8ª GIORNATA DELLO SPECIALIZZANDO IN NEUROLOGIA

Neurofisiologia dell'invecchiamento cerebrale

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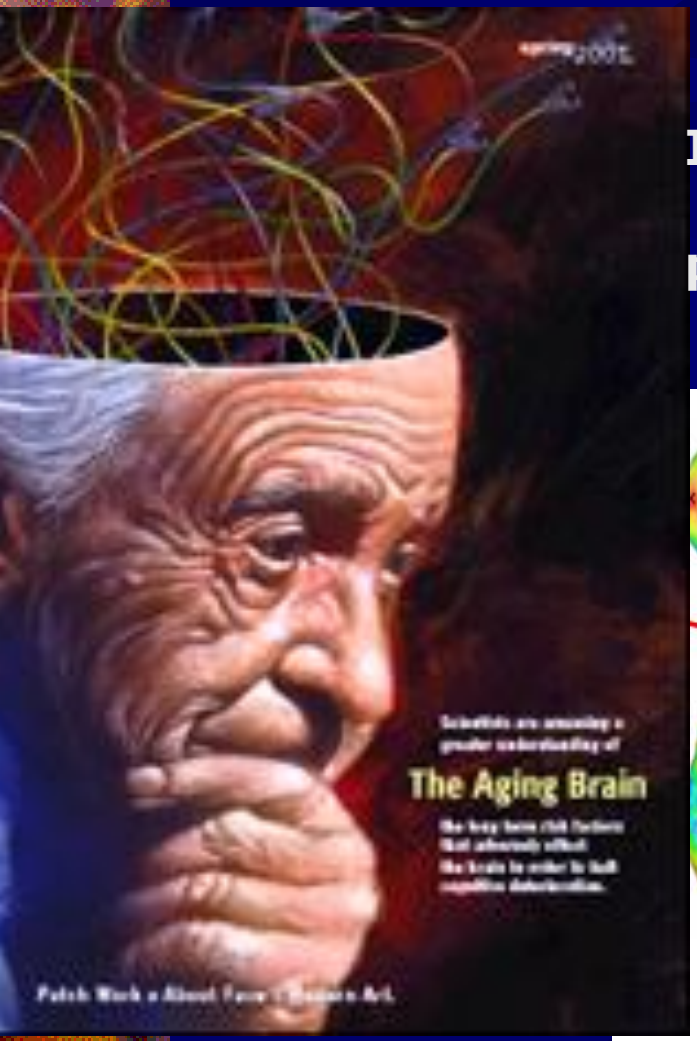
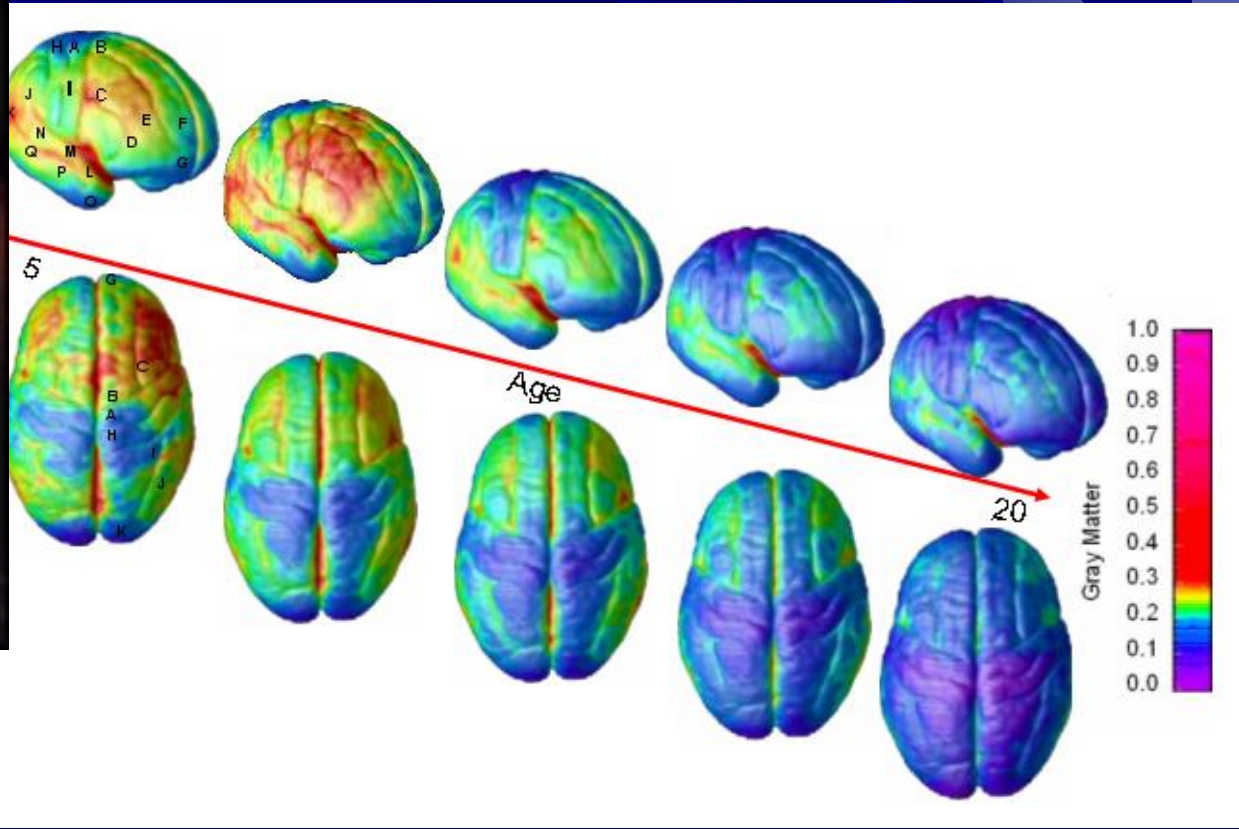
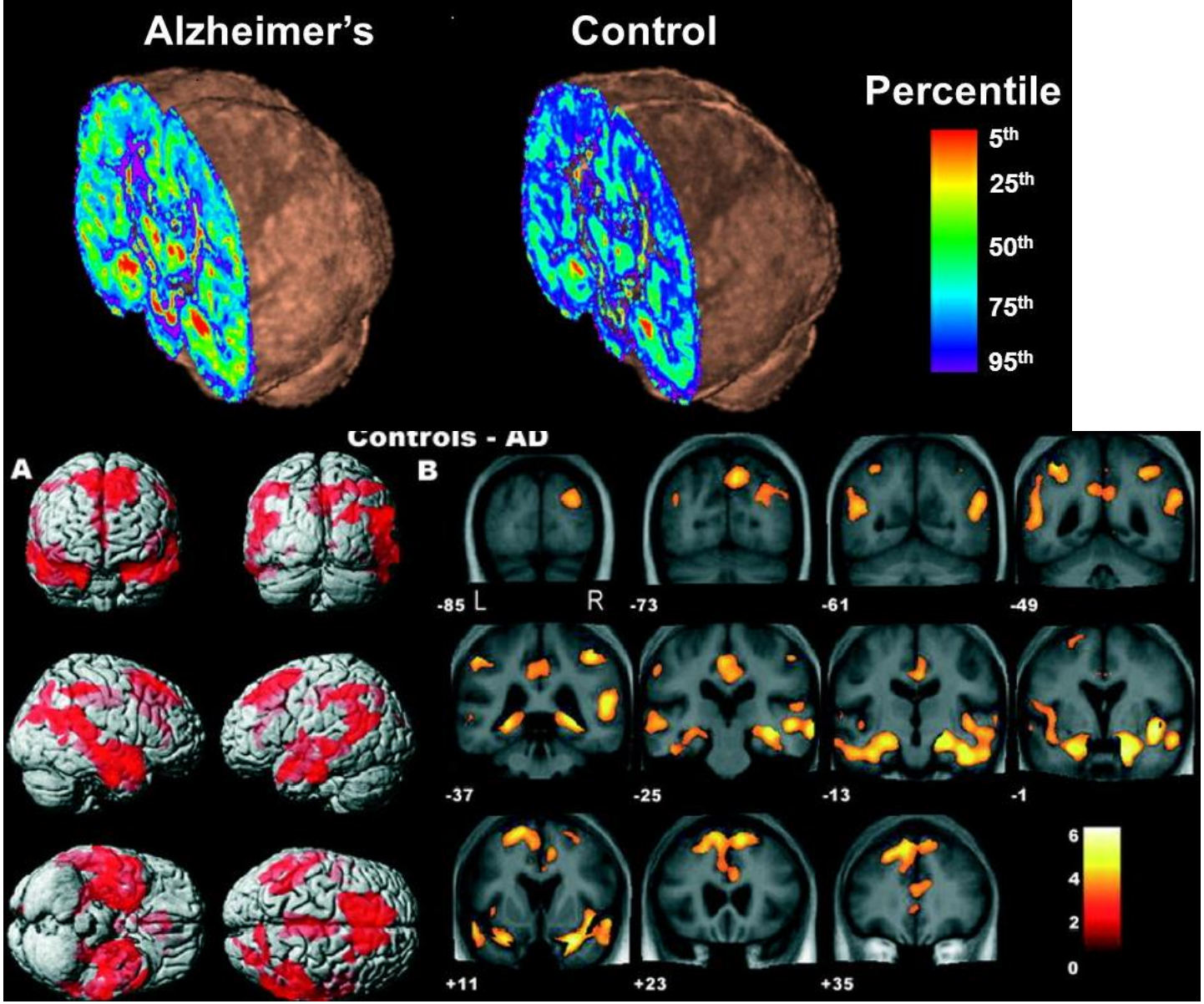
Time is acting on our brain with both maturative and degenerative processes



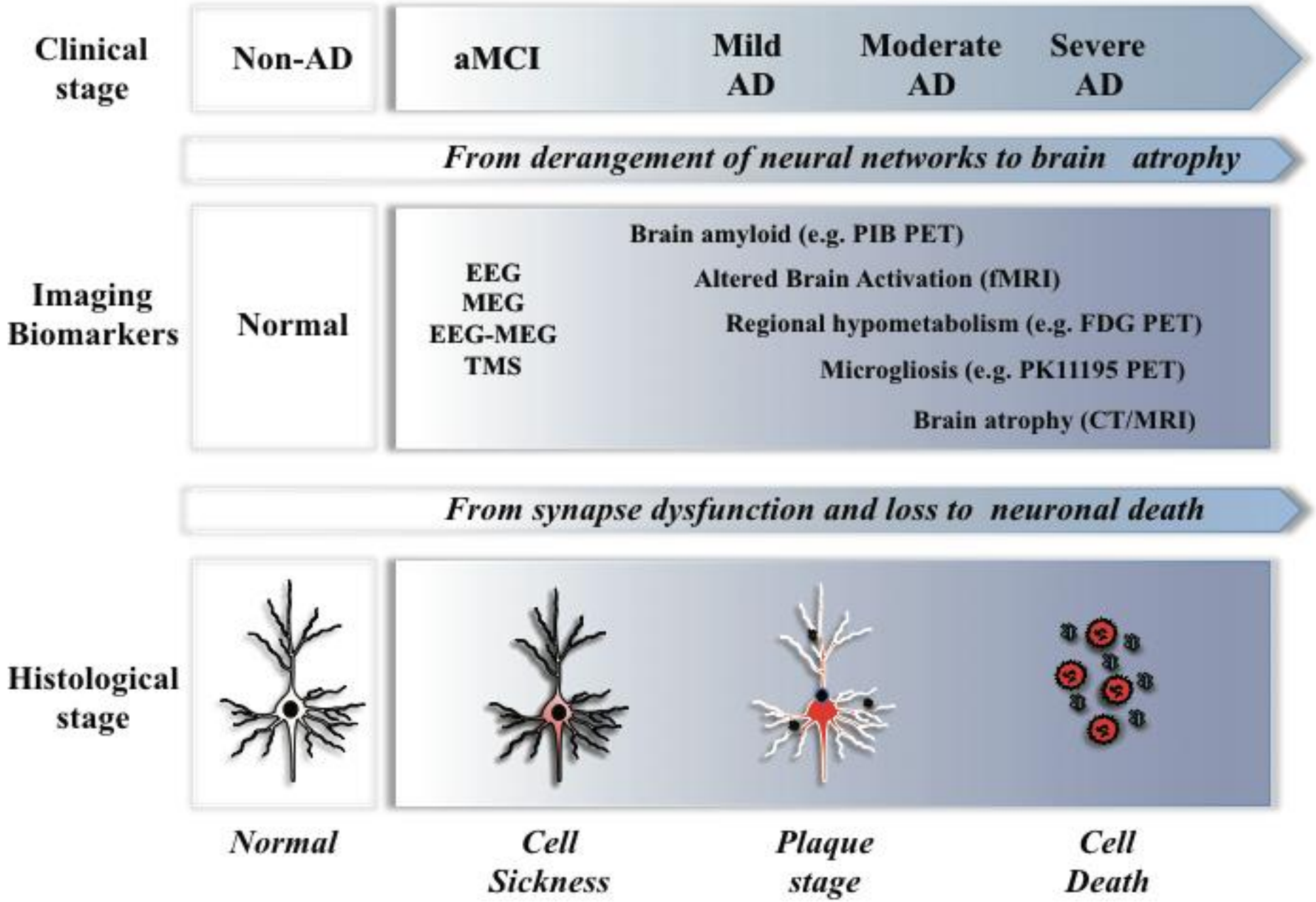
In recent decades >20% of the population in developed Countries is represented by people >65 y.o.



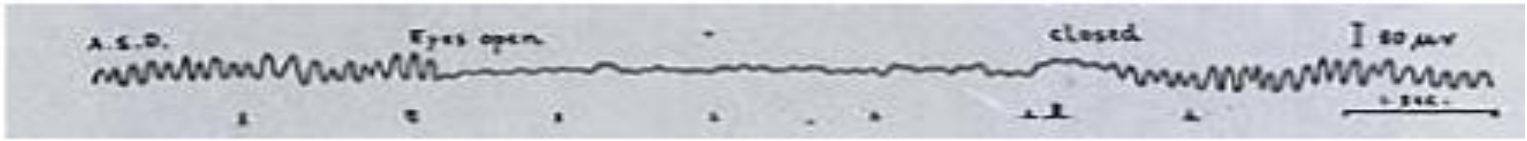


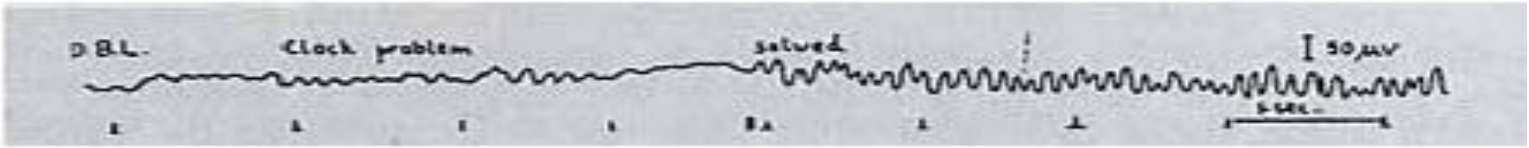
Voxel based Morphometry (Kinkingnehun et al. 2008)

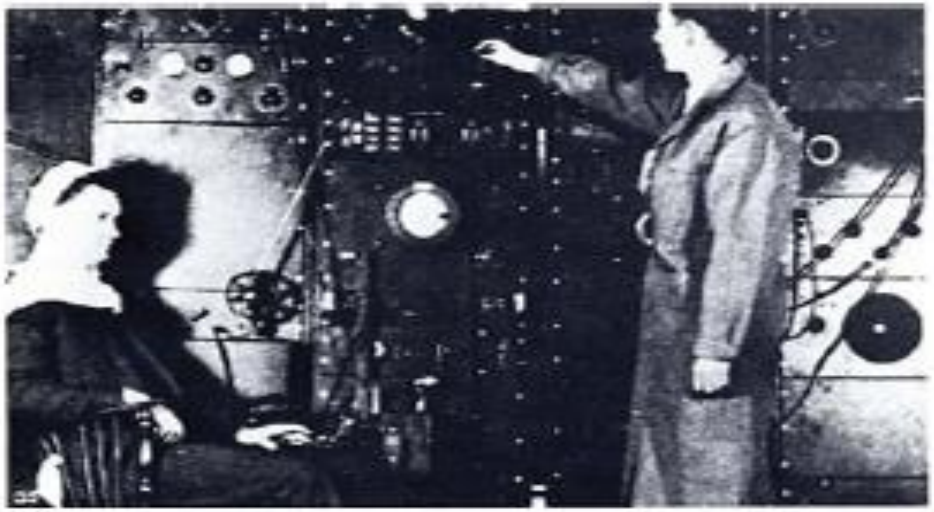
D'Amelio & Rossini Progr. Neurobiol. 2012



Cerebral EEG rhythms change in amplitude during sensorimotor and cognitive events



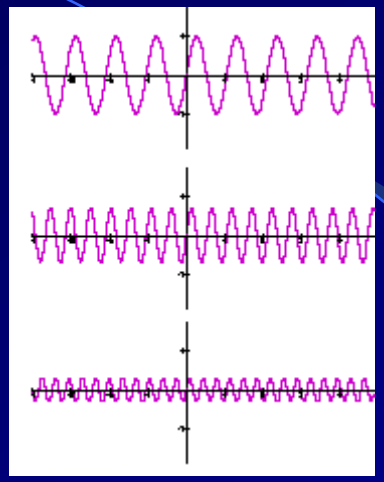




One-channel EEG device at the Harvard Medical School, 1934

Power of cortical EEG rhythms

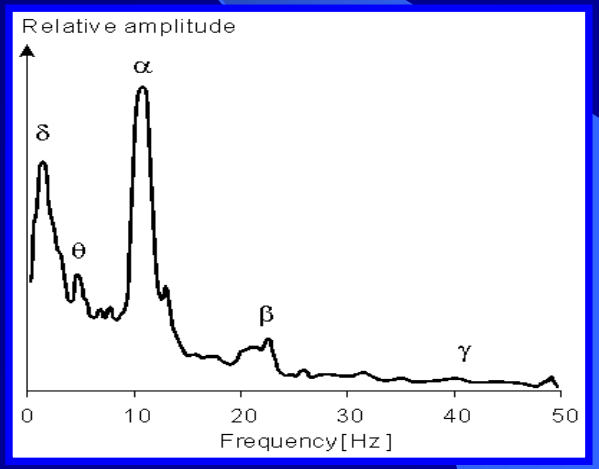
EEG signal can be divided in sinusoids at different frequencies

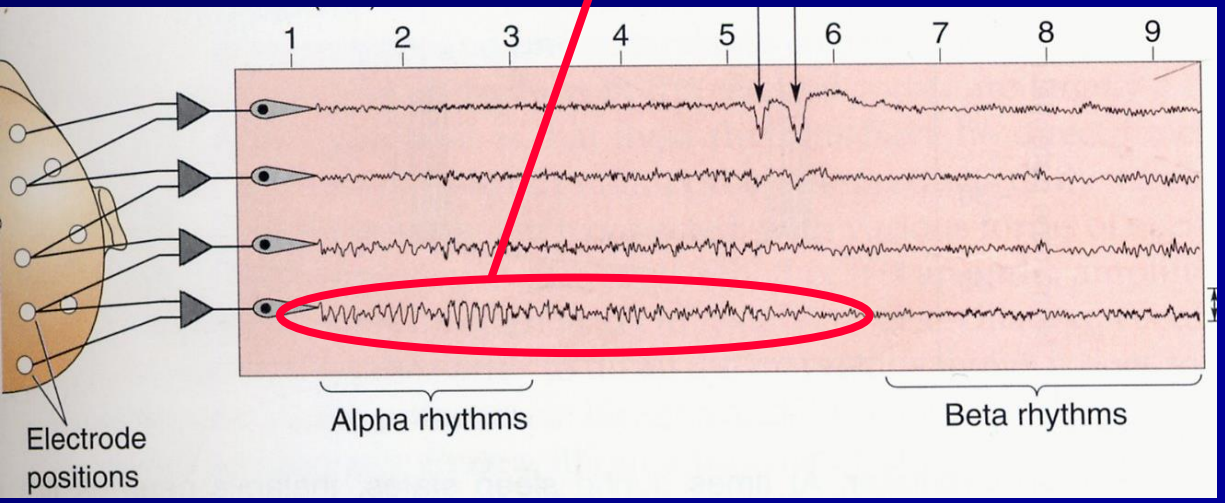


10 Hz α

20 Hz β

40 Hz γ



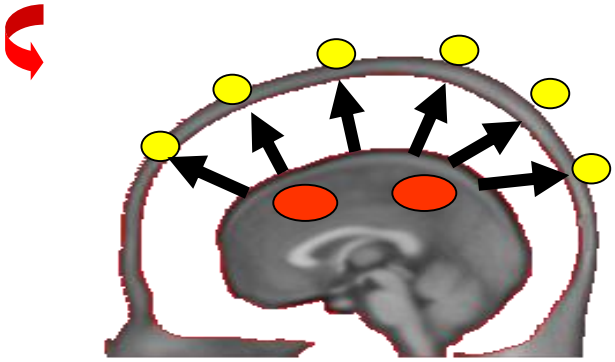


Magnitude of each EEG sinusoid is represented by spectral power at that frequency

CORTICAL SOURCES ANALYSIS of EEG RHYTHMS

Direct Problem: from cortical sources to EEG

Inverse Problem: from EEG to cortical sources (underdeterminated and ill-conditioned)

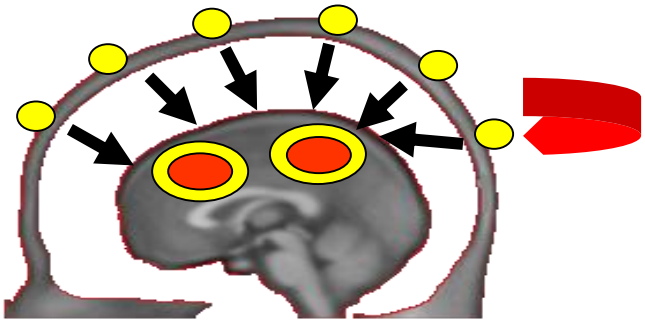


$$f = K \times J_{real}$$

(f) Scalp Potentials

(K) Transfer matrix

(J) "Real" current density



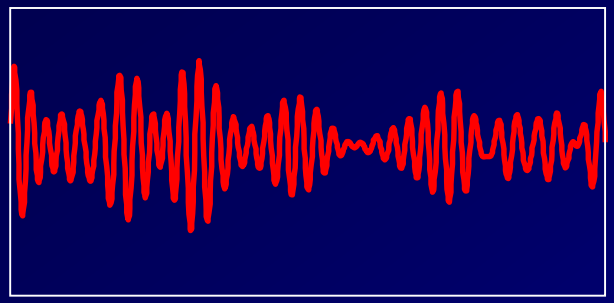
$$\hat{J}_{estim} = T \times f$$

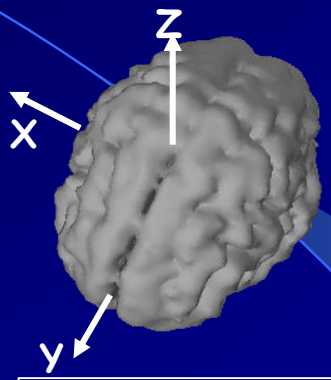
($\hat{J}$) "Estimated" current density

(T) Inverse of K matrix

(f) Scalp Potentials

From scalp EEG to its cortical sources: LORETA





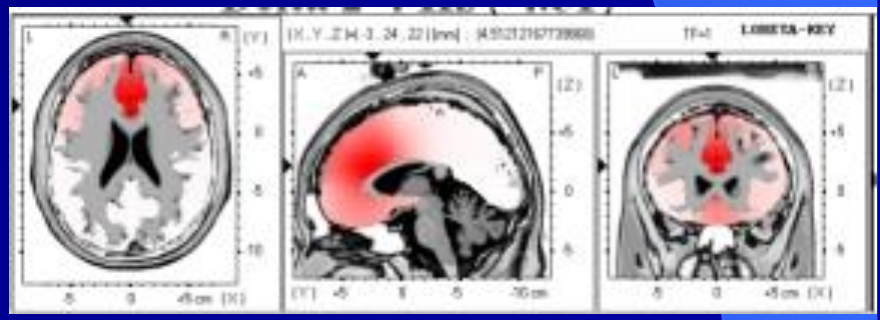
Spherical head model fitting a cortex model in the Talairach space

LORETA inverse linear estimation

Loreta computed 3-D linear solutions within a three shell-spherical head model including scalp, skull and brain compartments. This compartment included 2.394 voxels (7 mm resolution).

Matrix inversion regularization through minimization of the Laplacian solution

Visualization of 3-D LORETA solutions

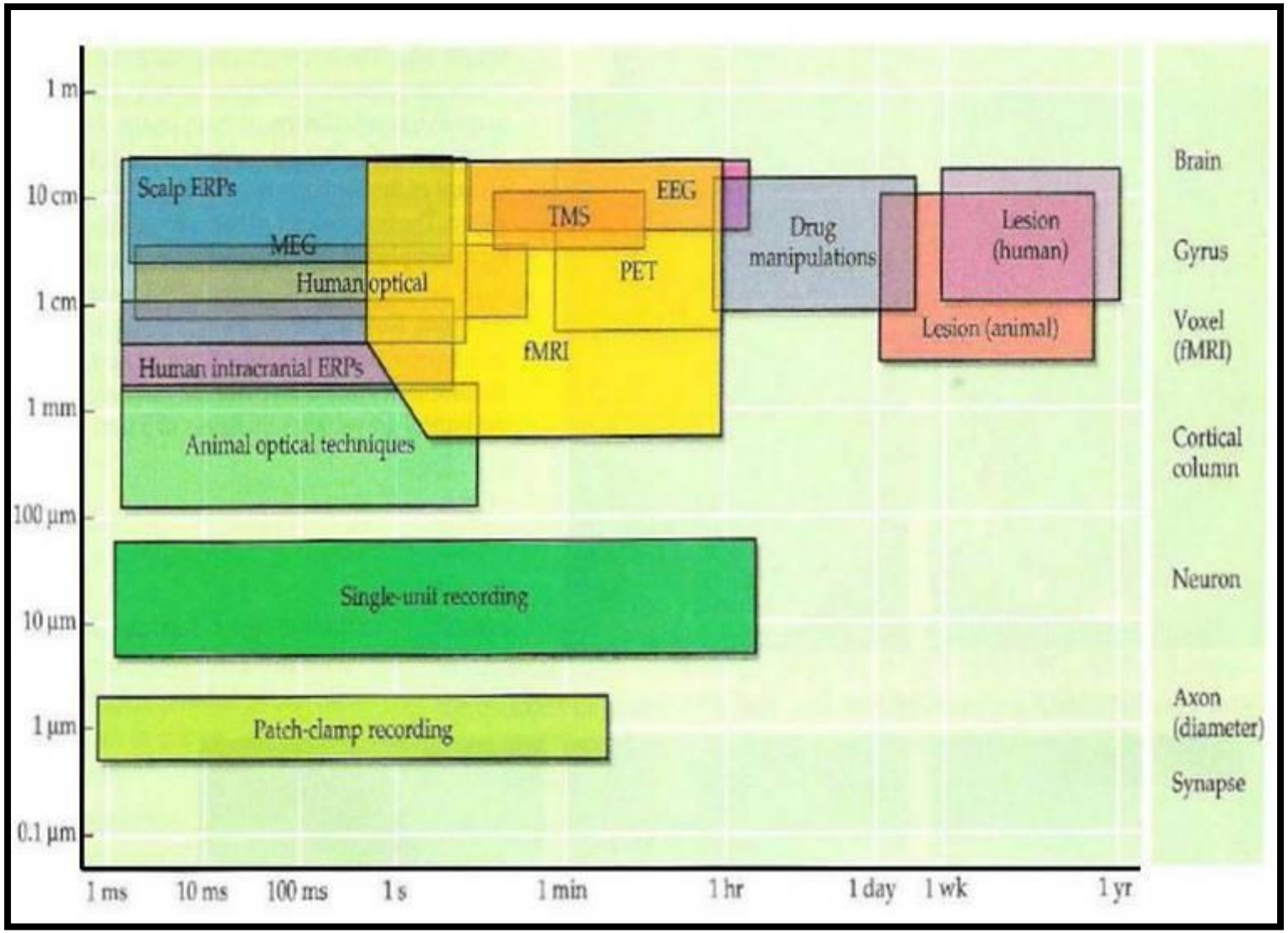


LORETA EEG CORTICAL SOURCES (Pascual-Marqui et al., 1994)

Axiale Sagittal Coronal

Spatiotemporal resolution of techniques for research in cognitive neuroscience

SPATIAL RESOLUTION

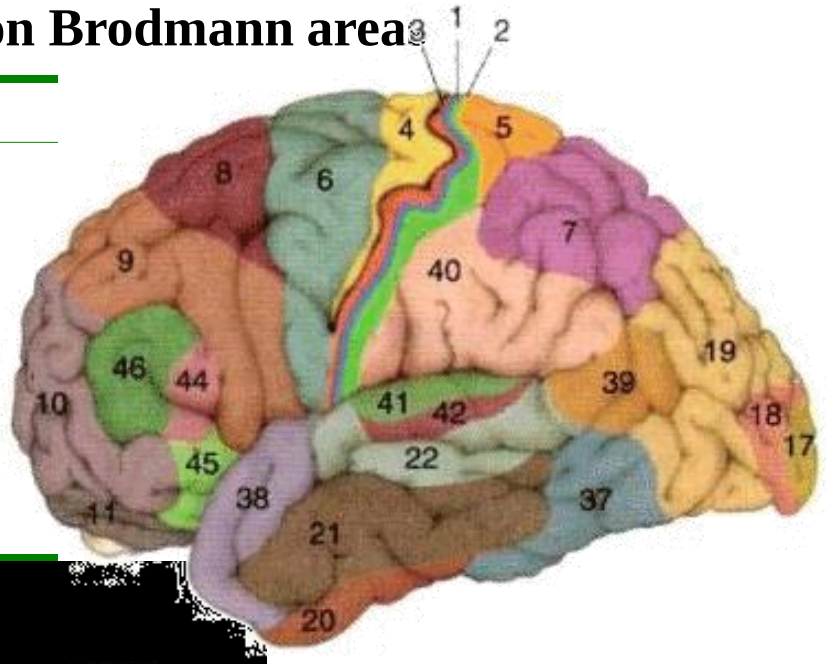


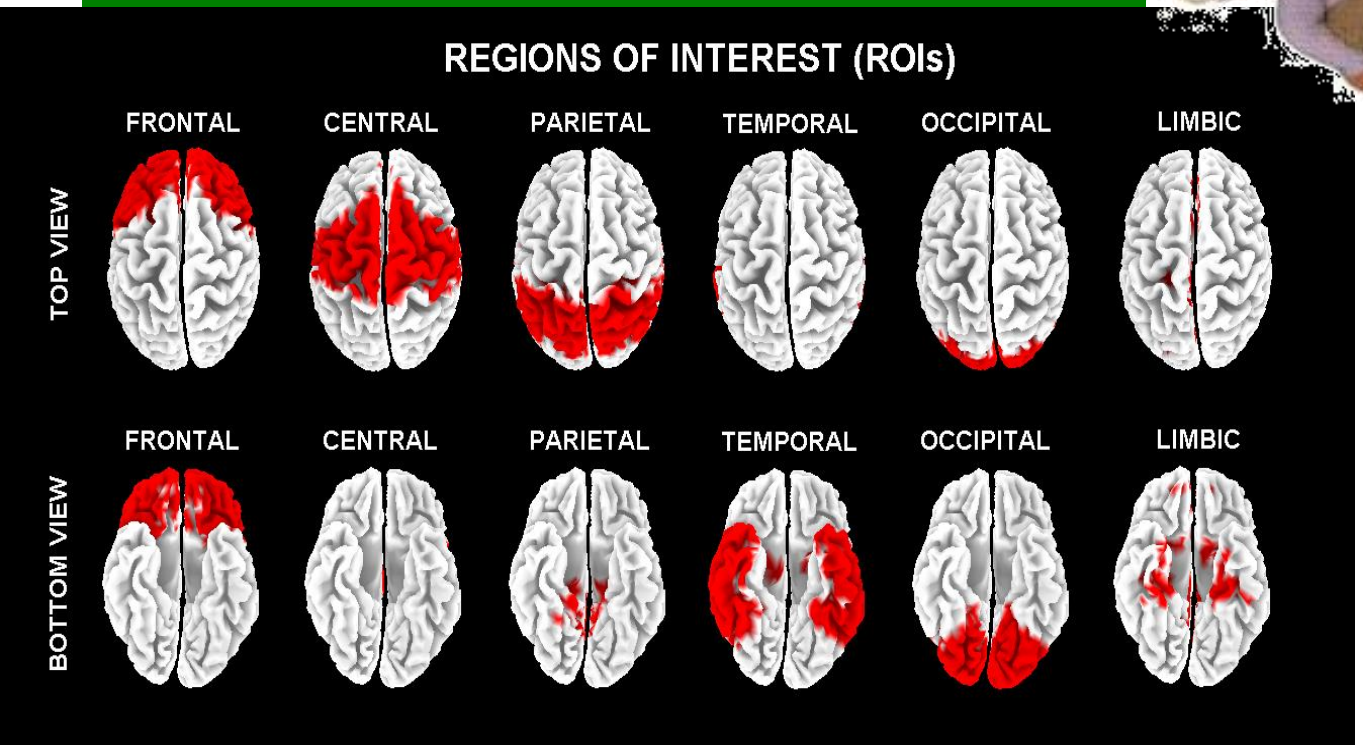
TEMPORAL RESOLUTION

A new approach to EEG via LORETA analysis:

MACROREGIONS based on Brodmann areas

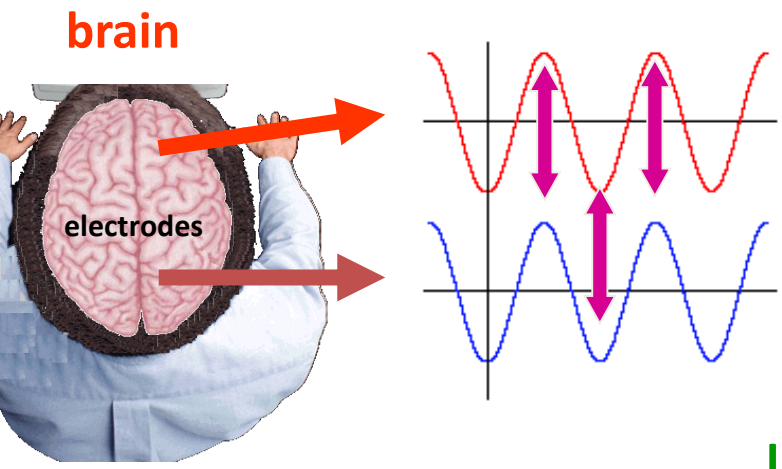
Regions of interest (ROIs)	
Frontal	(areas) 8, 9, 10, 11, 44, 45, 46, 47
Central	1, 2, 3, 4, 6
Parietal	5, 7, 30, 39, 40, 43
Temporal	20, 21, 22, 37, 38, 41, 42
Occipital	17, 18, 19
Limbic	12, 23, 24, 25, 26, 27, 28, 29, 31, 32, 33, 34, 35, 36





Linear temporal synchronization (coherence) of EEG rhythms at electrode pairs as an index of functional cortico-cortical coupling (information transfer)

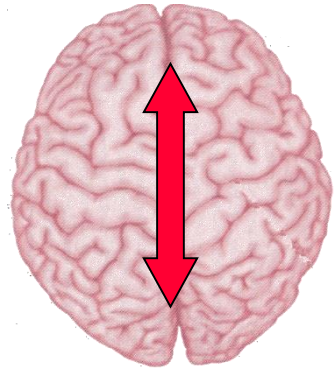
•It is defined as the square of the crossspectrum of the electrodes divided by the product of the power spectra of the individual electrodes.



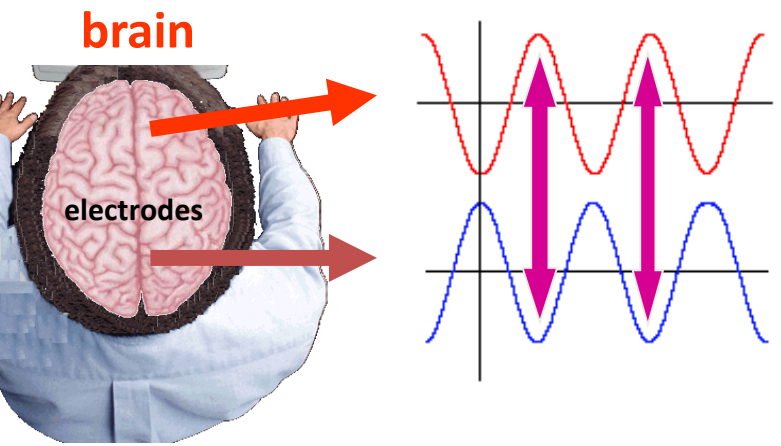
high spectral coherence

max coh = 1

= high information transfer



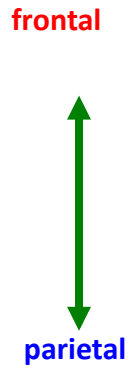
linear coupling



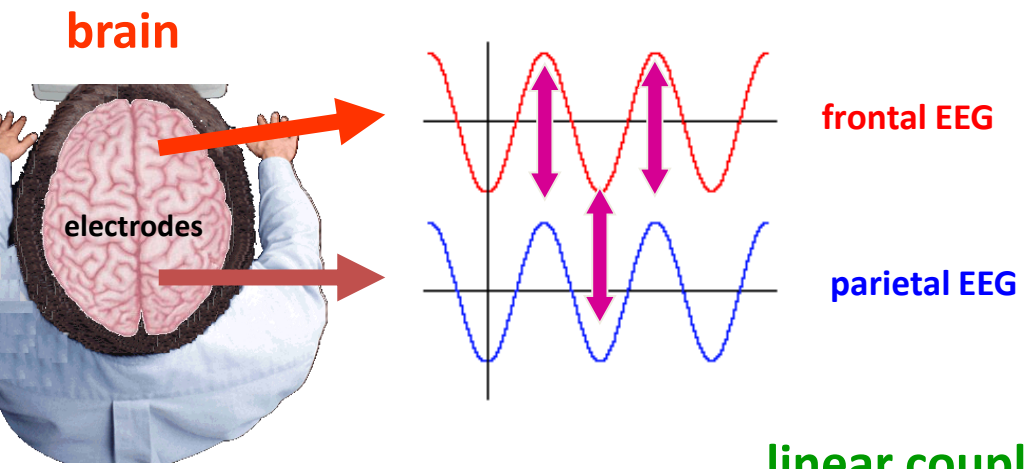
low spectral coherence

min coh = 0

= low information transfer



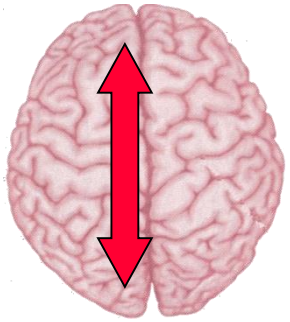
Linear temporal synchronization (coherence) of EEG rhythms at electrode pairs as an index of functional cortico-cortical coupling (information transfer)

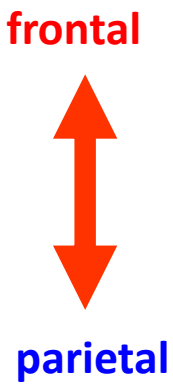


high spectral coherence

max coh = 1

= high information transfer



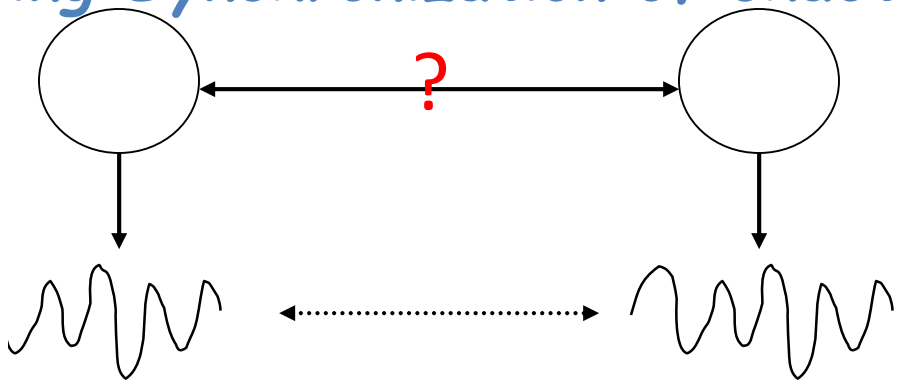


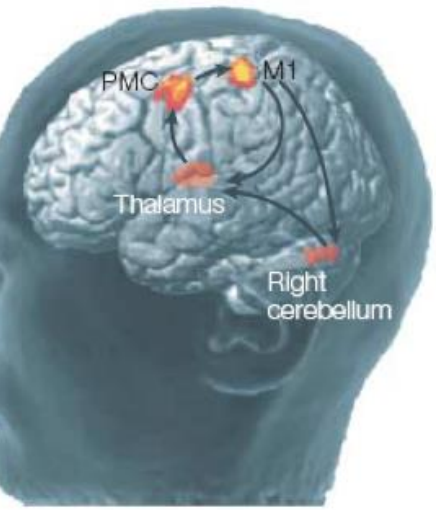
linear coupling

1. The brain shows local specialization
2. Complex tasks require cooperation between multiple brain areas
3. Rhythmic firing synchronization is a key mechanism for functional integration
4. Synchronization results in the formation of *functional networks* with *temporal* and *spatial* structure

How do distributed systems in the brain integrate their activity under normal and pathological conditions?

Time-varying Synchronization of chaotic oscillators





6–9 Hz movement discontinuities.

'Functional connectivity'

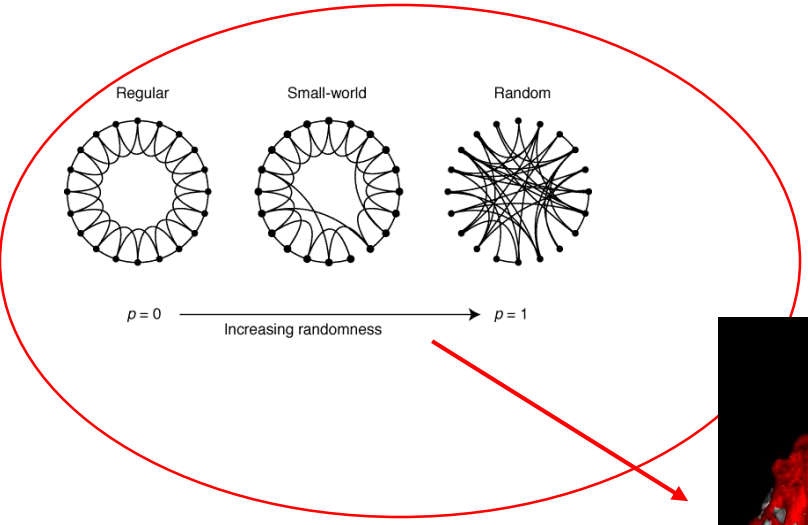
Dynamics of Synchronization:

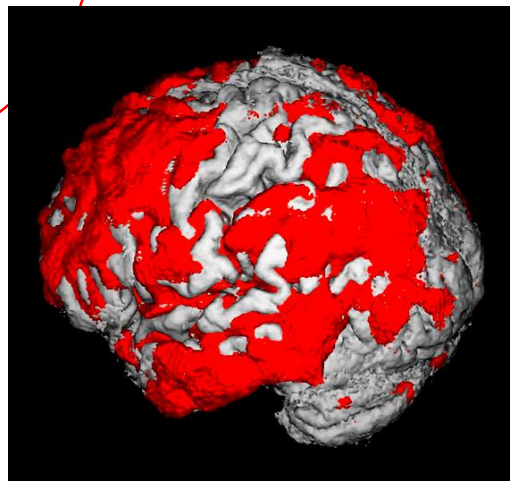
Excessive:
Epileptic paroxysms,Stroke

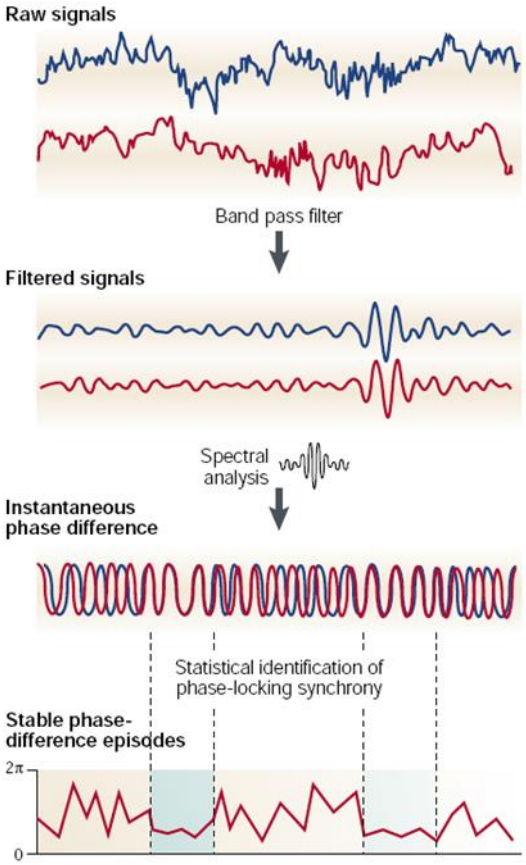
Normal:
'fragile binding'

Diminished:
Dysconnection / Cognitive dysfunction, Stroke

ALZHEIMER's DISEASE. A DYSCONNECTION SYNDROME?





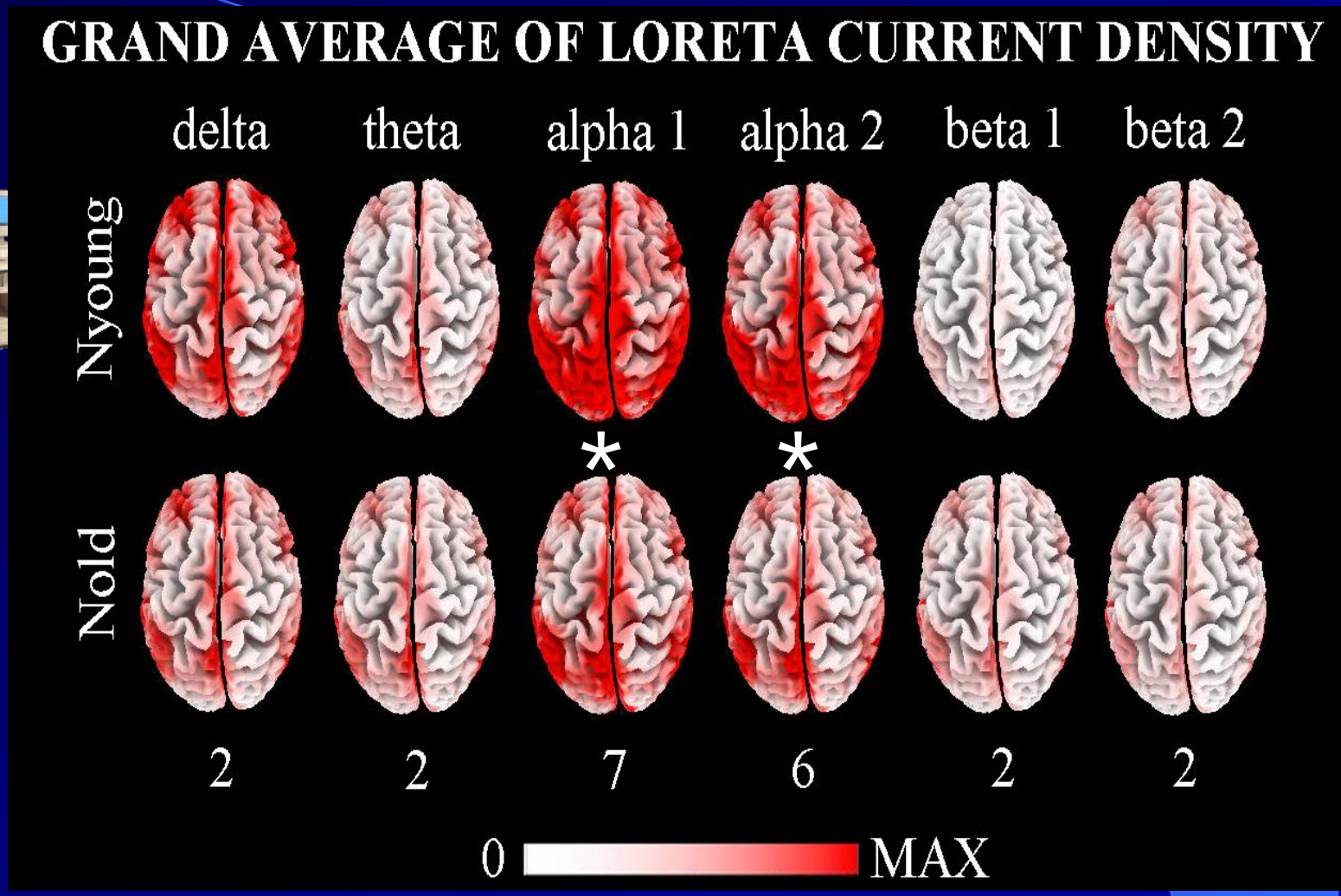


Cortical rhythms characterizing physiological aging

	Nyoung	Nold
N	108	107
Age (years)	27.3 (±7.3SD)	67.3 (±9.2 SD)
Gender (F/M)	56/52	67/40
MMSE	30	28.5 (±1.2 SD)
Education (years)	15.9 (±2.6 SD)	9.6 (±4.2 SD)

Diagnosis: DSM-IV and NINCDS-ADRDA criteria
EEG data: 5 min of resting EEG (closed eyes)
Data analysis: artifact rejection, LORETA at ROIs, statistical analysis (age and education as covariates)

Resting EEG data

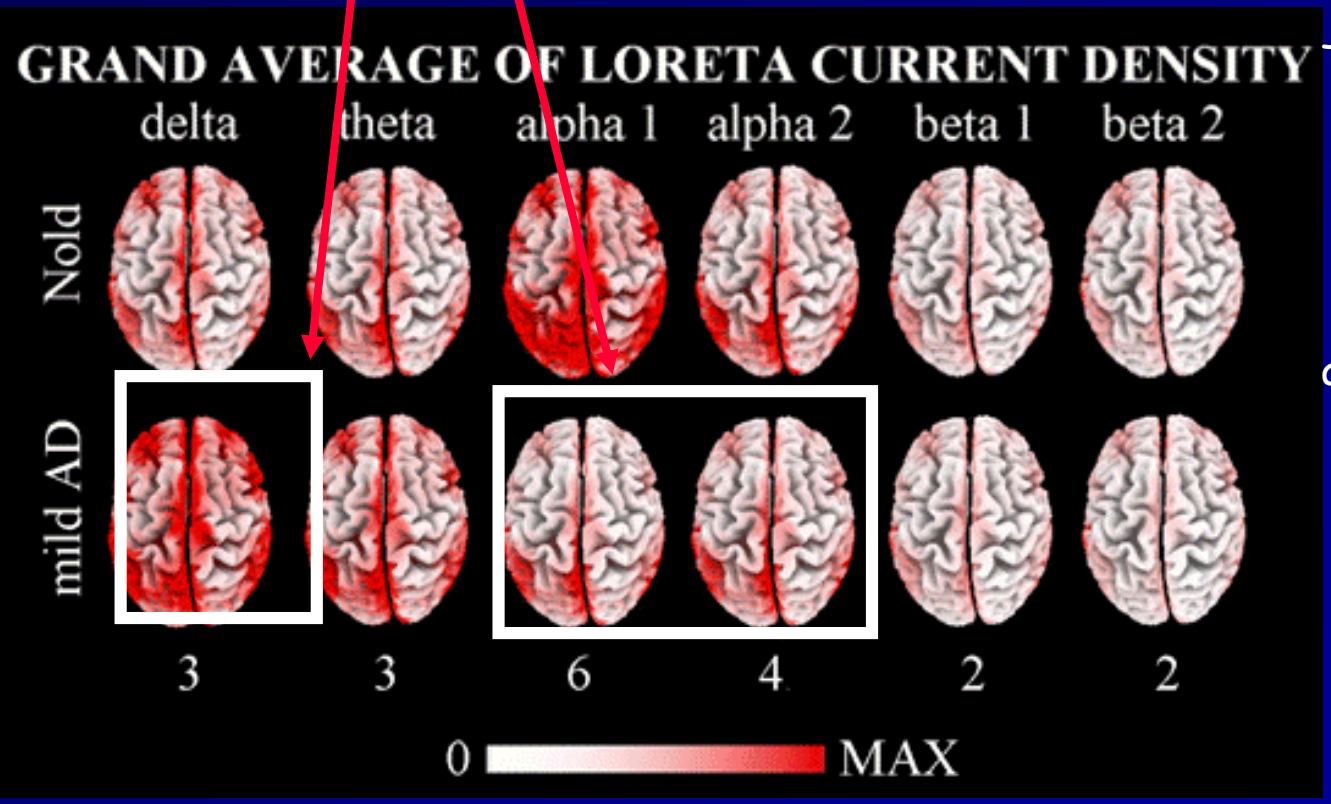


Babiloni C, Binetti G, Cassarino A, Dal Forno G, Del Percio C, Ferreri F, Ferri R, Frisoni G, Galderisi S, Hirata K, Lanuzza B, Miniussi C, Mucci A, Nobili F, Rodriguez G, Romani GL, and Rossini PM. Sources of cortical rhythms in adults during physiological aging: a multi-centric EEG study. *Human Brain Mapping* 2006

Alzheimer disease (AD) and EEG

- EEG rhythms reflects severity of Alzheimer disease (AD)

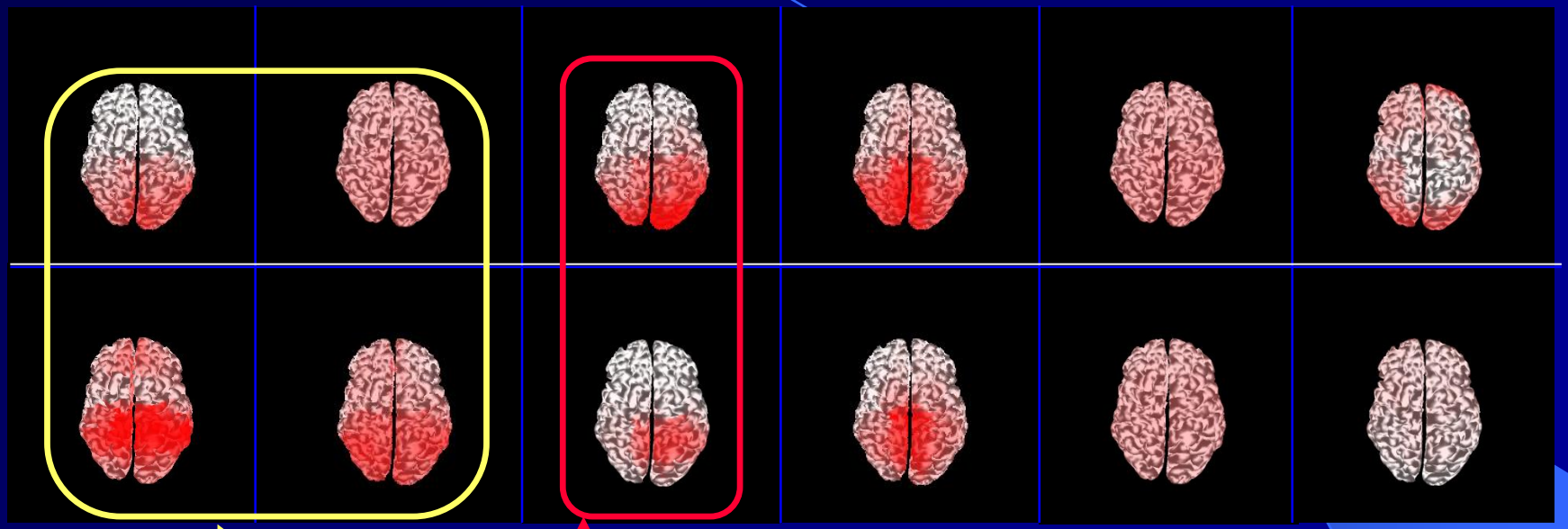
- Decline of cholinergic transmission in AD "slows" EEG rhythms
  - increases delta 2-4 Hz ("partial cortical disconnection")
  - decreases alpha 8-12 Hz ("abnormal cortical excitability")



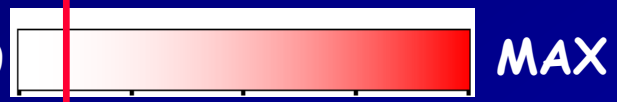
The EEG abnormalities in AD patients is a shift of the power spectrum to lower frequencies and a decrease in coherence of fast rhythms. These abnormalities are thought to be associated with functional disconnections among cortical areas resulting from death of cortical neurons, axonal pathology, cholinergic deficits, etc.

Cortical rhythms characterizing severity of mild AD

GRAND AVERAGE OF LORETA RELATIVE CURRENT DENSITY



3 | 2.5 | 3 | 2 | 1 | 1



posterior delta/theta increased

posterior alpha 1 increased

Babiloni C. et al., 2004; NeuroImage

Delta sources were stronger in magnitude with the maximal severity of the disease (mild AD- to mild AD+), along with an evident lesser magnitude of occipital alpha 1 sources.

Very mild AD - (N=23): **MMSE 24-21**
Mild AD + (N=25): **MMSE 20-17**

Mild cognitive impairment (MCI)

- Mild cognitive impairment (MCI) is a clinical condition characterized by memory (aMCI) or multimodal cognitive impairment not causing functional impairment and not yet encompassing the definition of dementia; it is a state of the elderly brain intermediate between normal cognition and dementia.
- MCI may be considered as a precursor of Alzheimer's Disease (AD). In normal aging (Nold) population annual conversion rate to AD ranges from 0.17% to 3.86%, while in MCI it is remarkably higher ranging between 6 and 35%.

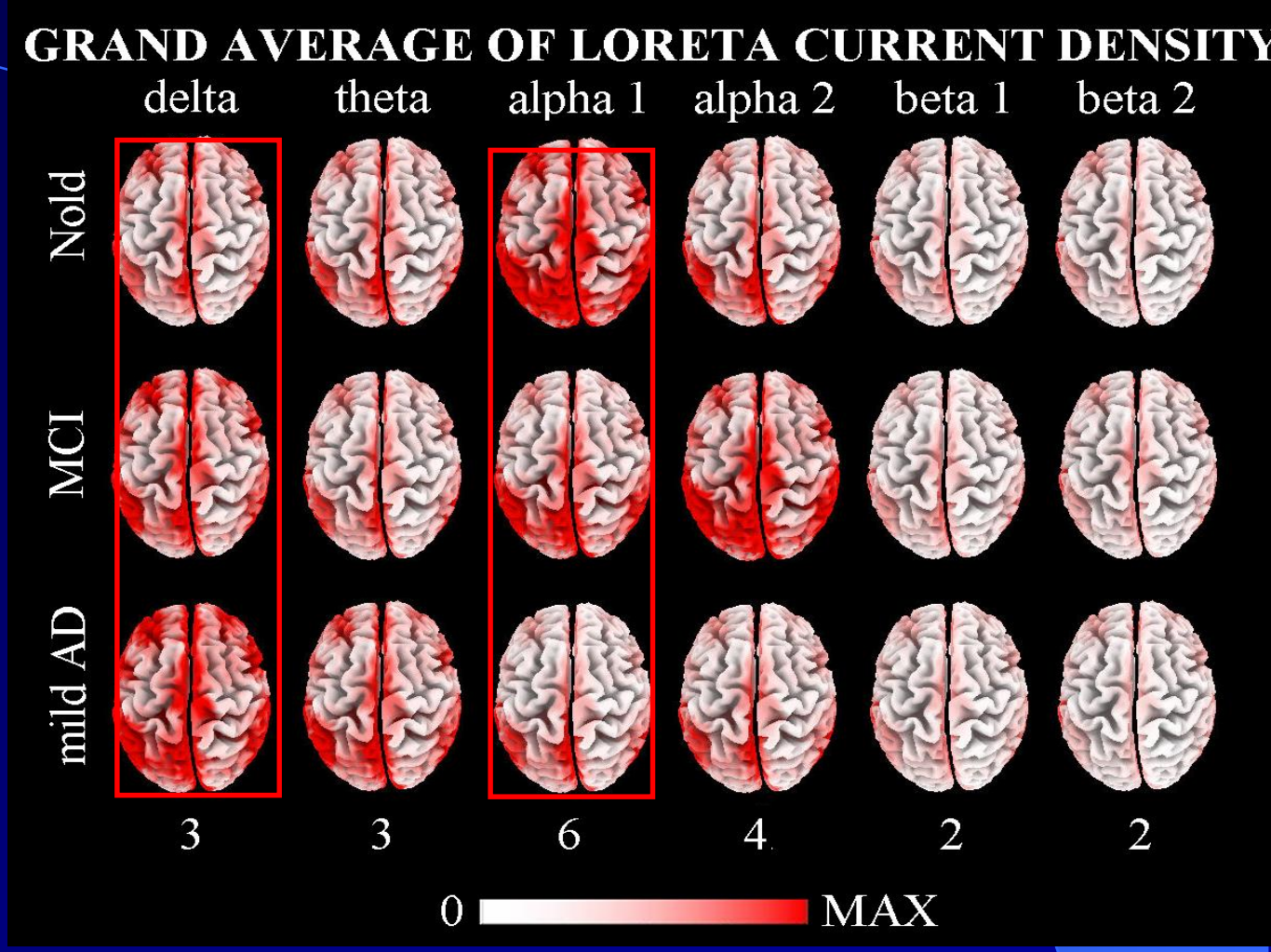
Age	Conversion Rate of Nold to AD	Conversion Rate of MCI to AD
65 - 69	0,17 %	From 6% to 35 %
85 - 89	3,86 %	

Resting EEG data:

126 Nold

155 MCI

193 mild AD



The Nold group presented alpha 1 sources with maximal values of the relative current density distributed in parieto-occipital regions. Delta, theta and alpha 2 sources had moderate relative current density values. Compared to Nold group, mild AD group showed an increase of delta sources, along with a dramatic reduction of parieto-occipital alpha 1 sources. With respect to Nold and mild AD groups, MCI group showed intermediate magnitude of delta and alpha 1 sources and greater magnitude of alpha 2 sources.

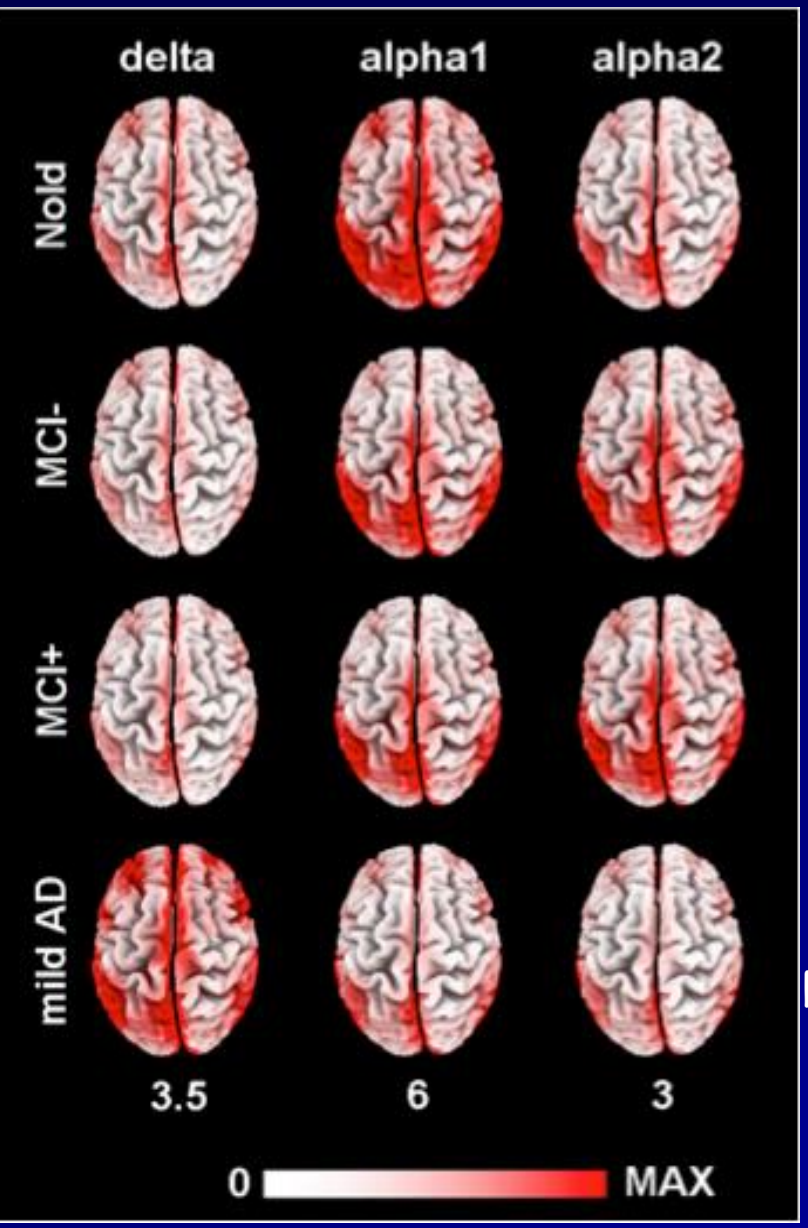
Babiloni C, Binetti G, Cassetta E, Dal Forno G, Del Percio C, Ferreri F, Ferri R, Frisoni G, Hirata K, Lanuzza B, Miniussi C, Moretti DV, Nobili F, Rodriguez G, Romani GL, Salinari S, and Rossini PM *Sources of cortical rhythms in subjects with mild cognitive impairment: a multi-centric study. Clinical Neurophysiology 2006*

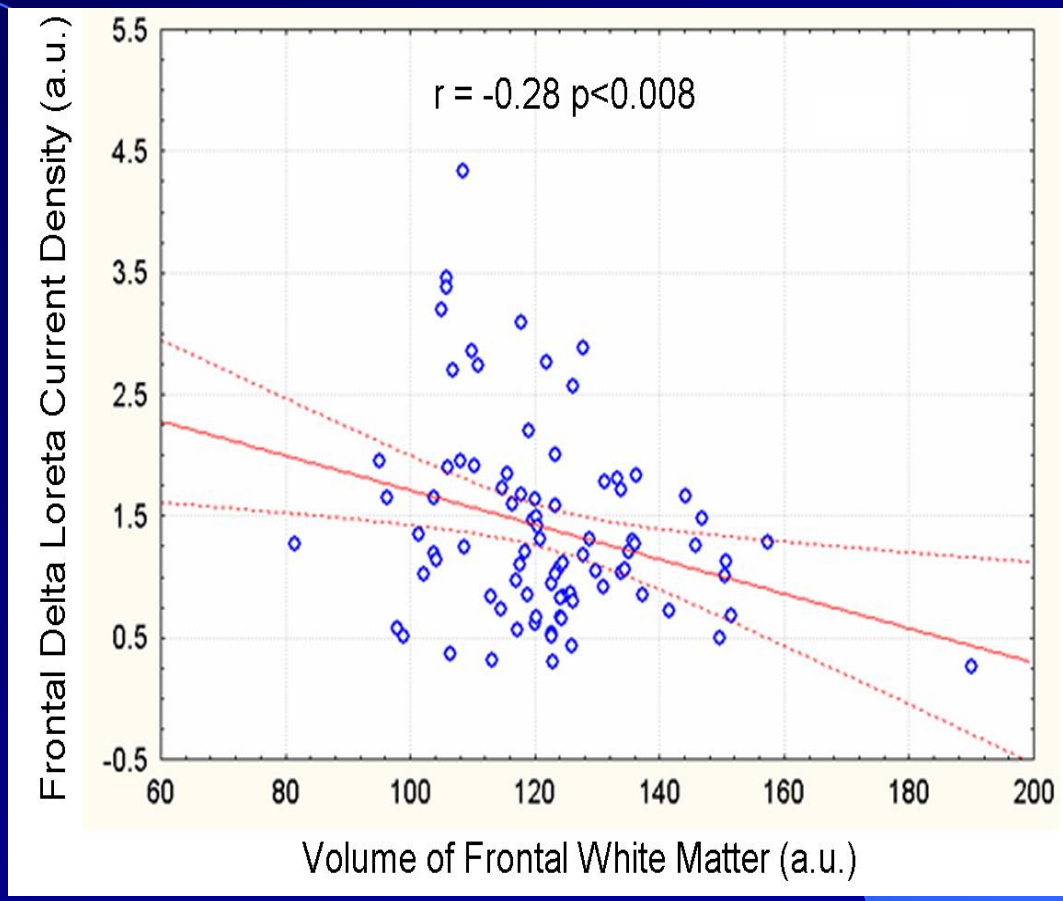
Relationships between LORETA sources of EEG rhythms correlating with pathological aging (MMSE) and local brain atrophy (MRI) in MCI and AD subjects

	Nold	MCI-	MCI+	mild AD
N	35	35	30	21
Age (years)	68.9 (±1.6 SE)	67.8 (±1.5 SE)	72 (±0.8 SE)	76.5 (±1.3 SE)
Gender (F\M)	18F/17M	28F/7M	15F/15M	16F/5M
MMSE	29 (±0.2 SE)	28.1 (±0.2 SE)	25 (±0.2 SE)	23.6 (±0.6 SE)
Education (years)	9.5 (±0.8 SE)	6.6 (±0.6 SE)	7.7 (±0.7 SE)	8.2 (±0.9 SE)

The MCI subjects were subdivided in
MCI+ (MMSE= 24-27) and MCI- (MMSE>27)

Scatterplot between the individual regional LORETA solution and white matter volume in





A significant negative correlation was observed between frontal WM and amplitude of the frontal delta LORETA sources in MCI and mild AD subjects considered as a single group.

Babiloni C, Frisoni G, Steriade M, Bresciani L, Binetti G, Del Percio C, Geroldi C, Miniussi C, Nobili F, Rodriguez G, Zappasodi F, Carfagna T, Rossini PM. e. Clin Neurophysiol. 2006

These results confirmed for the first time the hypothesis that the sources of resting delta rhythms (2–4 Hz) are correlated with lobar brain volume across MCI and AD subjects. We found a negative correlation between the frontal delta sources with global cognitive status (MMSE) and the volume of frontal white matter.

The present findings further support the 'transition hypothesis' of brain structural and functional continuity between MCI and mild AD, at least at group level.

ApoE4 genetic risk of late onset AD affects EEG rhythms in MCI and AD

	MCI		mild AD	
	non apoe4	apoe4	non apoe4	apoe4
N	58	31	51	52
Age (years)	70.9 (±1.2 SE)	70.7 (±1.2 SE)	74.9 (±1.3 SE)	75.2 (±1.2 SE)
Gender (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 (years)	7.3 (±0.5 SE)	8 (±0.8 SE)	6.7 (±0.2 SE)	5.8 (±0.4 SE)

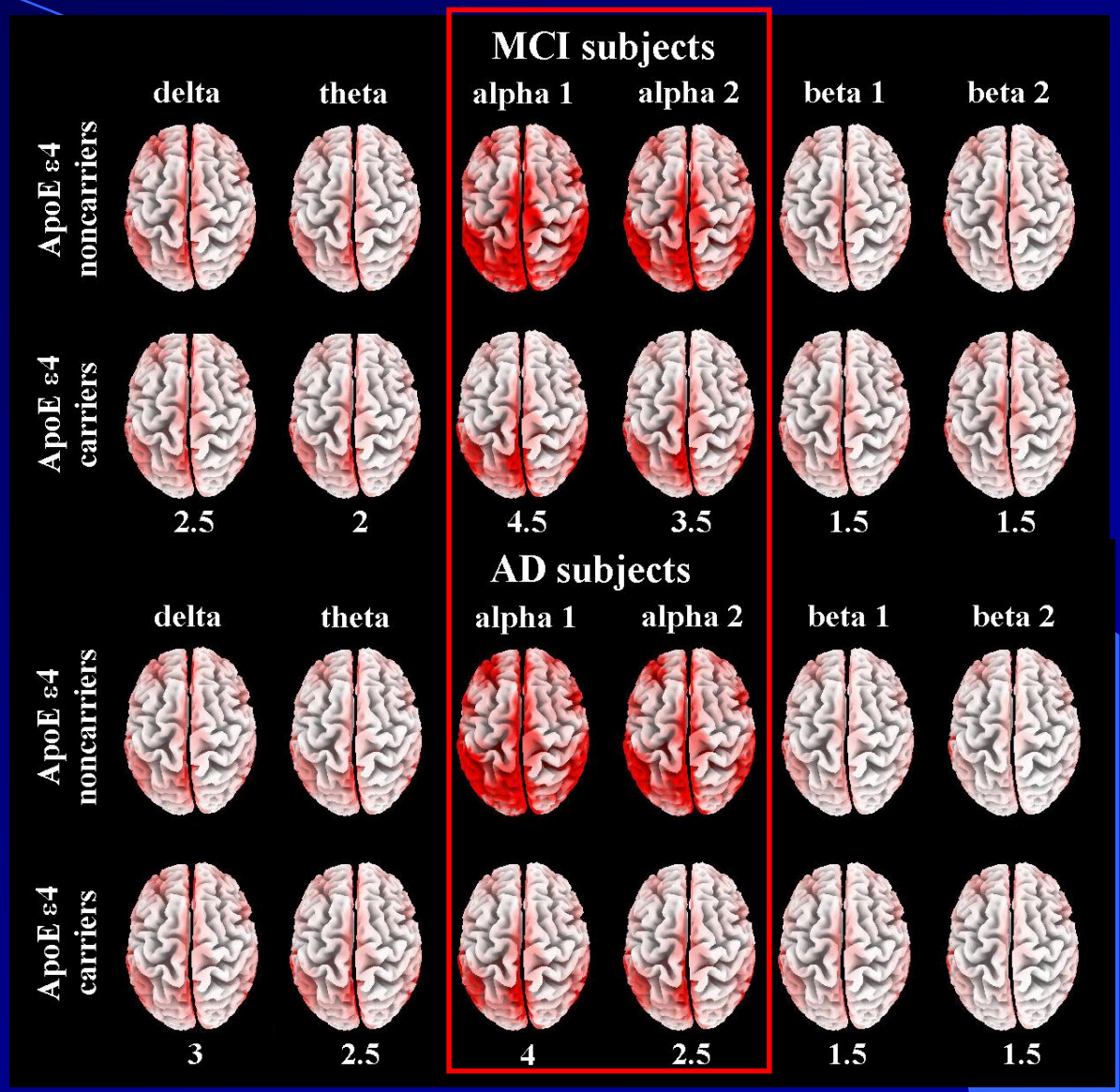
Diagnosis: DSM-IV, NINCDS-ADRDA, NINDS-AIREN criteria
EEG data: 5 min of resting EEG (closed eyes)
Data analysis: artifact rejection, LORETA, statistical analysis (age and education and IAF as covariates)

GRAND AVERAGE OF LORETA RELATIVE DENSITY CURRENT

Resting EEG data:

89 MCI (38% ε4)

103 AD (50% ε4)



Both MCI- and AD- groups presented alpha 1 sources with maximal values of the relative current density distributed in the parietooccipital regions. In comparison, MCI+ and AD+ ε4 carriers showed a reduction of alpha 1 and alpha 2 sources intensities.

Babiloni C, et al. Annals of Neurology (2006)

The challenge of the MCI research

MCI
(mild cognitive impairment)

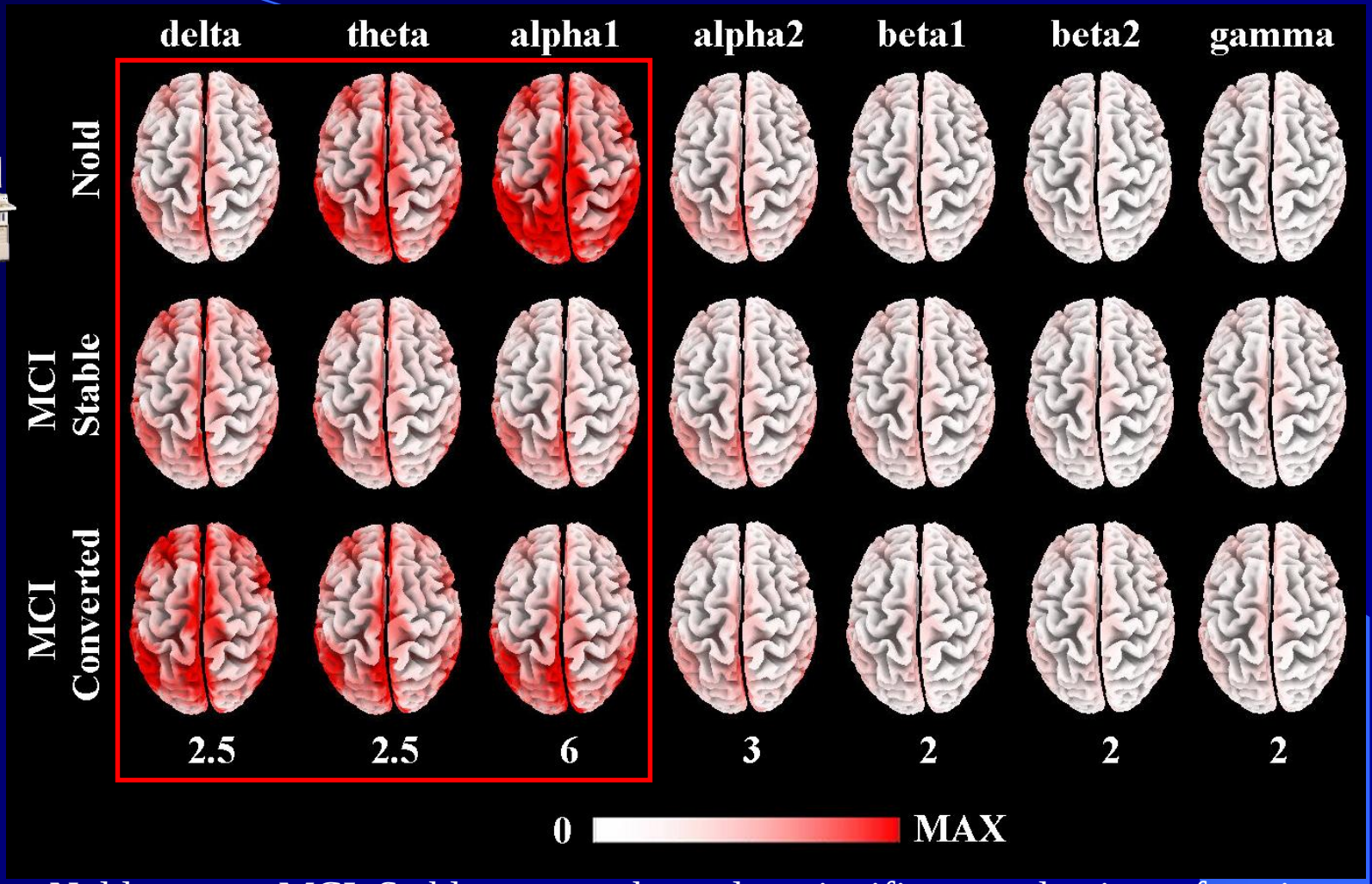
characterizes people with slight cognitive decline and enhanced risk to develop AD

AD
(Alzheimer's disease)

?

Diagnostic/prognostic value of a multimodal approach
genetic (ApoE-Cystatin)
phenotipic (EEG-MRI)

GRAND AVERAGE OF LORETA CURRENT DENSITY

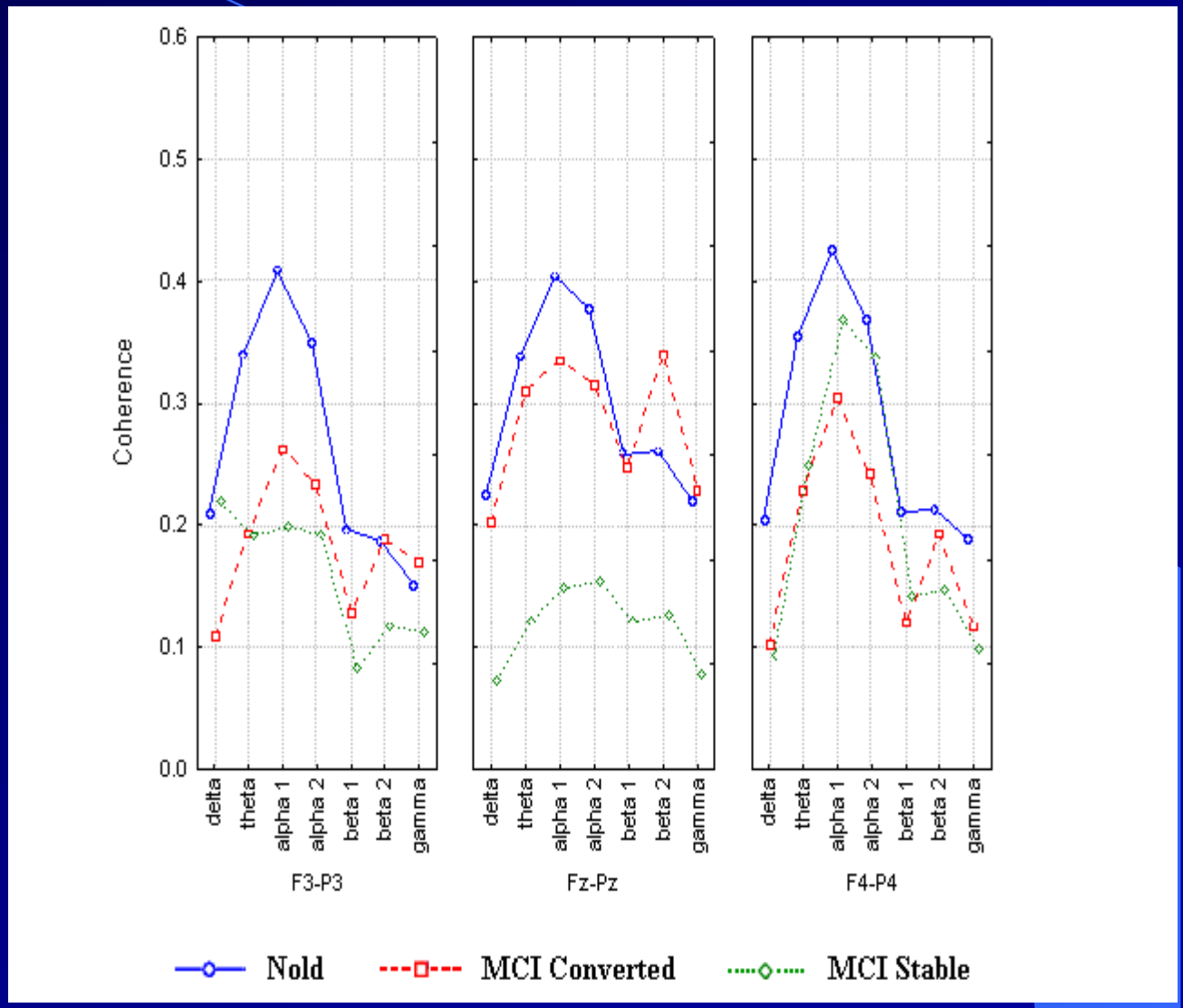


Compared to the Nold group, MCI Stable group showed a significant reduction of parieto-occipital alpha1, along with a reduction of occipital theta source strengths. With respect to the Nold and MCI stable, MCI Converted group showed intermediate magnitude of alpha 1 source and significantly stronger delta source strengths. Finally, compared to MCI stable, MCI converted showed an increase of occipital theta source.

ANOVA ANALYSIS OF EEG SPECTRAL COHERENCE

- ANOVA analysis of EEG spectral coherence showed a statistical interaction ($p<0.000001$) among the factors Group, Band, and Pair.

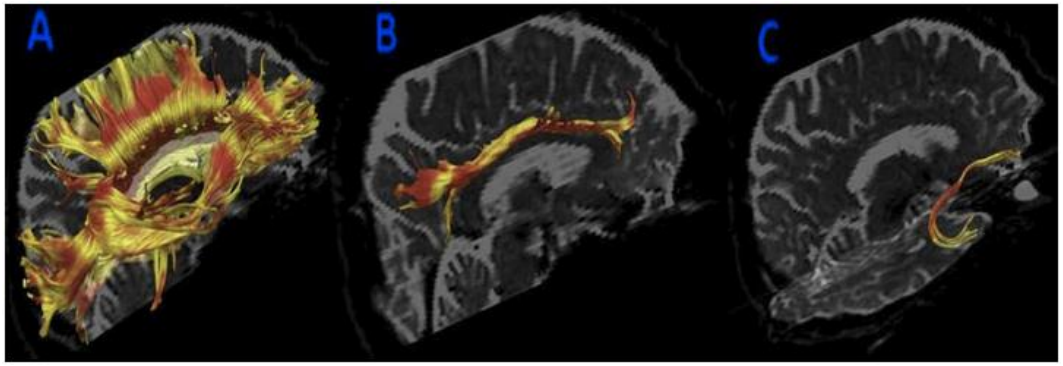
- Difference between MCI stable and MCI Converted were found especially in the medial fronto-parietal coupling.

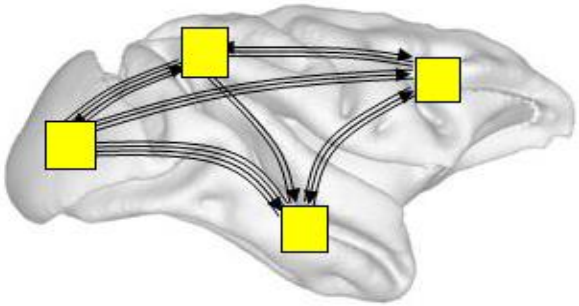


Rossini PM, Del Percio C, Pasqualetti P, Cassetta E, Binetti G, Dal Forno G, Ferreri F, Frisoni G, Chiovenda P, Miniussi C, Parisi L, Tombini M, Vecchio F, Babiloni C. *Conversion from MCI to Alzheimer's disease is predicted by sources and coherence of brain EEG rhythms*. Neuroscience, 2006

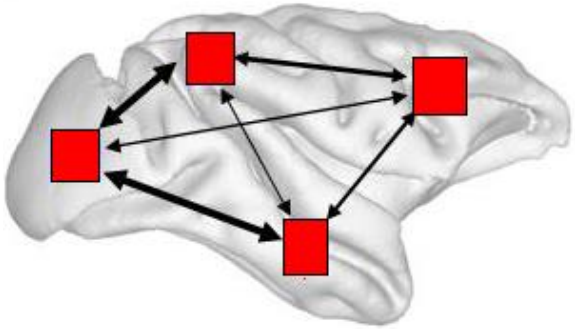
Cortico-cortical connectivity

- STRUCTURAL

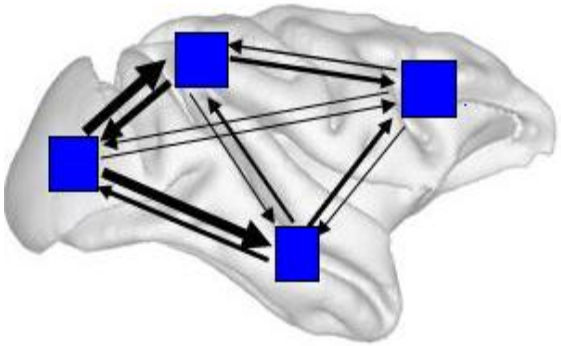


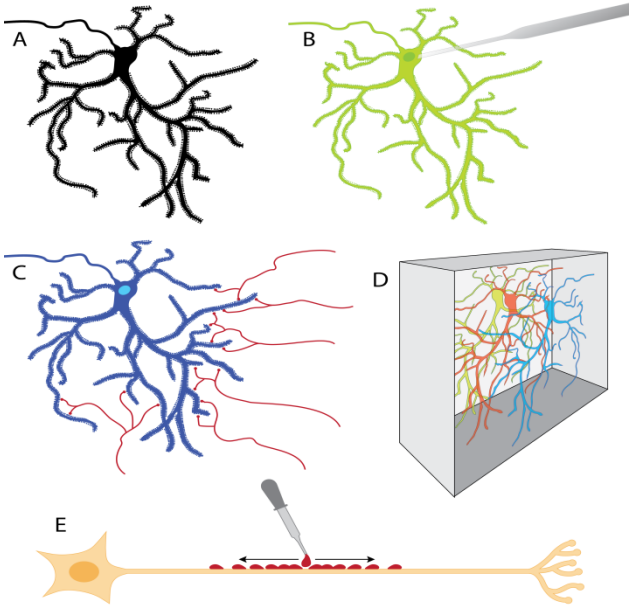


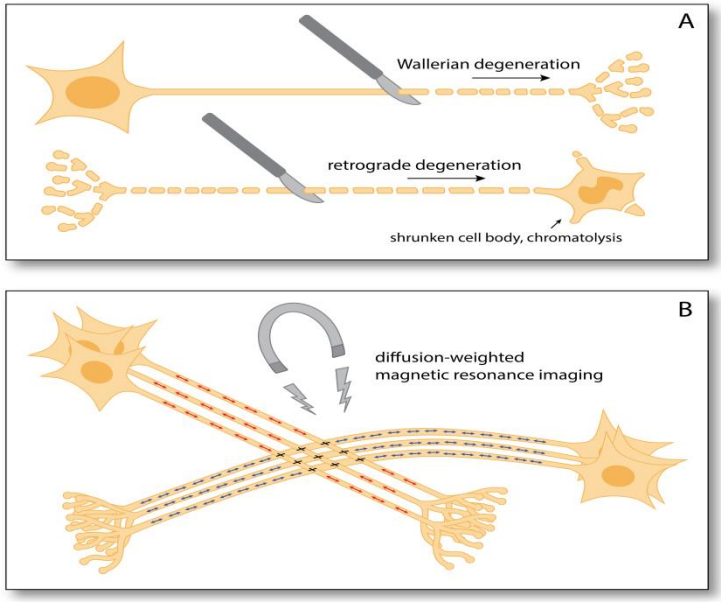
- FUNCTIONAL

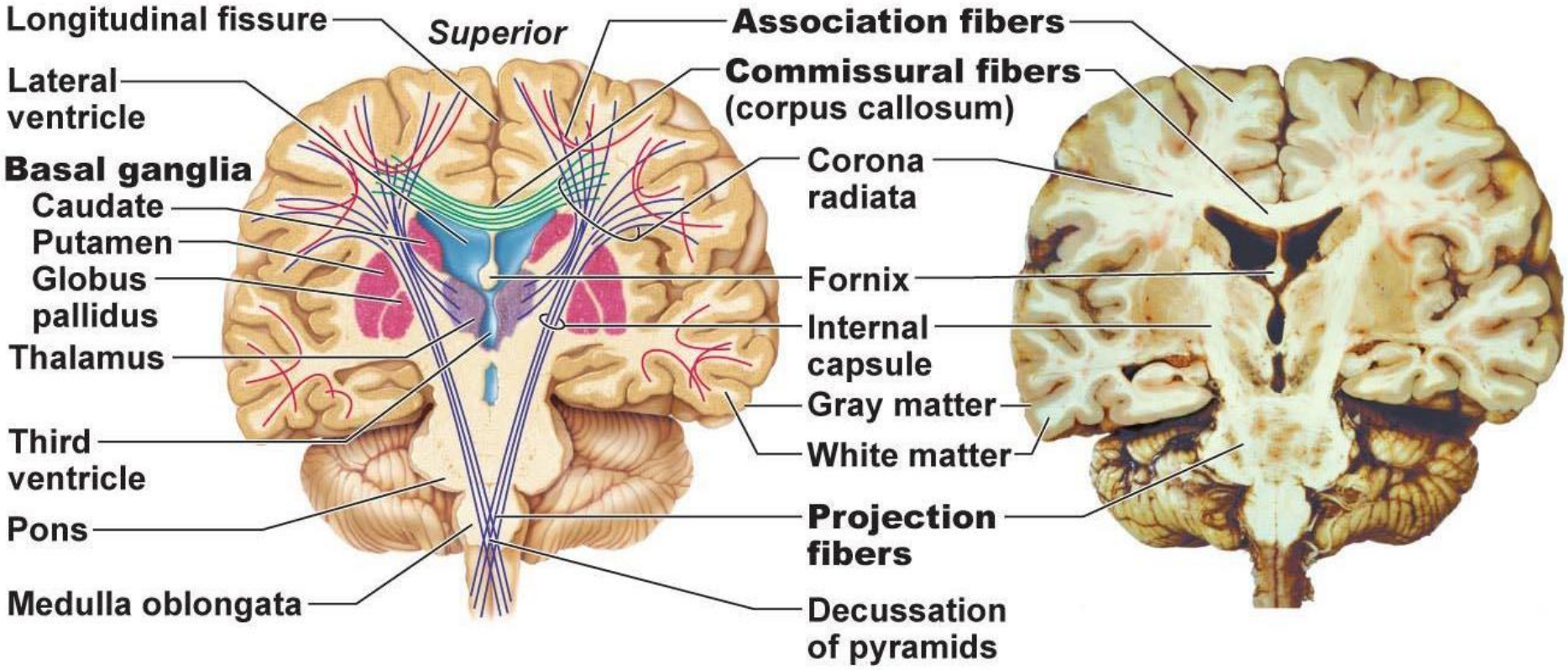


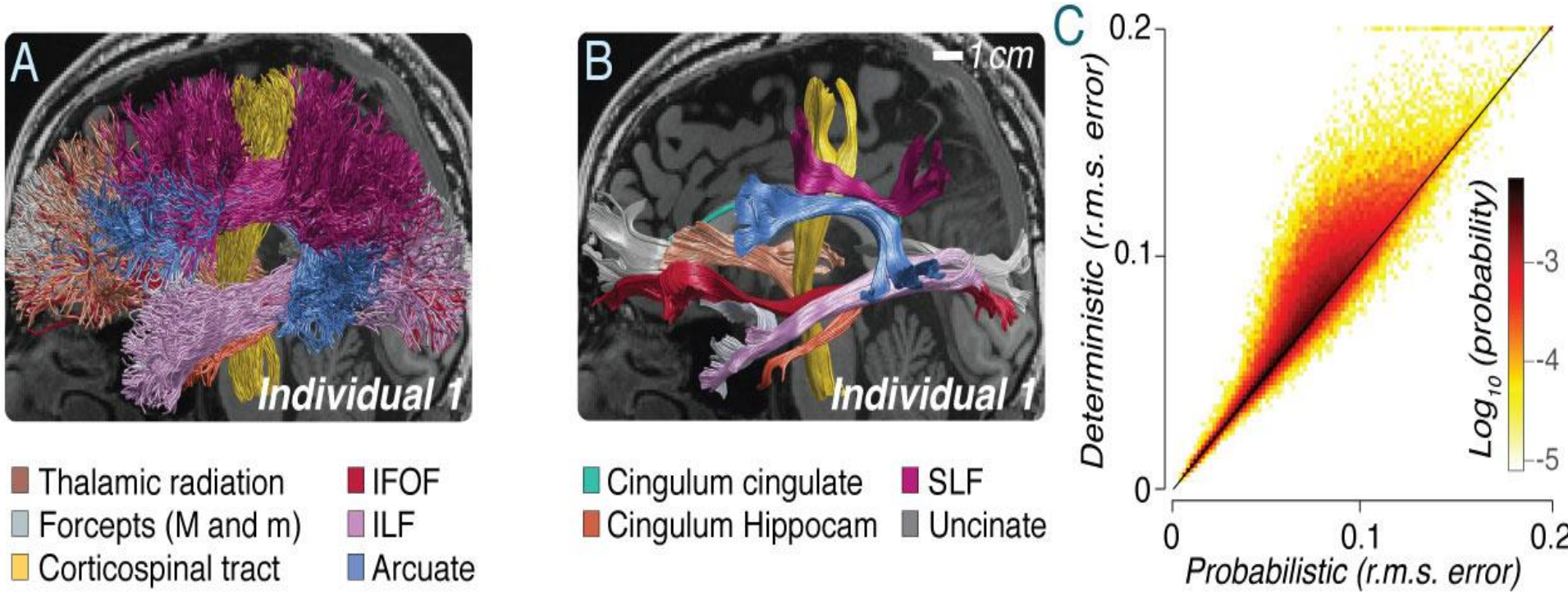
- EFFECTIVE (time-varying, dynamic !)




A
B
C
D
E

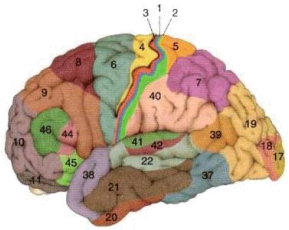

A
Wallerian degeneration
retrograde degeneration
shrunken cell body, chromatolysis
B
diffusion-weighted
magnetic resonance imaging


Longitudinal fissure
Superior
Lateral ventricle
Basal ganglia
Caudate
Putamen
Globus pallidus
Thalamus
Third ventricle
Pons
Medulla oblongata
Association fibers
Commissural fibers (corpus callosum)
Corona radiata
Fornix
Internal capsule
Gray matter
White matter
Projection fibers
Decussation of pyramids



- **The problem of tractography for precision connectomics. Major human WM tracts generated using probabilistic (A) and deterministic (B) tractography algorithms differ for a single individual. C. Comparison of the r.m.s. errors of the connectomes in A and B in predicting the diffusion signal using the LiFE method. Probabilistic tractography (A) shows a smaller prediction error in a majority of voxels for this individual.**

Proposed application of advanced EEG techniques for the evaluation of brain connectivity

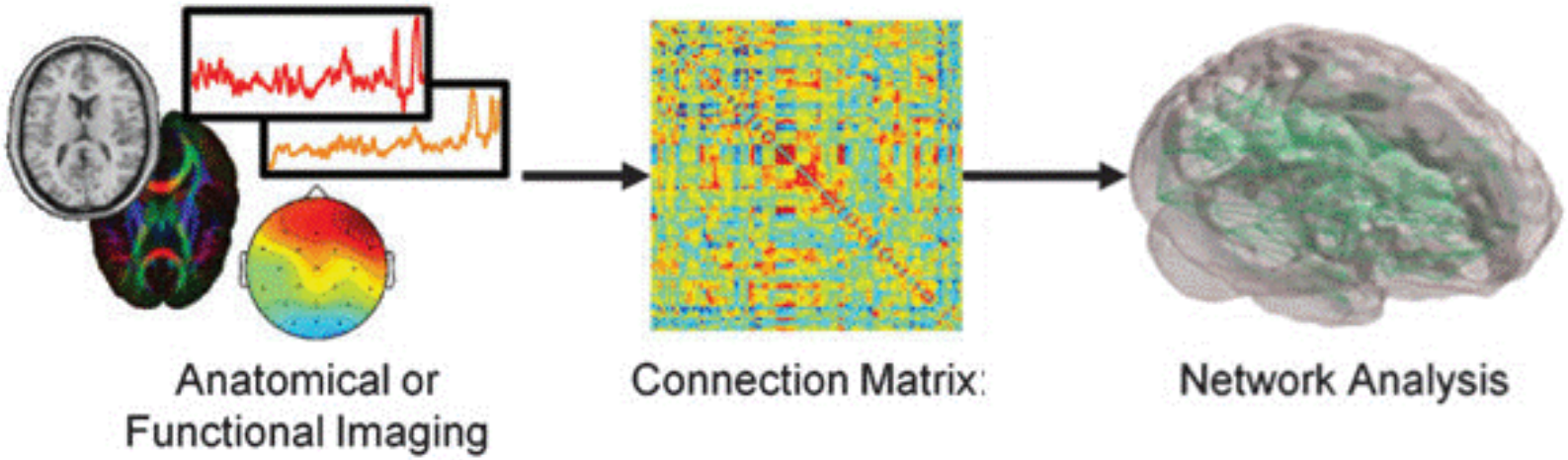


GRAPH THEORY

Describing a real-world system, a graph provides an abstract and a mathematical representation of the system's elements and their interactions.

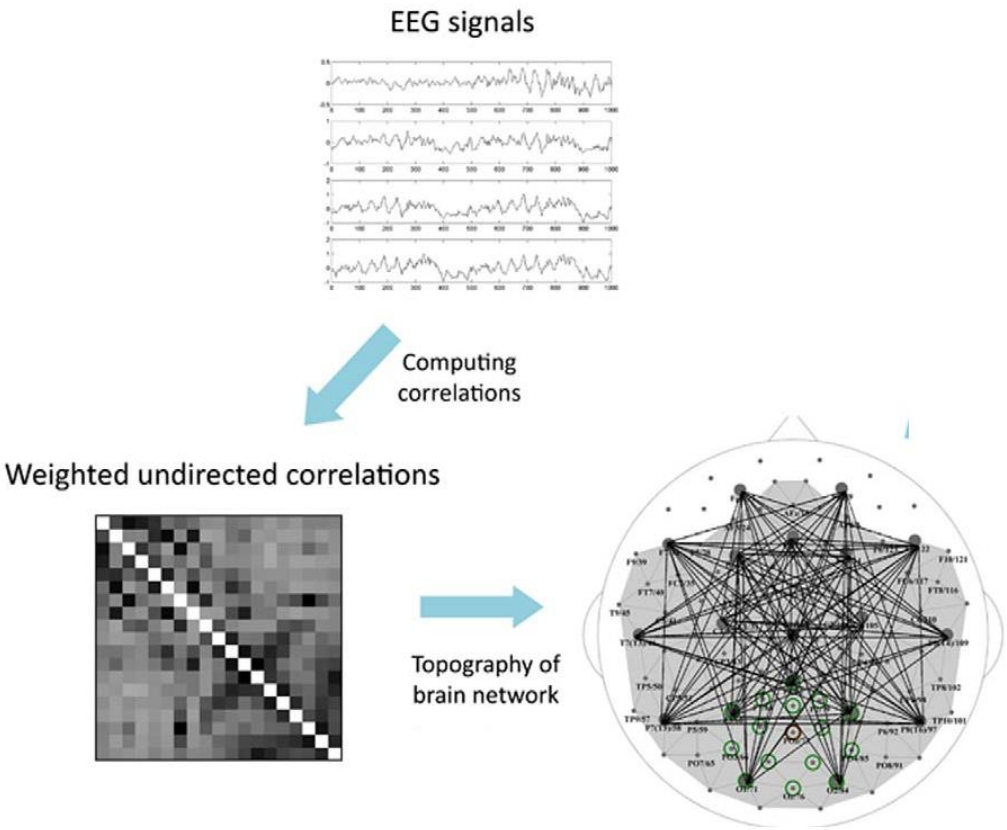
Graph theory describes mathematical methods applied to representations of networks reduced to their essence: vertices (nodes) and edges (connections).

NODES: neural elements; EDGES: structural or functional connectivity



Network analysis schematic. Anatomical and functional brain imaging data are analyzed to produce a connection matrix, denoting the strength of connection between nodes. From the matrix, various graph metric analyses can be performed.

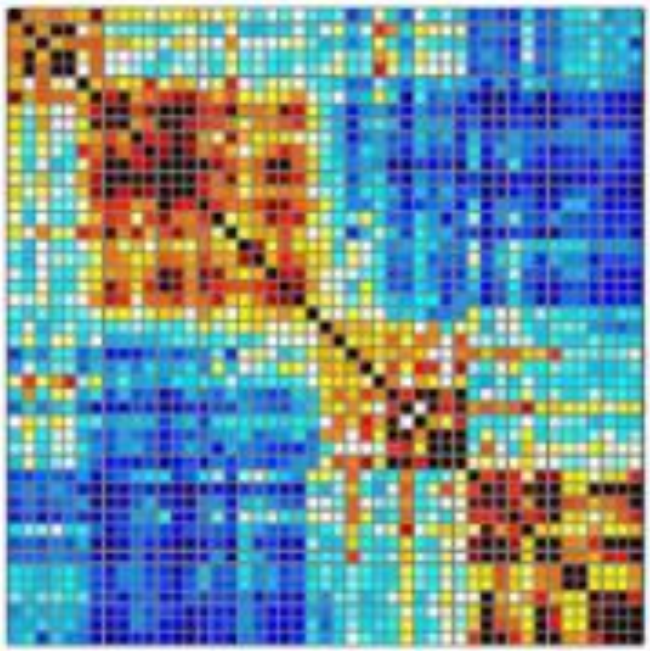
FUNCTIONAL COUPLING GRAPH ANALYSES



GRAPH ANALYSIS :

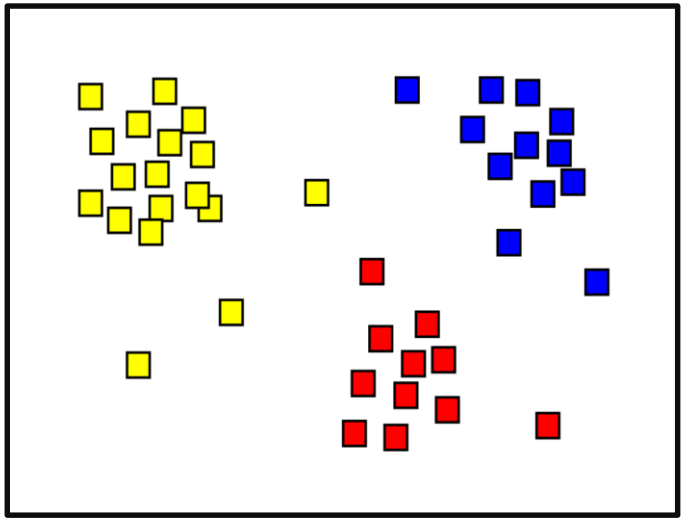
Undirected and weighted network based on the connectivity between eLORETA ROIs.

The nodes of the network are ROIs, the edges of the network are weighted by the lagged linear connectivity.



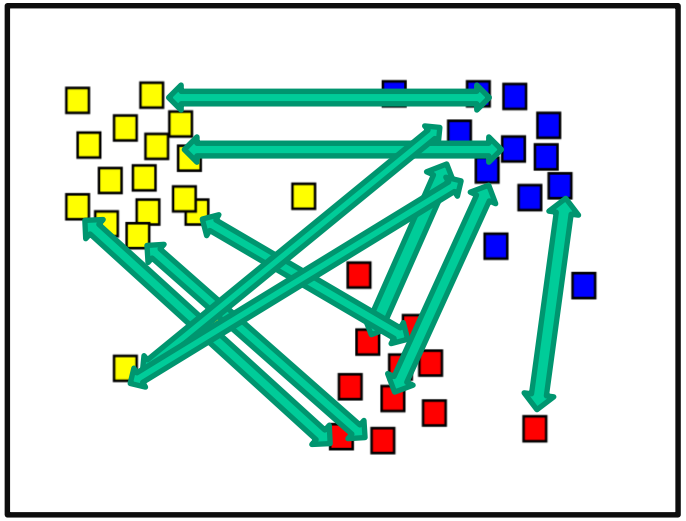
NETWORK PROPERTIES ARE DESCRIBED BY TWO MAIN PARAMETERS

SEGREGATION



CLUSTERING COEFFICIENT (C)

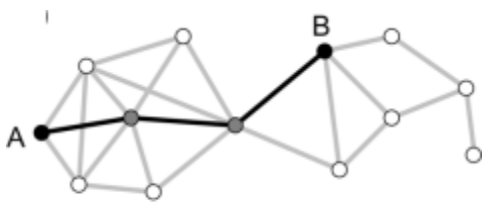
INTEGRATION



PATH LENGTH COEFFICIENT (L)

GRAPH THEORY PARAMETERS

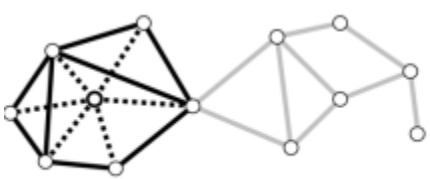
PRINCIPAL PARAMETERS: there is a broad range for the rewiring probability p where networks have clustering that is similar to that of the regular network, and a path length that is similar to that of the random network.



path length and distance

$$L = \frac{1}{n}\sum_{i \subset N} L_i = \frac{1}{n}\sum_{i \subset N} \frac{\sum_{j \subset N, j \neq i} d_{ij}}{n-1}$$

measure of integration, global connectedness



clustering coefficient

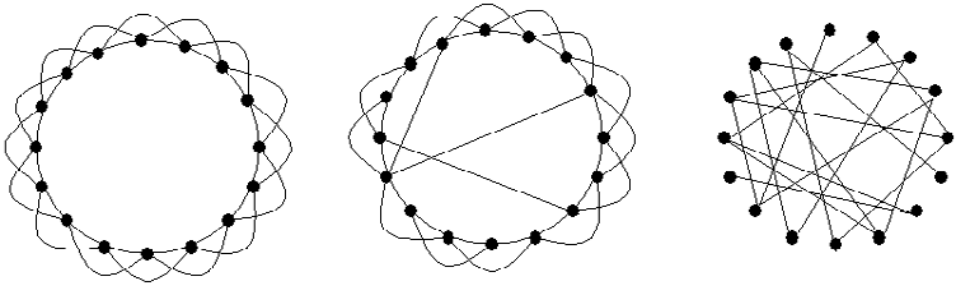
$$C = \frac{1}{n}\sum_{i \subset N} C_i = \frac{1}{n}\sum_{i \subset N} \frac{2t_i}{k_i(k_i-1)}$$

measures of segregation, local interconnectedness

GRAPH THEORY PARAMETERS

Watts and Strogatz [1998] showed that C and L parameters represent non-trivial aspects of connection patterns along the dimension ranging from highly ordered graphs (lattices, regular networks) to fully randomized graphs. Ordered graphs are characterized by high C and long L. Random graphs have short L and low C. By starting from ordered graphs and randomly reconnecting single edges with a rewiring probability P, Watts and Strogatz showed that while the average path length L drops quickly, clustering coefficient C showed resilience against reconnection. Within this range, networks exhibit **small-world attributes = σ = Cn/Ln** >1

SW = [C/Crand] / [L/Lrand] where Crand and Lrand are the mean clustering coefficient and the characteristic path length of the m random networks (Bassett and Bullmore, 2006). Regular network in which all cells are only coupled to their nearest neighbors (left). Small world network in which small numbers of connections are broken and rewired in order to make long-distance connections at random locations (middle). Random network with more long-distance connections (right), the network loses the property that most connections are local.



ordered small-world random

The study of connectivity in physiological aging

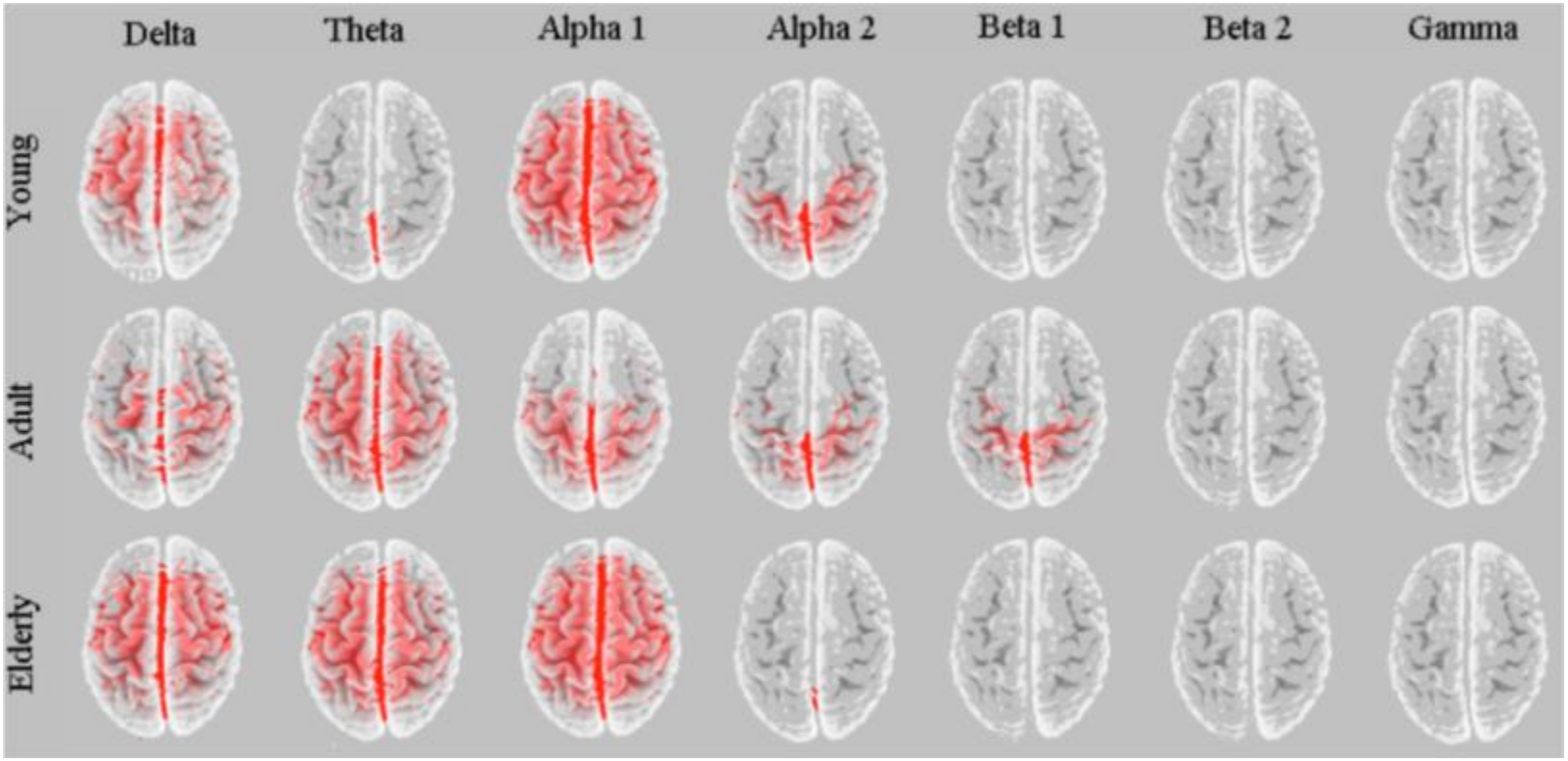
Human brain networks in physiological aging

Lagged linear coherence

$$LagR^2_{xy\omega} = \frac{[ImCov(x,y)]^2}{Var(x) \times Var(y) - [ReCov(x,y)]^2}$$

113 subjects

- 36 Young (aged 18-45 years)
- 46 Adult (aged 50-70 years)
- 31 Elderly (aged over 70 years).



Functional connectivity of cortical sources of cerebral rhythms

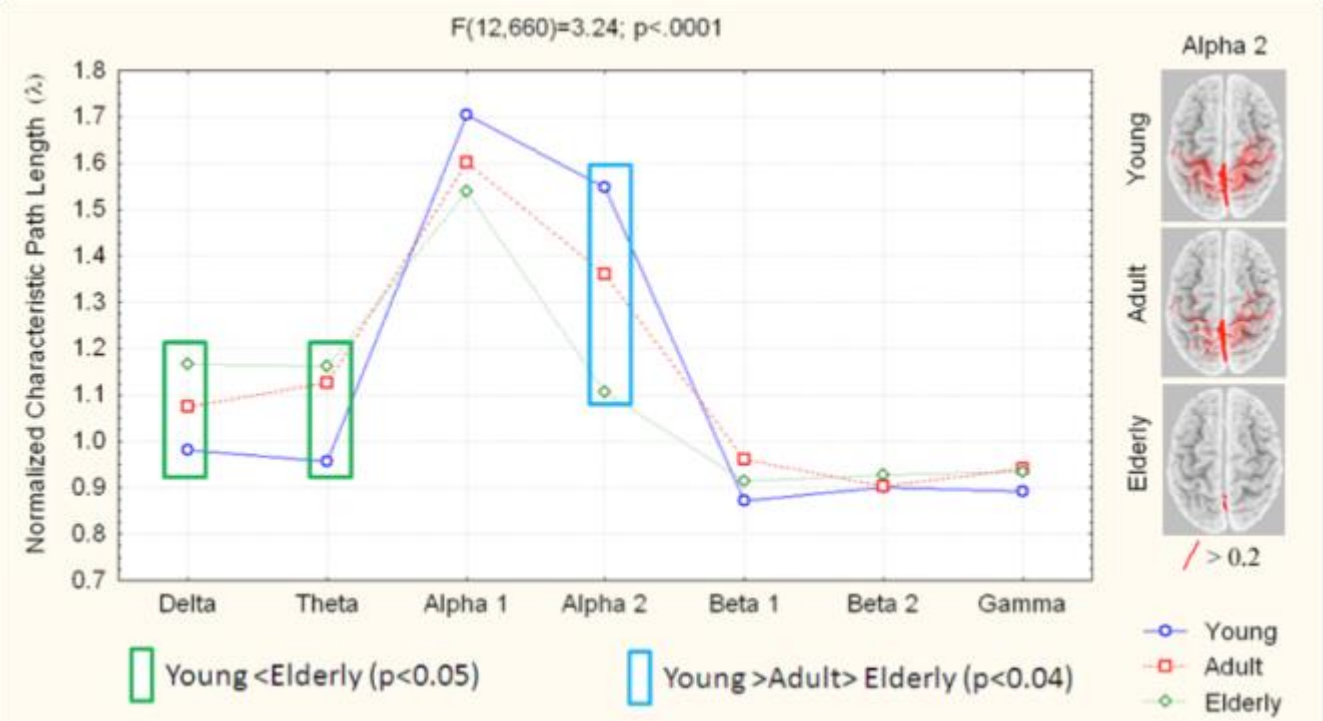
Human brain networks in physiological aging

Characteristic Path Length

113 subjects

- 36 Young (aged 18-45 years)
- 46 Adult (aged 50-70 years)
- 31 Elderly (aged over 70 years).

Vecchio F, Miraglia F, Bramanti P, Rossini PM. Human brain networks in physiological aging: a graph theoretical analysis of cortical connectivity from EEG data. J Alzheimers Dis. 2014 Jan 1;41(4):1239-49.



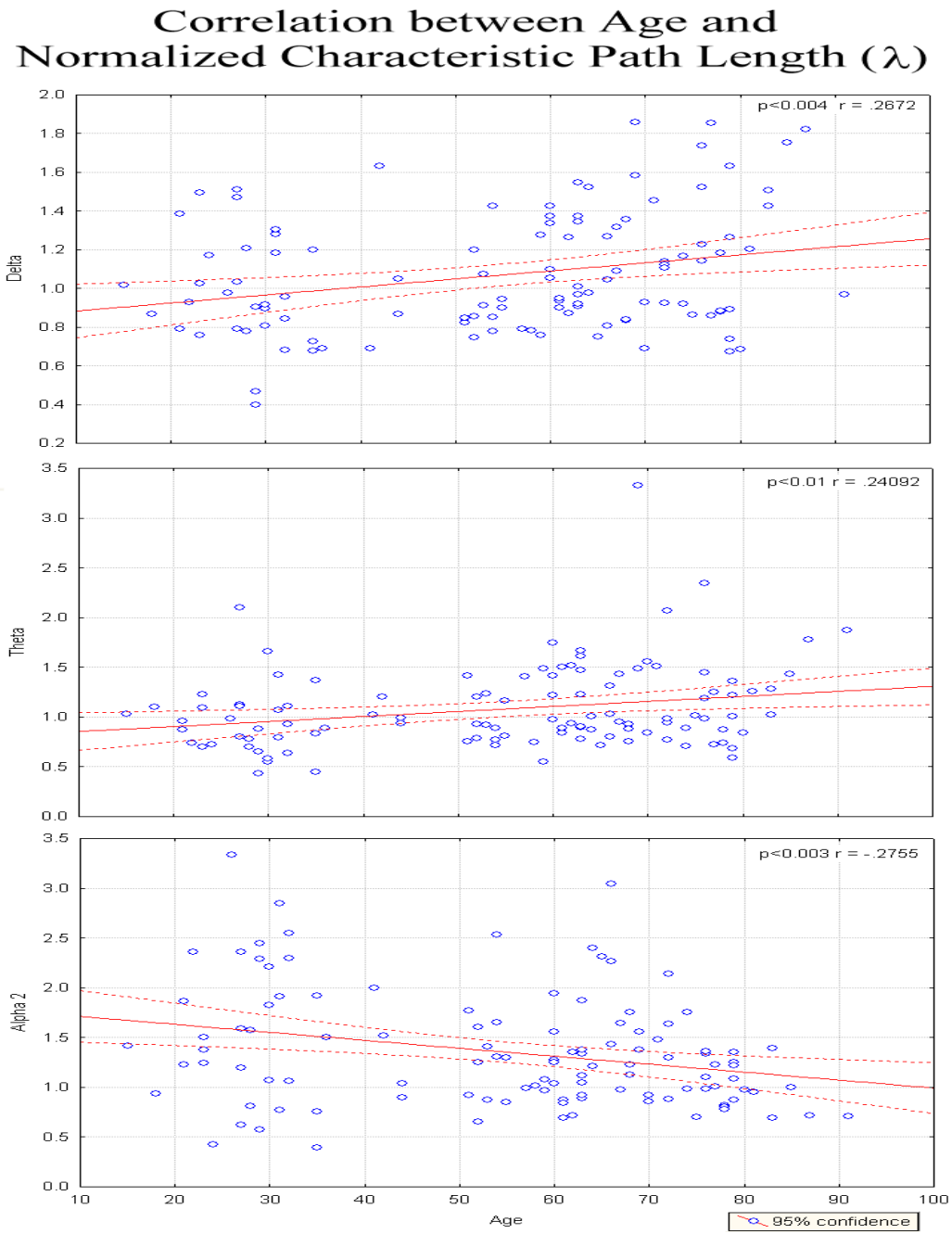
Aging processes provoke progressive disconnection among brain areas, as revealed by the increase of low and decrease of high frequency characteristic path length (λ), measuring the average shortest path length of a network as global index of how easy it is to travel from one part of the network to another.

Human brain networks in physiological aging

113 subjects

- 36 Young (aged 18-45 years)
- 46 Adult (aged 50-70 years)
- 31 Elderly (aged over 70 years).

Scatterplots showing the correlation between age and λ in the delta, theta and alpha 2 bands for all subjects as a whole group. The r- and p-values relative to the Pearson's correlation are reported within the diagram.



The study of connectivity in pathological aging

Rossini PM, Di Iorio R, Granata G, Miraglia F, Vecchio F.

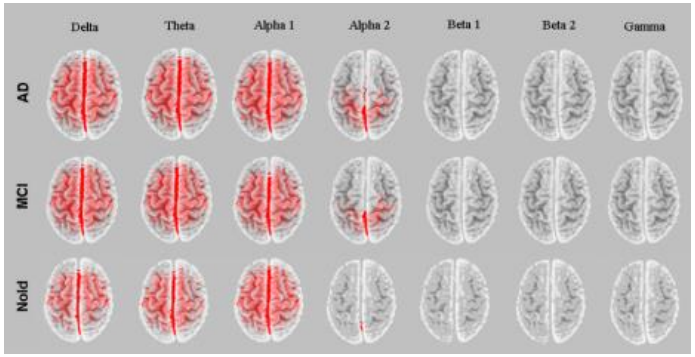
From Mild Cognitive Impairment to Alzheimer's Disease: A New Perspective in the "Land" of Human Brain Reactivity and Connectivity.

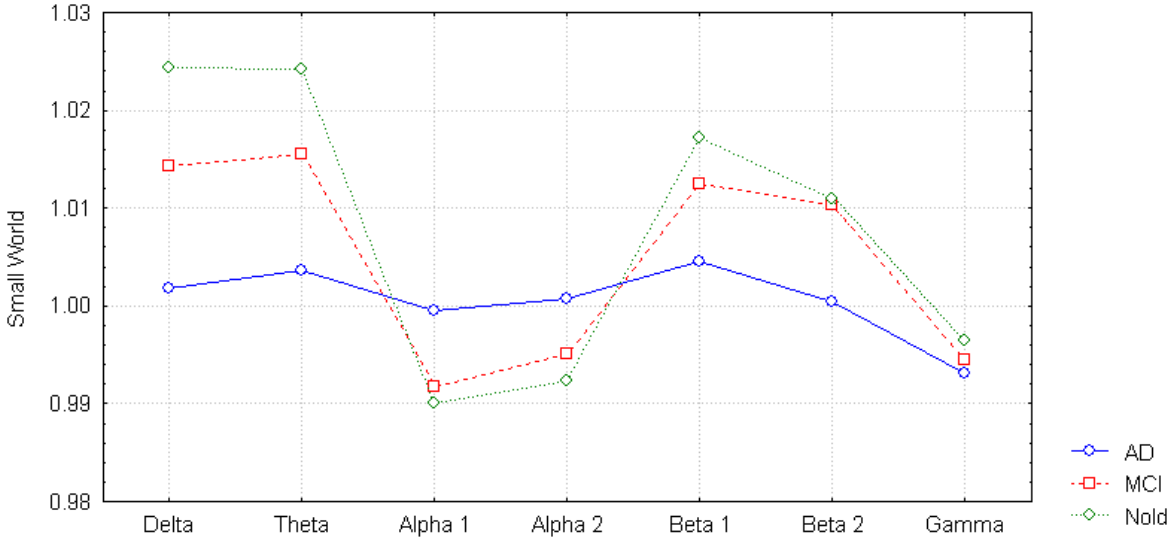
J Alzheimers Dis. 2016

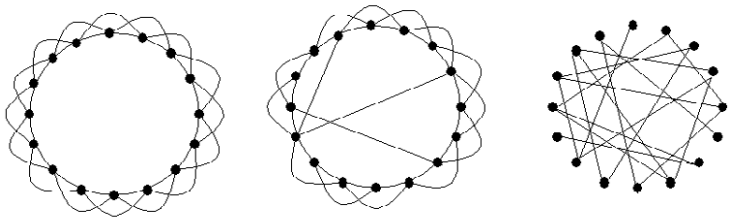
Human brain networks in cognitive decline

378 subjects

- 174 AD (MMSE 20.1)
- 154 MCI (MMSE 26.1)
- 50 Nold (MMSE 28.4)





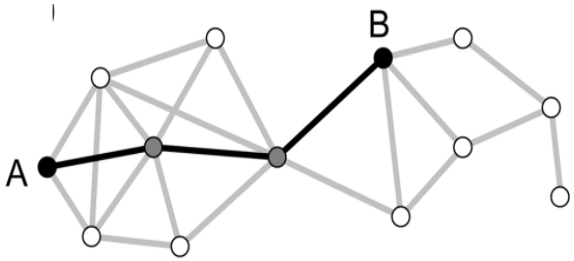


Vecchio F, Miraglia F, Marra C, Quaranta D, Vita MG, Bramanti P, Rossini PM. Human brain networks in cognitive decline: a graph theoretical analysis of cortical connectivity from EEG data. J Alzheimers Dis. 2014

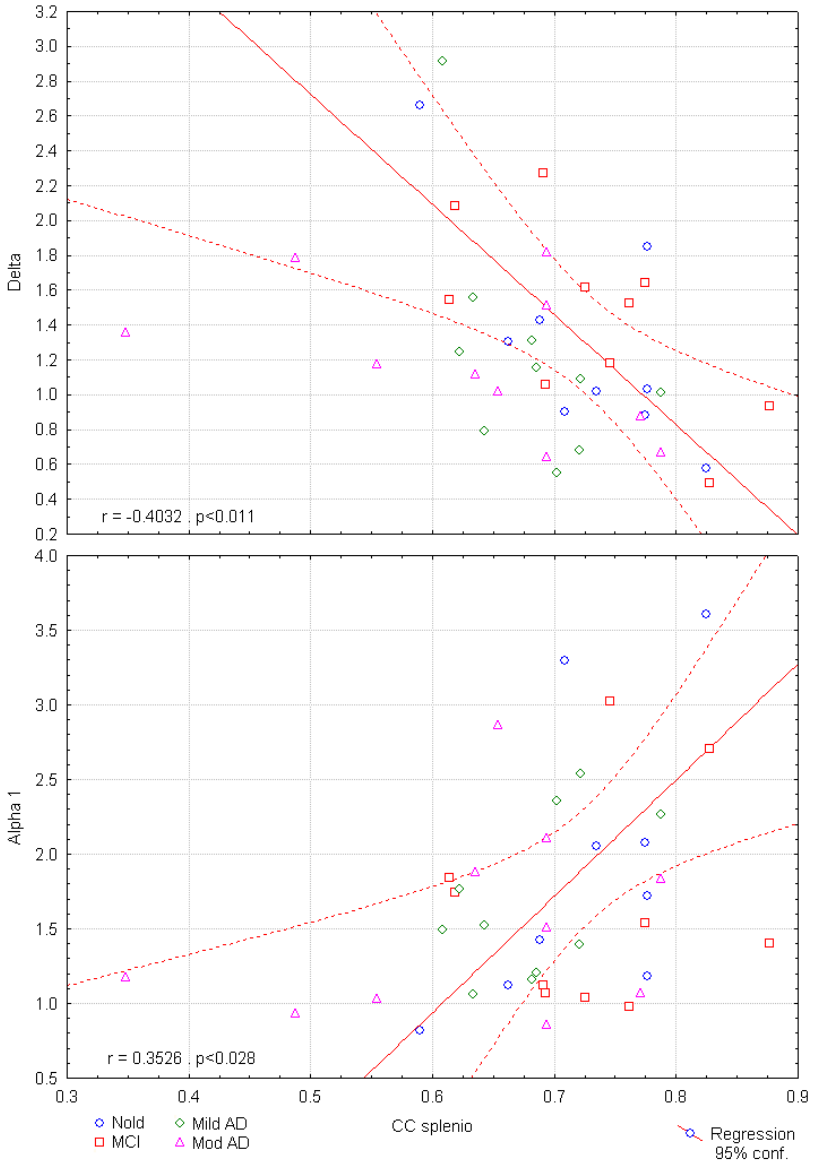
Vecchio F, Miraglia F, Quaranta D, Granata G, Romanello R, Marra C, Bramanti P, Rossini PM. Cortical connectivity and memory performance in cognitive decline: a study via graph theory from EEG data. Neuroscience. 2016

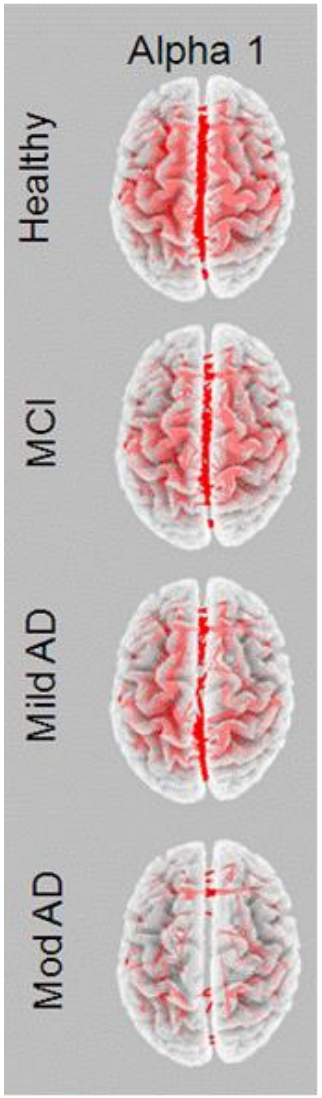
Human brain networks in cognitive decline

- 11 Moderate AD
- 10 Mild AD
- 10 MCI
- 9 Healthy



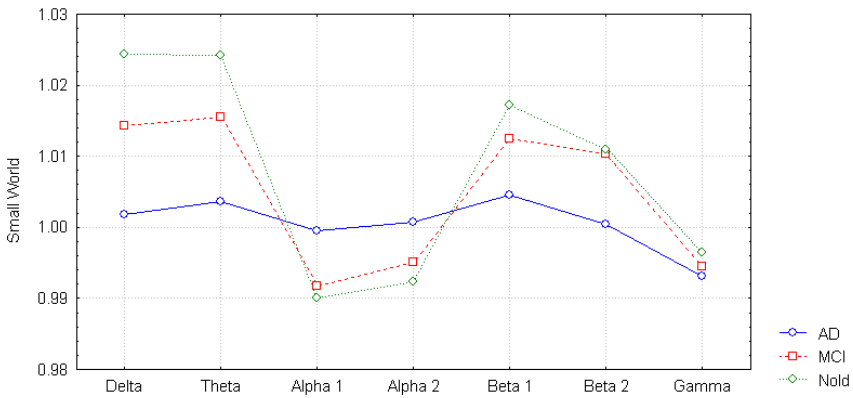
path length and distance

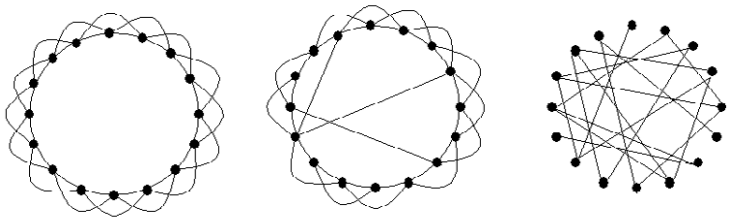




Vecchio F, Miraglia F, Curcio G, Altavilla R, Scrascia F, Giambattistelli F, Quattrocchi CC, Bramanti P, Vernieri F, Rossini PM. Cortical brain connectivity evaluated by graph theory in dementia: a correlation study between functional and structural data. J Alzheimers Dis. 2015

Human brain networks in cognitive decline

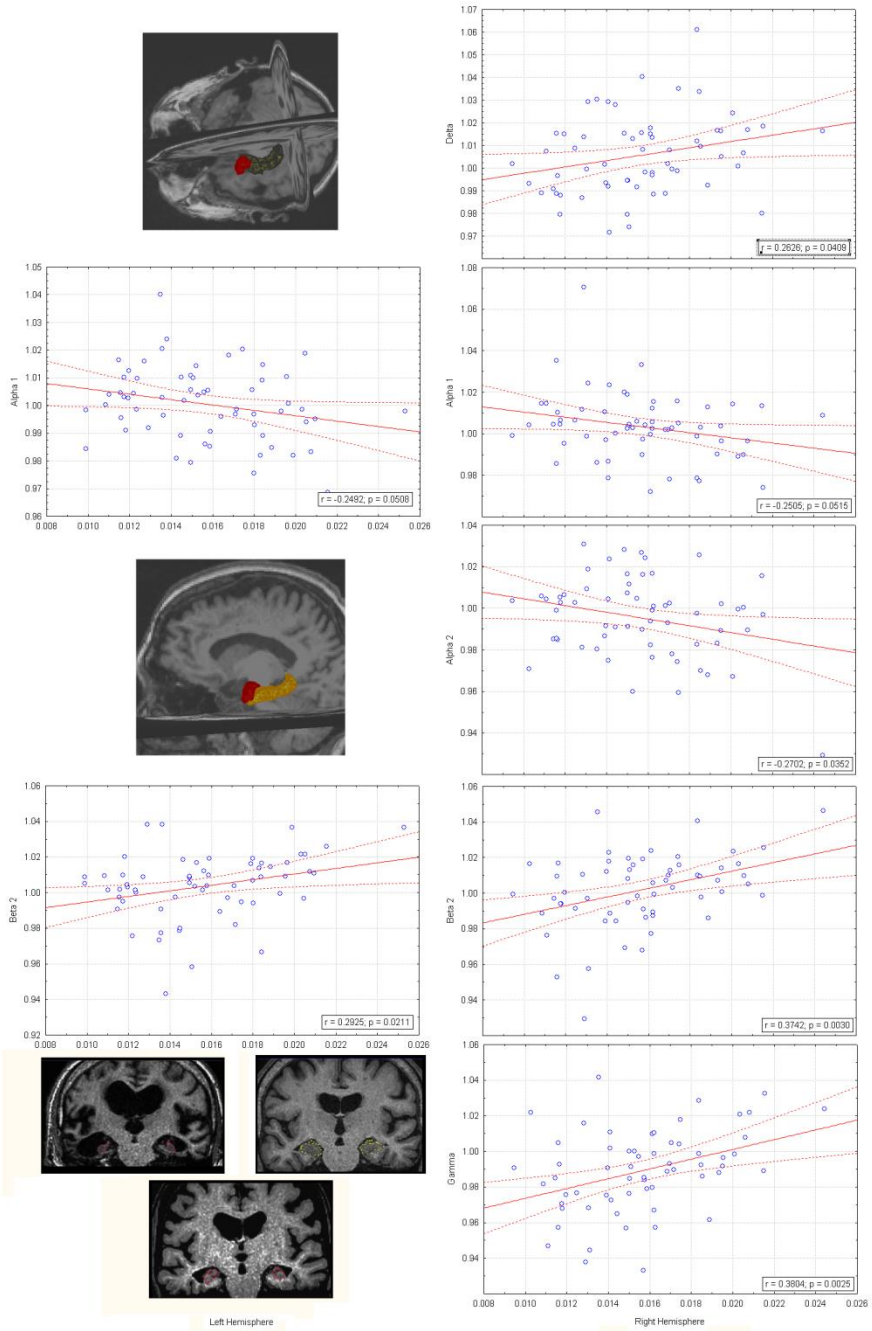




ordered small-world random

Demographic data of the participants

	Age (years)	Gender	Education (years)	MMSE
Nold	73.2 ±0.5	80 F	7 ±0.4	21.3 ±0.3
AD	73.8 ±0.7	20 F	8 ±0.9	29.8 ±0.3

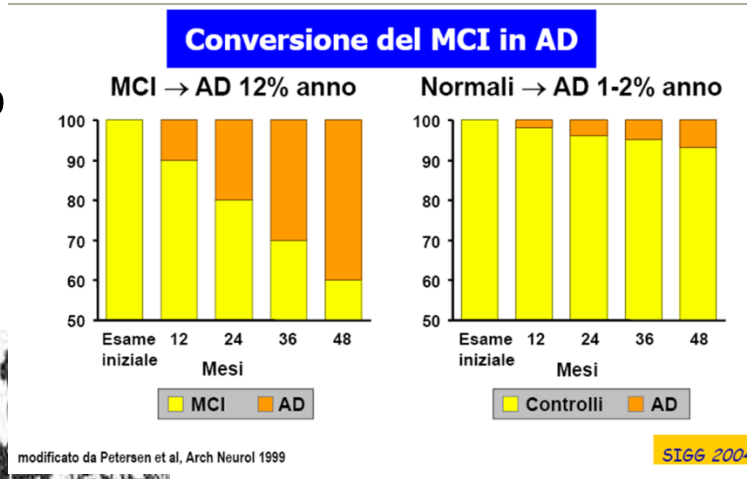


Vecchio F, Miraglia F, Piludu F, Granata G, Romanello R, Caulo M, Onofrj V, Bramanti P, Colosimo C, Rossini PM. "Small World" architecture in brain connectivity and hippocampal volume in Alzheimer disease: a study via graph theory from EEG data. Brain Imaging Behav. 2016

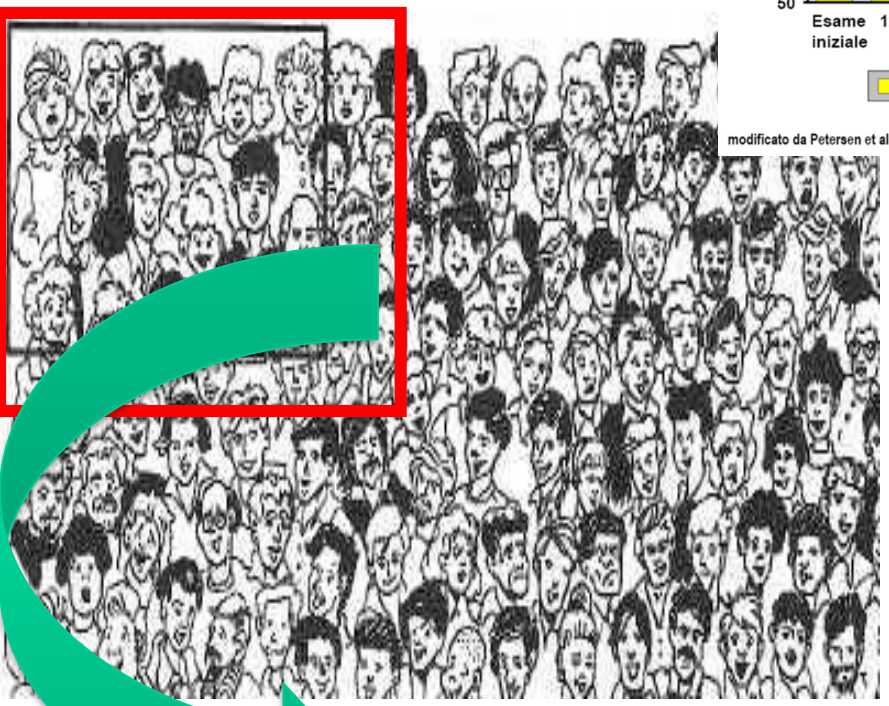
La grande sfida!

MCI (mild cognitive impairment) → **AD (Alzheimer's disease)**

Persone con lieve ma oggettivo declino cognitivo con elevato rischio di progredire verso l'Alzheimer



?

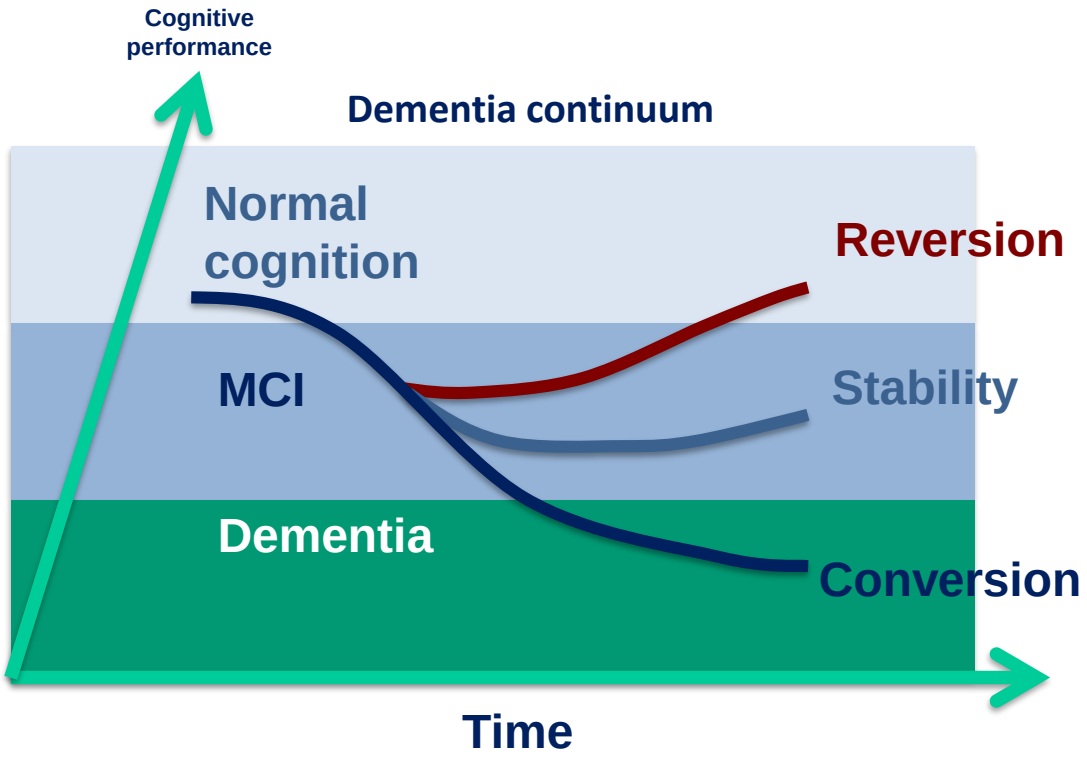


Età	% di conversione Anziani ad AD	% di conversione MCI ad AD
65 - 69	0,17 %	da 6% ad 38 %
85 - 89	3,86 %	

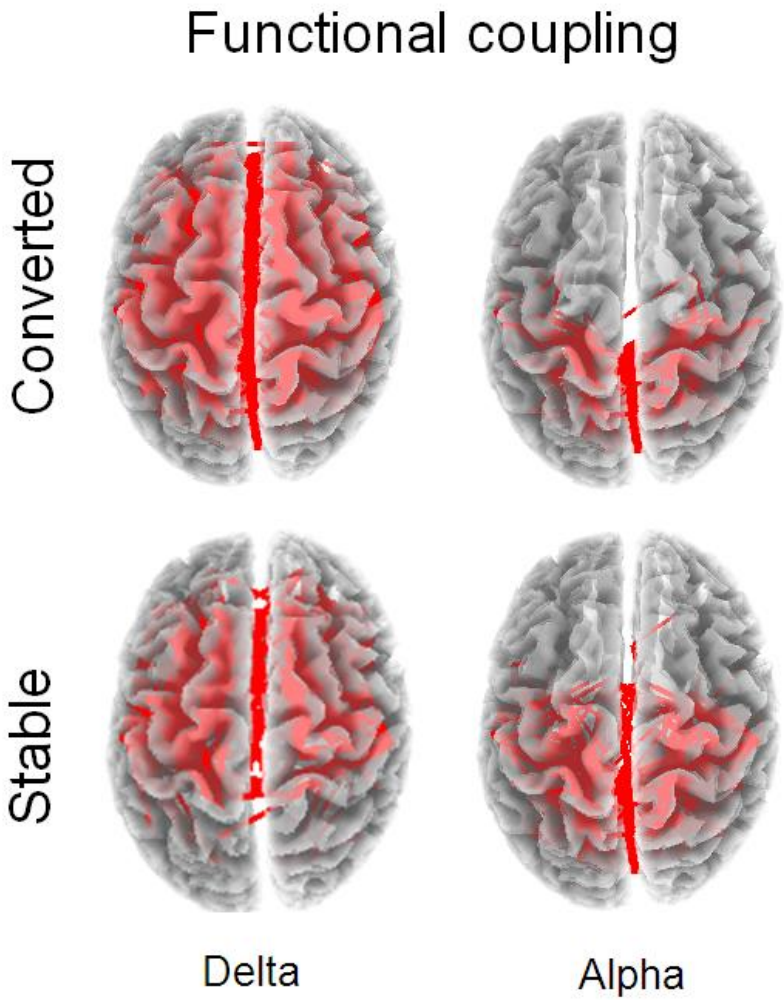
7-800.000 in Italia di cui il 50% prodromici di AD

An AD diagnosis is often made late in the disease continuum

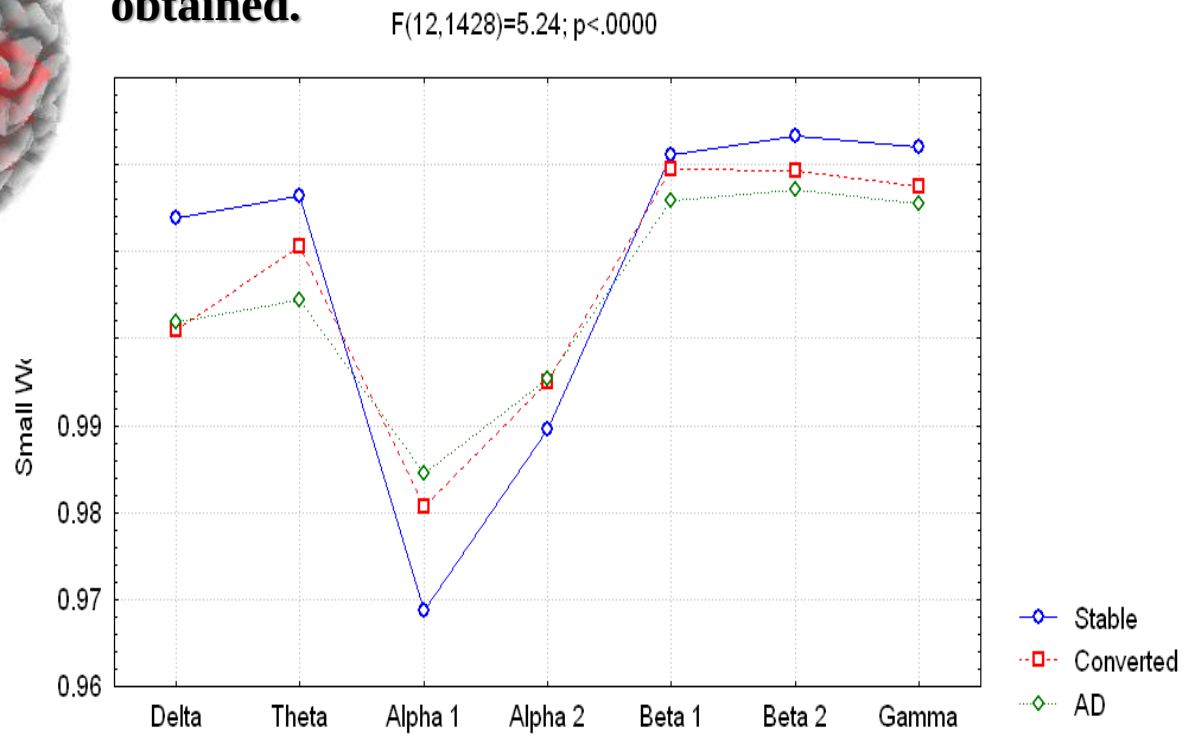
- **A more timely and accurate dementia diagnosis may reduce the impact of misdiagnosis**
- **A timely and accurate diagnosis provides access to a pathway of care and enables patients and their families to plan for the future**
- **Early diagnosis and treatment of patients with AD can improve the efficacy of the presently available drugs and of lifestyle changes leading to a better QoL, a longer patient's autonomy and reduced direct and indirect costs**

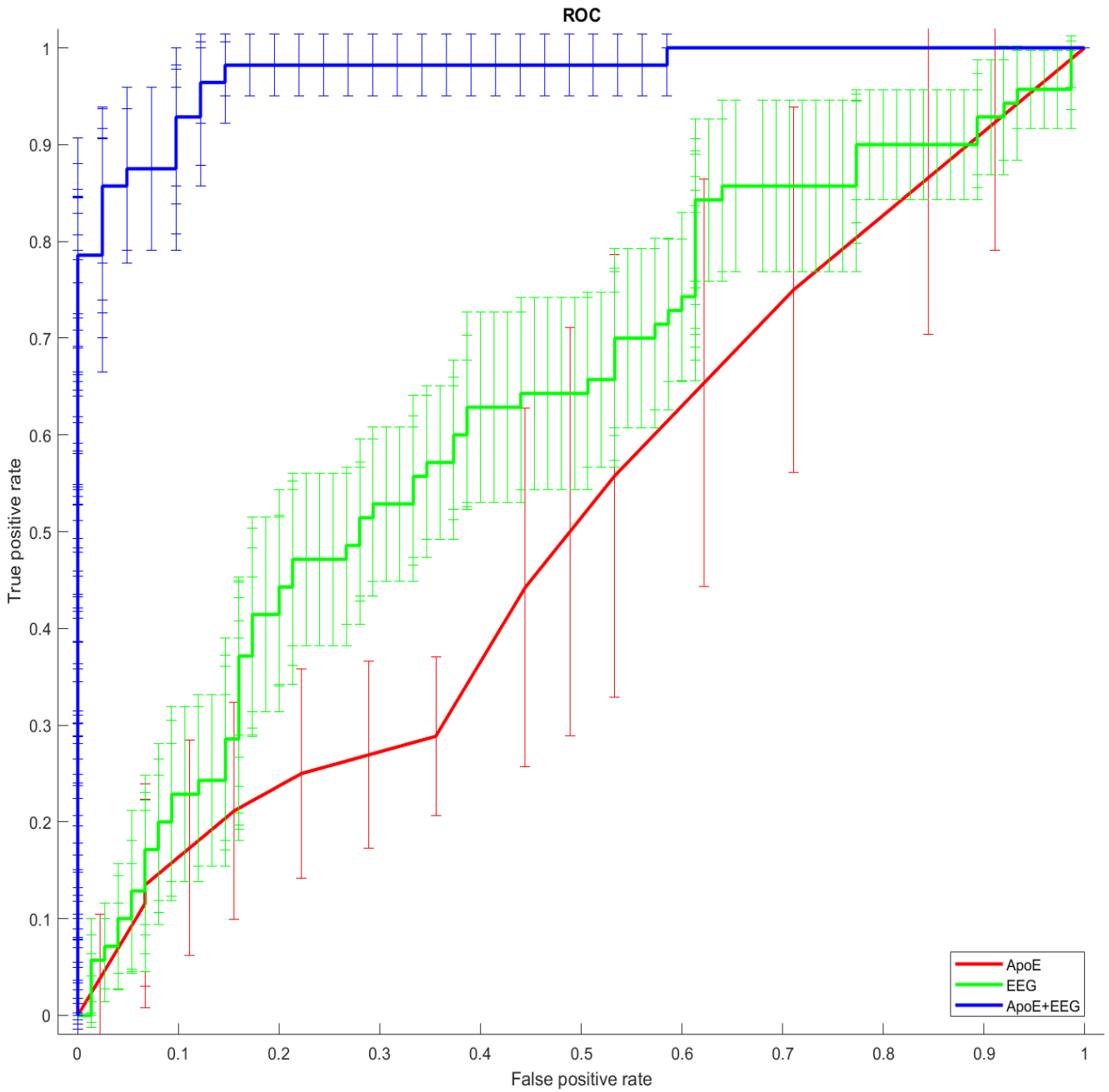


REVERSIONE ALLA NORMALITÀ DEL MILD COGNITIVE IMPAIRMENT: REVISIONE SISTEMATICA DELLA LETTERATURA E METANALISI. *Canevelli Marco, Grande Giulia, Lacorte Eleonora, Mariani Claudio, Bruno Giuseppe, Vanacore Nicola*



145 aMCI according to clinical follow-up, classified as Converted to AD (C-MCI, 71) or Stable (S-MCI, 74). Significant differences in SW organization in delta, alpha1, alpha2, beta2 and gamma bands, with C-MCI organization in baseline similar to that in AD. Receiver Operating Characteristic (ROC) curves, based on first-order polynomial regression of SW. In 97 MCI, ApoE alleles testing was also available. By adding this genetic risk factor, 96.7% sensitivity, 86% specificity and 91.7% accuracy (AUC=0.97) were obtained.




ROC
True positive rate
False positive rate
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
1
ApoE
EEG
ApoE+EEG

Biomarkers

Parte della “caratterizzazione” fenotipica di malattia:

- Modificazioni strutturali del cervello “visibili” alla RMN
- Modificazioni molecolari “visibili” con la PET
- Modificazioni molecolari “visibili” nel LCR
- Connettività EEG alterata
- Fattori di rischio geneticamente determinati
- Altri…

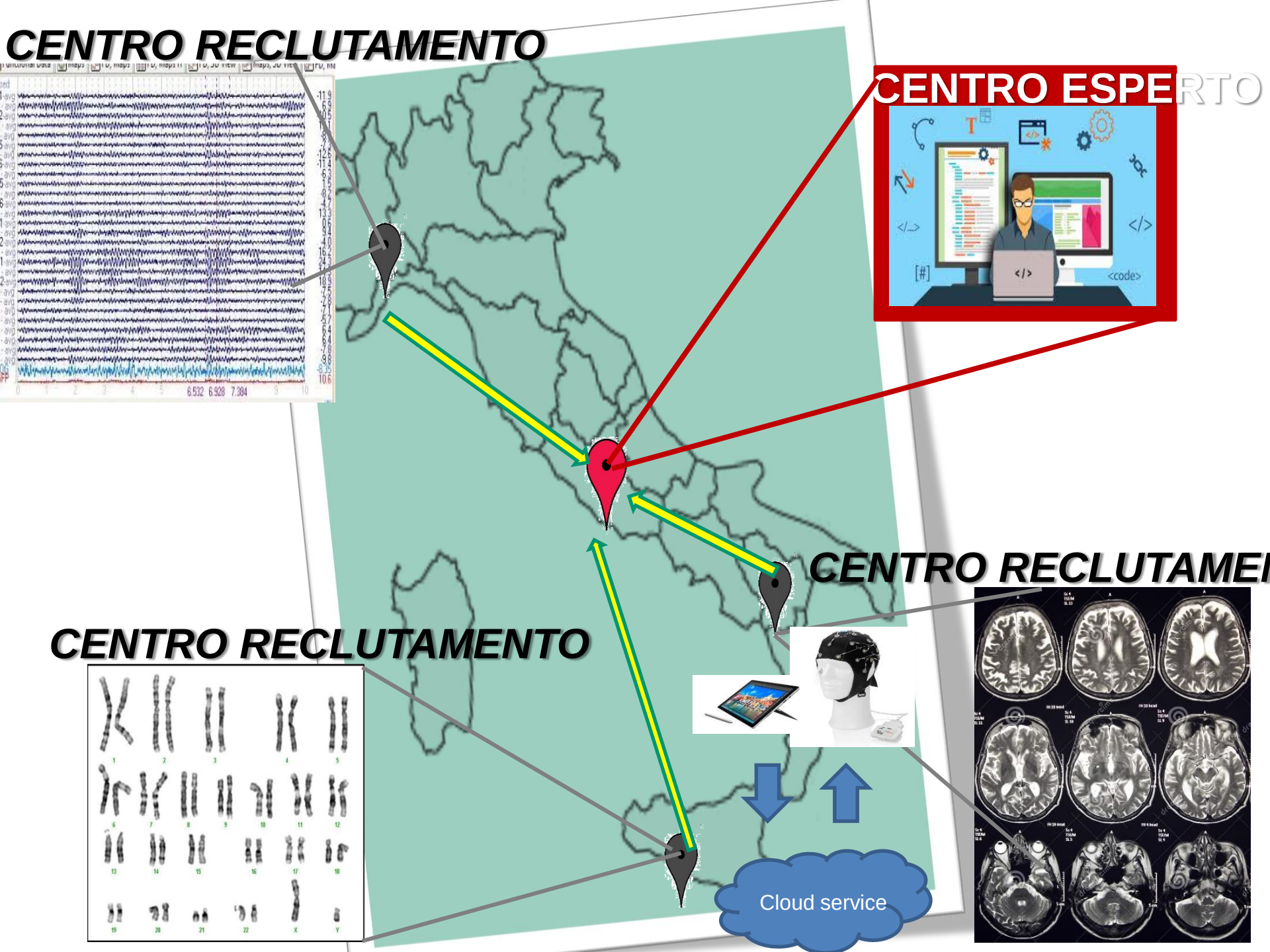
Strategic project promoted by Italian Medicines Agency (AIFA)

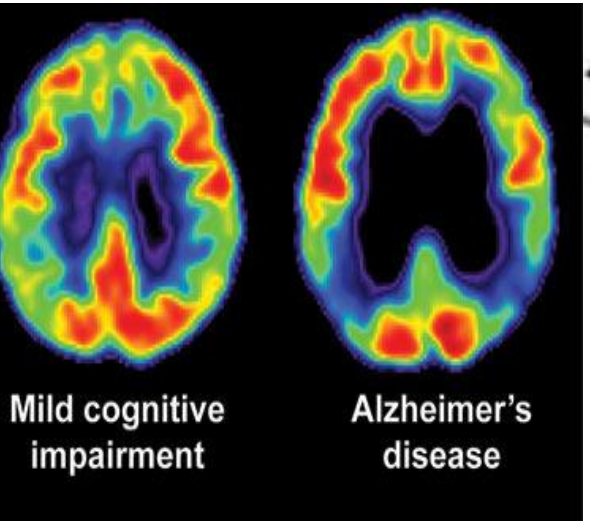
INTERCEPTOR PROJECT

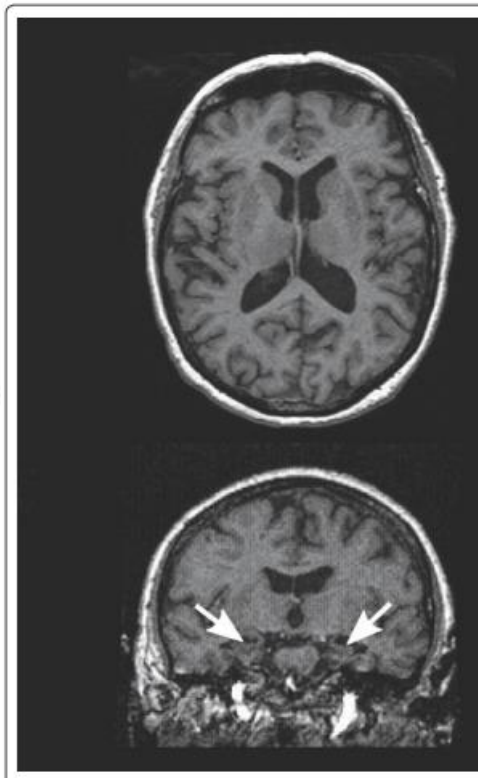
ON THE EARLY DIAGNOSIS OF THE PRODROMAL STAGE OF ALZHEIMER DISEASE. THE PROGRESSION FROM MILD COGNITIVE IMPAIRMENT (MCI) TO DEMENTIA: THE ROLE OF BIOMARKERS IN THE EARLY INTERCEPTION OF PATIENTS TO WHOM PROVIDE FUTURE DISEASE-MODIFYING DRUGS

Biomarcatore Ideale

alta accuratezza (specificità/sensibilità), non-invasività, disponibilità su territorio, costo sostenibile


CENTRO RECLUTAMENTO
CENTRO ESPERTO
CENTRO RECLUTAMENTO
CENTRO RECLUTAMENTO
Cloud service


Mild cognitive impairment
Alzheimer's disease




Cloud service

ENDPOINT PRIMARIO: Percentuale di conversione in 3.5 anni a demenza di Alzheimer. Inoltre si valuterà il biomarcatore o l'insieme di biomarcatori in grado di prevedere con la migliore accuratezza tale progressione e si validerà la piattaforma digitale per trasferimento dati.

Endpoints Secondari: Rapporto costi/benefici del biomarcatore o dell'insieme di biomarcatori in termini di previsione di progressione e di loro sostenibilità finanziaria, disponibilità sul territorio nazionale e non-invasività per i pazienti. Biorepository per testare nuovi biomarcatori.

Studio di coorte, longitudinale, multicentrico su un gruppo di pa.zienti MCI ed in cui si confrontano le caratteristiche al baseline e le risposte ai biomarcatori tra coloro che convertiranno nei 3.5 anni successivi a demenza di Alzheimer e quelli che rimarranno in una condizione di stabilità o di reversione al profilo cognitivo normale.

Numero di MCI da reclutare 400 da 20 Centri sul territorio nazionale. Durata del protocollo complessiva 54 mesi. Entro 60 giorni dal T0 (reclutamento sulla base della batteria dei TNP e dell'acquisizione del 'consenso informato') si debbono eseguire i seguenti marcatori "standard":

Neuropsicologici (MMSE e DRF – FCSRT)

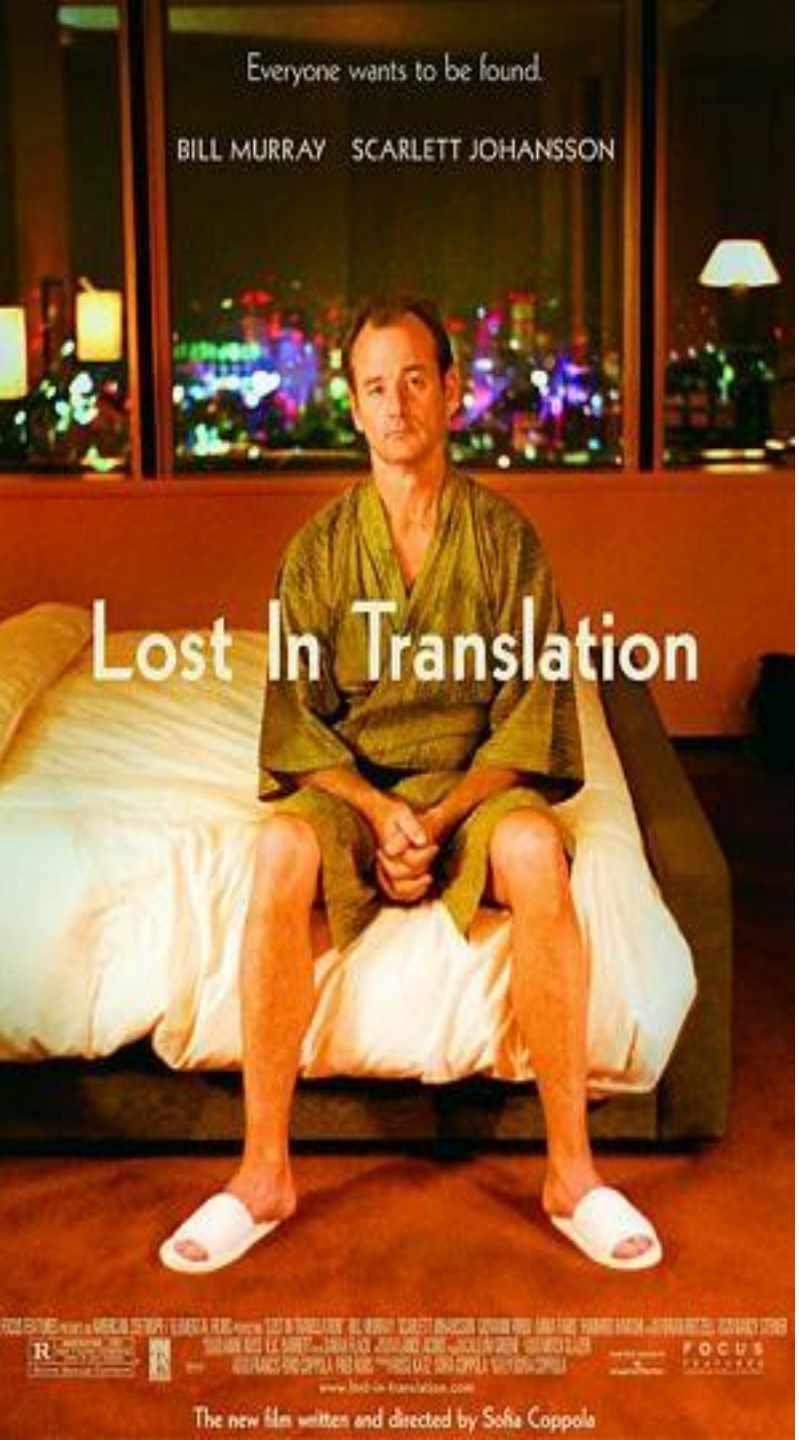
Neuroimmagini strutturali con MRI con volumetria ippocampale

Neuroimmagini funzionali con PET-FDG

Prelievo liquor per metaboliti Beta amiloide e Tau

Estrazione DNA e tipizzazione ApoE

EEG x studio connettività cerebrale



Rossini PM, Di Iorio R, Granata G, Miraglia F, Vecchio F.

From Mild Cognitive Impairment to Alzheimer's Disease: A New Perspective in the "Land" of Human Brain Reactivity and Connectivity.

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...lost in connection?