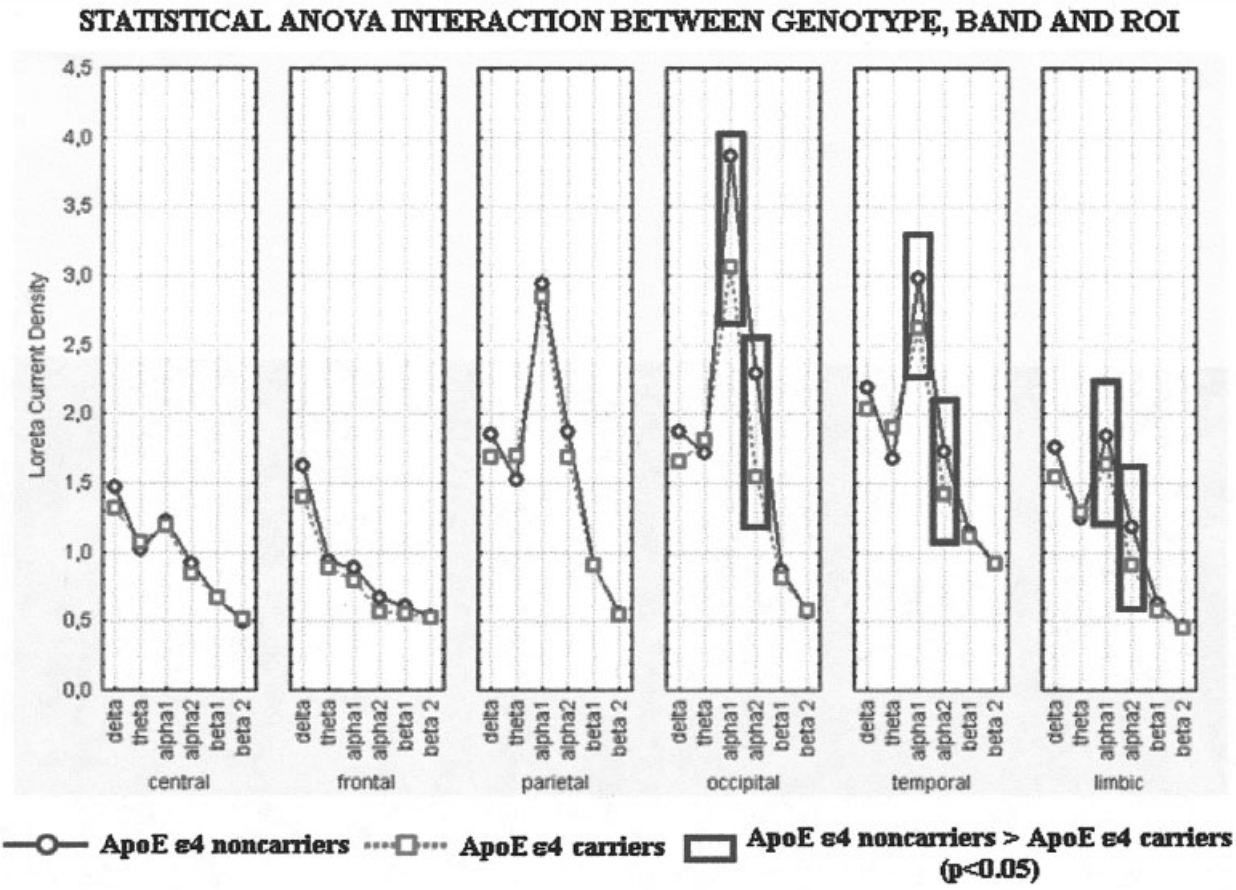

Fig 2. Regional low-resolution brain electromagnetic tomography (LORETA) solutions (mean across subjects) relative to a statistical analysis of variance (ANOVA) interaction among the factors Genotype (presence or absence of the ApoE ε4 allele), Band (delta, theta, alpha 1, alpha 2, beta 1, beta 2), and region of interest (ROI; central, frontal, parietal, occipital, temporal, limbic). This ANOVA design used the normalized relative current density values at ROI level as a dependent variable. Subjects' age, education, and individual alpha frequency peak (IAF) were used as covariates. Regional LORETA solutions modeled the electroencephalographic (EEG) relative power spectra as shown by a sort of "virtual" intracranial macroelectrodes located on the macrocortical regions of interest. (rectangles) *Cortical regions and frequency bands in which LORETA solutions presented statistically significant different values between subjects plus or minus for the apolipoprotein E (ApoE) ε4 allele* ($p < 0.05$, *planned Duncan post hoc testing). See Materials and Methods for further details.*

shown a reduction of resting posterior cortical activity in cognitively intact individuals with ApoE ε4 allele compared with subjects not carrying ApoE ε4 allele.[41,42]

The open question of this study is why alpha sources were lower in amplitude in MCI and AD ε4 carriers. We do not have a conclusive explanation at this early stage of research. As introductory basic aspects, it is well known that brain of an individual with AD exhibits extracellular plaques of aggregated β-amyloid protein (Abeta) and intracellular neurofibrillary tangles that contain hyperphosphorylated tau protein. In brain, degradation and clearance of Abeta would imply insulin-degrading enzyme, which is reduced by approximately 50% at hippocampus of ε4+ AD patients compared with ε4- patients and controls.[115] Furthermore, AD is characterized by a profound loss of basal forebrain cholinergic neurons that innervate hippocampus and neocortex.[116] Finally, alpha rhythms are mainly modulated by thalamocortical and corticocortical interactions.[117–119] Within extended alpha band (8–13Hz), low-band alpha would be mainly related to subject's global attentional readiness, whereas high-band alpha would reflect the engagement of specific neural channels for the elaboration of sensorimotor or semantic information.[80,85,86] At rest, the voltage of the alpha rhythms would be inversely correlated with the cortical excitability and level of attention processes depending on the novelty and importance of the stimulus.[80,85,86,117,120,121] For this reason, it has been suggested that the amplitude of alpha rhythms and corresponding cortical excitability reflect at least in part the time-varying inputs of forebrain cholinergic pathways.[122]

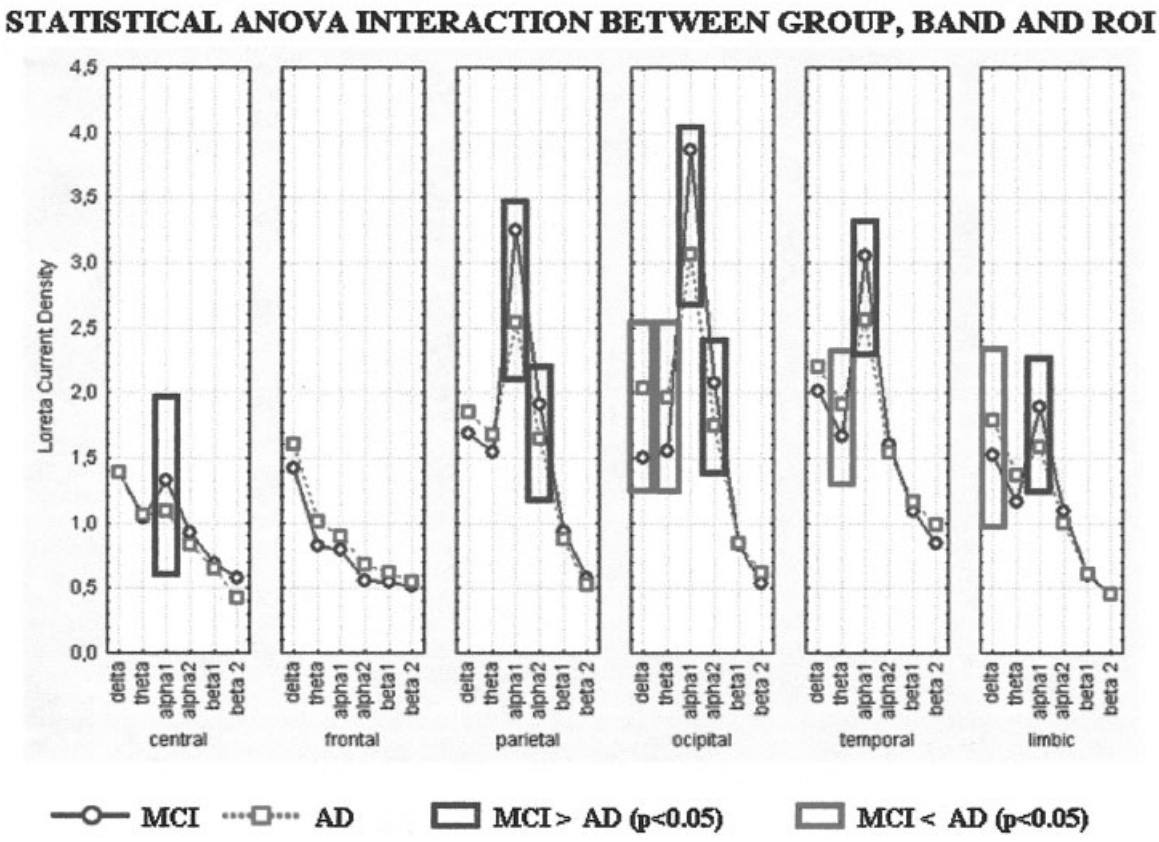

Fig 3. Regional low-resolution brain electromagnetic tomography (LORETA) solutions (mean across subjects) relative to a statistical analysis of variance (ANOVA) interaction among the factors Group (mild cognitive impairment [MCI], Alzheimer's disease [AD]), Band (delta, theta, alpha 1, alpha 2, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic). This ANOVA design used the normalized relative current density values at ROI level as a dependent variable. Subjects' age, education, and individual alpha frequency peak (IAF) were used as covariates. The rectangles indicate the cortical regions and frequency bands in which LORETA solutions presented statistically significant different values between MCI and AD groups ($p < 0.05$, *planned Duncan post hoc testing). See Materials and Methods for further details.*

Keeping in mind these data, it could be speculated that, in MCI and AD ε4 carriers, reduced degradation and clearance of Abeta would result in a great impairment of the cholinergic basal forebrain, hippocampal, and thalamocortical networks. As a consequence, there would be an increase of the excitatory activity of the cholinergic brainstem pathway,[123–125] which would desynchronize resting posterior alpha rhythms[126] and might enhance cortical excitability in AD patients.[127–132] Previous studies have indeed shown that resting EEG rhythms, including alpha, are lowered by experimental or clinical impairment of the cholinergic basal forebrain.[13–15,17,76,77,133–135] In contrast, brainstem cholinergic innervations of the thalamus are relatively spared in AD patients.[134–139]

Conclusions

The present EEG study evaluated cortical rhythms in MCI and AD subjects carrying or not a factor risk of dementia such as ApoE ε4 allele. Amplitude of alpha sources in occipital, temporal, and limbic areas was lower in subjects carrying the ApoE ε4 than noncarrier individuals ($p < 0.01$). This was true for both MCI and AD subjects. The results suggest that at group level, cortical delta and alpha rhythms of MCI subjects were qualitatively affected by similar pathological mechanisms impinging upon the generation of cortical rhythms in mild AD subjects. In this sense, they are in favor of the hypothesis that most of the MCI subjects with ApoE genetic risk suffer from a preclinical stage of AD. However, this might not be true for all MCI subjects with ApoE genetic risk. Furthermore, the cognitive decline might not have the same progression in all these subjects. Therefore, these results motivate a follow-up study testing whether MCI subjects with ApoE genetic risk convert to mild AD as a function of the baseline alteration of cortical delta and alpha rhythms.

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