PATOGENESI MOLECOLARE DELLA MALATTIA DI ALZHEIMER: LA CASCATA DI AMILOIDE - NUOVE IPOTESI PATOGENETICHE: UN RUOLO PER LA DISFUNZIONE SINAPTICA NELLE FASI PRECOCI

Monica DiLuca
Universita’ di Milano
Brain Diseases in Europe

WHO- Global Burden of Diseases
50% disability world wide is related to brain diseases

In Europe
35% of the global burden is linked to brain diseases

The cost of brain diseases in Europe has been estimated in 2005: 386 billion euros

Cost of Brain Disorders in Europe
The cost of brain diseases in Europe in 2010 has been estimated 798 billion euros (Consensus document on European brain research DiLuca et al, EJN, 2011)

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Estimates in 2010</th>
<th></th>
<th>Estimates in 2004</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Number of subjects (million)</td>
<td>Costs per subject (€PPP, 2010)</td>
<td>Total costs (million €PPP, 2010)</td>
<td>Number of subjects (million)</td>
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<tr>
<td>Addiction</td>
<td>15.5</td>
<td>4227</td>
<td>65,684</td>
<td>9.2</td>
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<tr>
<td>Anxiety disorders</td>
<td>51.3</td>
<td>1076</td>
<td>65,995</td>
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<tr>
<td>Brain tumor</td>
<td>0.24</td>
<td>21,590</td>
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<td>Dementia</td>
<td>6.3</td>
<td>16,584</td>
<td>105,163</td>
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<td>Epilepsy</td>
<td>2.6</td>
<td>5221</td>
<td>13,800</td>
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<tr>
<td>Migraine</td>
<td>49.9</td>
<td>370</td>
<td>18,463</td>
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<tr>
<td>Mood disorders</td>
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<td>3406</td>
<td>113,405</td>
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<td>Multiple sclerosis</td>
<td>0.54</td>
<td>26,974</td>
<td>14,559</td>
<td>0.38</td>
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<td>Parkinson’s disease</td>
<td>1.2</td>
<td>11,153</td>
<td>13,933</td>
<td>1.2</td>
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<tr>
<td>Psychotic disorders</td>
<td>5.0</td>
<td>5805</td>
<td>29,007</td>
<td>3.7</td>
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<tr>
<td>Stroke</td>
<td>1.3</td>
<td>21,000</td>
<td>26,641</td>
<td>1.1</td>
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<tr>
<td>Traumatic brain injury</td>
<td>1.2</td>
<td>4209</td>
<td>5085</td>
<td>0.71</td>
</tr>
<tr>
<td>Total</td>
<td>178.5</td>
<td>2672</td>
<td>476,911</td>
<td>127.0</td>
</tr>
</tbody>
</table>

1Referred to as “affective disorders” in 2005, 2includes only incident cases in 2010, 3weighted mean from all countries and diagnoses including also persons with zero costs, 4excluding indirect costs, 7excluding PTSD.
Outline

✓ The amyloid cascade: pros and cons
✓ Do we have new clues? Is Alzheimer Disease a synaptopathy?
✓ How basic research can help patients’ management: biomarkers?
La patogenesi: la cascata di amiloide
Alzheimer’s Disease: 100 years and beyond

- 1906: Aβ sequence
- 1984: APP gene
- 1986: -tau
- 1987: γ-secretase complex
- 1995: PS1-PS2
- 1999: BACE, ADAM10

Case closed. Plaques and tangles in the brain of Auguste D.
APP metabolism: a mutually exclusive dance between alpha- and beta-secretase
amyloid plaque

amyloid precursor protein (APP)

CD147
(γ-secretase regulatory subunit)

β-secretase cleavage site

Psn, Nct, APH-1, PEN-2
(γ-secretase core complex)

amyloid β-peptide
Nicastrin modulates presenilin-mediated notch/glp-1 signal transduction and βAPP processing

Figure 1. γ-Secretase processing of amyloid precursor protein C-terminal fragments (CTFs). A cartoon illustration shows the various subunits of the γ-secretase enzyme complex performing a number of different putative γ-secretase–mediated proteolytic activities. Assembly and maturation of the γ-secretase enzyme begins with the formation of a binary complex between nicastrin and anterior pharynx defective 1 (Aph-1). Presenilin (PS) binds to the nicastrin/Aph-1 complex to form an inactive trimeric complex. The PS N-terminal fragment (NTF) and the PS-CTF collectively harbor the catalytic center of γ-secretase, which requires binding of presenilin enhancer 2 (Pen-2) for the endoproteolysis of PS into the PS-NTF and the PS-CTF, rendering activation of γ-secretase.
ALL proteolytic products have a functional role
A novel gene in the amyloid cascade

Rogaeva et al., 2007 … when SORL1 is underexpressed, APP is sorted into Abeta-generating compartments
The complexity of amyloid hypothesis

Tanzi and Bertram, Cell, 2005
Is amyloid the main cause of Alzheimer’s disease?
Is amyloid the main cause of Alzheimer’s disease?

Why addressing the question?
Dissociation of Neuropathologic Findings and Cognition

Case Report of an Apolipoprotein E ε2/ε2 Genotype

Daniel J. Berlau, PhD; Kristin Kahle-Wrobleski, PhD; Elizabeth Head, PhD; Matthew Goodus, BA; Ronald Kim, MD; Claudia Kawas, MD

Figure 2. Neuropathologic features in the case subject compared with matched control subjects. Note pathologic features in area CA1/subiculum of the hippocampus in the case subject. β-Amyloid deposition is absent in area CA1/subiculum in a 90-year-old woman without dementia (A), primarily diffuse in the case subject (B), and associated with extracellular neurofibrillary tangles and compact neuritic plaques in a 94-year-old woman with dementia (C). Neurofibrillary tangle accumulation was sparse in the control subject without dementia (D) but was comparable in the case subject (E) and the control subject with dementia (F). AD indicates Alzheimer disease; bar, 100 μm.
Is amyloid the main cause of Alzheimer’s disease?
# Pros and Cons

<table>
<thead>
<tr>
<th>Pros</th>
<th>Cons</th>
</tr>
</thead>
<tbody>
<tr>
<td>A limited number of patients with Abeta accumulation fail to show cognitive deficits</td>
<td>All familial forms of AD are linked to mutation of genes involved in the amyloid cascade</td>
</tr>
<tr>
<td>Transgenic animals characterized by Abeta accumulation not always show a parallel cognitive impairment</td>
<td>Clearance of Abeta in mice reverts cognitive deficits</td>
</tr>
<tr>
<td>Abeta 1-42 is found in CSF of non-demented individuals</td>
<td>Different forms of Abeta are neurotoxic in in vitro and in vivo models</td>
</tr>
</tbody>
</table>
Is amyloid the main cause of Alzheimer’s disease?

✓ Which?
✓ How much?
✓ Where?
Which?

A key element of complexity: Abeta assembly
Which?

A key element of complexity: Abeta assembly

<table>
<thead>
<tr>
<th>Oligomeric assembly</th>
<th>Characteristics</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protofibril (PF)</td>
<td>Intermediates of synthetic Aβ fibrillization; up to 150 nm in length and ~5 nm in width; β-sheet structure: bind Congo red and Thioflavin T</td>
<td>54–57</td>
</tr>
<tr>
<td>Annular assemblies</td>
<td>Doughnut-like structures of synthetic Aβ; outer diameter of ~8–12 nm; inner diameter of ~2.0–2.5 nm</td>
<td>58,59</td>
</tr>
<tr>
<td>Aβ-derived diffusible ligands (ADDLs)</td>
<td>Synthetic Aβ oligomers smaller than annuli; might affect neural signal-transduction pathways</td>
<td>60,61</td>
</tr>
<tr>
<td>Aβ*56</td>
<td>Apparent dodecamer of endogenous brain Aβ; detected in the brains of an APP transgenic mouse line and might correlate with memory loss</td>
<td>62</td>
</tr>
<tr>
<td>Secreted soluble Aβ dimers and trimers</td>
<td>Produced by cultured cells; resistant to SDS; resistant to the Aβ-degrading protease IDE; alter synaptic structure and function</td>
<td>63,64,69</td>
</tr>
</tbody>
</table>

Aβ, amyloid β-protein; APP, β-amyloid precursor protein; IDE, insulin-degrading enzyme.

Haass and Selkoe *Nature Reviews Molecular Cell Biology*, 2007
Is amyloid the main cause of Alzheimer’s disease?

✓ Which? The effect of different forms of Abeta aggregates on excitatory synaptic function

✓ Where?

✓ How much?
The glutamatergic synapse

The positive effect of monomers on synaptic function
Brief Communications

β-Amyloid Monomers Are Neuroprotective

María Laura Gluffrida, Filippo Caraci, Bruno Pignataro, Sebastiano Cataldo, Paolo De Bona, Valeria Bruno, Gemma Molinari, Giuseppe Pappalardo, Angela Messina, Angelo Palmiglione, Domenico Garozzo, Ferdinando Nicoletti, Enrico Rizzarelli, and Agata Copani
A key element of complexity: Abeta assembly

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</tr>
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Aβ, amyloid β-protein; APP, β-amyloid precursor protein; IDE, insulin-degrading enzyme.

Haass and Selkoe *Nature Reviews Molecular Cell Biology*, 2007
Mnomers vs oligomers:

When friends become enemies
Aβ star: a light onto synaptic dysfunction?

Aβ '56, most likely a tetramer of Aβ trimers, is the prime candidate suspected of causing early memory deficits in a mouse model of Alzheimer disease.

Lesne et al., Nature, 2006
Soluble A beta extracted from Alzheimer’s disease brain alter hippocampal synaptic physiology

Shankar et al, Nat Med 2008
How much?
Is amyloid the main cause of Alzheimer’s disease?

✓ Which? Dimers, oligomers
✓ How much? In the range of nanomol
✓ Where?
Fig. 1 Immunochemical analysis of cerebellum and frontal cortex sections using antibodies specific for Aβ40 or Aβ42. a, b Cerebellum from CTRL-2 (control case, black square in Table 1), c, d cerebellum from AD-1 (sporadic AD case, white circle in Table 1), e, f cerebellum from APPswe-1 (Swedish APP case, white triangle in Table 1).

Hypothesis that memory loss in AD is an oligomer-induced glutamatergic synaptic failure

ADDLs (small, diffusible oligomers of Abeta) colocalize with synapses. (Lacor et al., 2004).

A role for Aβ soluble oligomeric and protofibrillar forms in dysregulation of synaptic function (Walsh et al., Nature 2002; Lesne et al., Nature, 2006)
Palop & Mucke, Nat. Neuroscience 2010
Amyloid is the main cause of Alzheimer’s disease
The tau protein

Querfurth H W, La Ferla FM, NEJM, 2010
The amyloid cascade hypothesis for Alzheimer's disease: an appraisal for the development of therapeutics

Karran et al, Nature Reviews Drug Discovery 10, 698-712 (September 2011)
Le nuove ipotesi: alla ricerca di nuovi target farmacologici
The ups and downs of Aβ  
Dennis J Selkoe, Nat Med 2006  

Complexity of AD molecular pathogenesis  
Several molecular pathways are involved  

1906  
1984  
1986  
1987  
1988  
1989  
1991  
1995  
2002  

Aβ sequence  
APP gene  
tau  
BACE, ADAM10  

1987  
1991  
1995  
1999  

Synapse loss is the major correlate of cognitive impairment.  
AD is a synaptic failure  

Alzheimer disease  
as a synaptopathy  

Nongenetic elevators of Aβ levels:  
- Neuronal electrical activity in general  
- Synaptic activity, with enhanced vesicle exocytosis  

Genetic elevators of Aβ levels:  
- APP mutations  
- APP duplication  
- Presenilin mutations  
- Apolipoprotein E4 allele  

Exocytic vesicle containing Aβ peptides  
Trimer of Aβ  
Dimmer of Aβ  
Released Aβ in interstitial fluid  

APP  
β-secretase cut  
γ-secretase cut
A correct functioning of a synapse is achieved via highly ordered biochemical organization. Modifications can underlie dysregulation associated with neurological and psychiatric disorders. “Synaptopathy” is a way of describing disease states that are primarily driven from dysfunction in the synapse or in a given synaptic sub-compartment.
The glutamatergic synapse

Glu

AMPA

NMDA

mGluR

Na⁺

Ca²⁺

Gardoni et al, JN 2001; JBC 2003; JBC 2004; JN 2006; JN 2009
Annu. Rev. Biochem. 76:823–47
A number of synaptic genes/proteins mutated in neurological and psychiatric disorders are known

Mutations in the gene encoding the synaptic scaffolding protein SHANK3 are associated with autism spectrum disorders

A new function for the fragile X mental retardation protein in regulation of PSD-95 mRNA stability

Neuroligin-3-deficient mice: model of a monogenic heritable form of autism with an olfactory deficit.
How/if early stages of neurodegenerative diseases involve synaptic “traits”? 
A key element of complexity: Abeta assembly
Hypothesis that memory loss in AD is an oligomer-induced glutamatergic synaptic failure

**ADDLs** (small, diffusible oligomers of Abeta) colocalize with synapses. (Lacor et al., 2004).

A role for Aβ soluble oligomeric and protofibrillar forms in dysregulation of synaptic function (Walsh et al., Nature 2002; Lesne et al., Nature, 2006)
The synapse in Alzheimer Disease

Figure 3. Synaptic Dysfunction in Alzheimer’s Disease.

Querfurth H W, La Ferla FM, NEJM, 2010
Oligomers: how do they work on spines and synapses?

Natural Oligomers of the Alzheimer Amyloid-β Protein Induce Reversible Synapse Loss by Modulating an NMDA-Type Glutamate Receptor-Dependent Signaling Pathway

Ganesh M. Shankar,1,2 Brenda L. Bloodgood,1 Matthew Townsend,2 Dominic M. Walsh,3 Dennis J. Selkoe,2 and Bernardo L. Sabatini
J Neurosci, 2007

Amyloid-β protein dimers isolated directly from Alzheimer’s brains impair synaptic plasticity and memory

Ganesh M Shankar1,2, Shaomin Li1, Tapan H Mehta1, Amaya Garcia-Munoz3, Nina E Shepardson1, Imelda Smith4, Francesca M Brett5, Michael A Farrell3, Michael J Rowan4, Cynthia A Lemere1, Ciaran M Regan3, Dominic M Walsh4, Bernardo L Sabatini2 & Dennis J Selkoe1

NATURE MEDICINE VOLUME 14 | NUMBER 8 | AUGUST 2008
Spine loss in AD
Spines and patients

La spina dendritica: il sito delle sinapsi eccitatorie
# Timing Is Everything, Even for Cholinergic Control

**Darwin K. Berg**

Neuron, 2011

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**Figure 1.** Schematic showing the timing dependence of cholinergic input from the septum in determining the kinds of synaptic plasticity found at glutamatergic synapses formed by SCs onto CA1 pyramidal neurons in the hippocampus.

<table>
<thead>
<tr>
<th>Induction Protocol (IP)</th>
<th>PSC before IP</th>
<th>PSC after IP</th>
<th>Outcome</th>
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<tr>
<td>Chol/SC</td>
<td></td>
<td></td>
<td>LTP (a7-nAChR)</td>
</tr>
<tr>
<td>CHOL/SC</td>
<td></td>
<td></td>
<td>STD (a7-nAChR)</td>
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<tr>
<td>CHOL/SC</td>
<td></td>
<td></td>
<td>No change</td>
</tr>
<tr>
<td>CHOL/SC</td>
<td></td>
<td></td>
<td>LTP (mAChR)</td>
</tr>
</tbody>
</table>
La spina dendritica: sede dei fenomeni di plasticità
Spine and synapse dynamic

Linking new genes to pathophysiology of AD

Sleegers K. et al, Trends in genetics 2009
Model of molecular mechanisms involved in spine pathology in Alzheimer's disease

Penzes et al, Nat Neurosci 2011
Elements of the amyloid cascade are located at the synapse
ADAM10 binds SAP97

**ADAM10**
- Signal peptide
- Pro-region
- Disintegrin domain
- Metalloproteinase domain
- Zn
- EGF repeat
- Cytoplasmic tail

**SAP97**
- PDZ1
- PDZ2
- PDZ3
- SH3
- GK
- 911

Marcello, Gardoni et al., J Neurosci, 2007
**ADAM10**

- It is a type I *membrane* protein, a member of the disintegrin and metalloproteases family,

- It is a component of the excitatory post synaptic density, where it binds a member of PSD-MAGUK, SAP97

- It is a shaddase towards many substrates, i.e. **APP**, **Notch**, **N-cadherins**, ILR6

The complexity of spine formation and synapse stability

ADAM10/SAP97: quale impatto sulla patologia?
Neuropathological features in ADAM10/SAP97 uncoupled mice

Epis, Marcello et al, *Brain* 2010
LTP and LTD modulate ADAM10 interaction with its binding partners.
ADAM10/SAP97/AP2 in human: a true novel pathogenic pathway?

Dalla biologia alla clinica
ADAM10/ SAP97/AP2 interaction in AD patients

Marcello, Epis et al., Neurobiology of Aging, 2012

Marcello et al, submitted
Verifying the impact of key elements of the amyloid cascade ex vivo
I marcatori biologici per la demenza e il loro ruolo

Pubmed search: Biomarkers and Alzheimer

# of publications

<table>
<thead>
<tr>
<th>Years</th>
<th># of Publications</th>
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<td>2001-2003</td>
<td>600</td>
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<td>2004-2006</td>
<td>800</td>
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<td>2007-2009</td>
<td>1400</td>
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A biomarker is defined as a biological signature objectively measured and evaluated as an indicator of a normal biological process, a pathogenic process, or a pharmacological response to a therapeutic intervention.
What’s a biomarker

Indicator of normal and abnormal biological processes:

✓ precise and reliable (accurate >80%)
✓ distinguishable between normal and interested disease
✓ differential between different diseases

Detecting a fundamental feature of neuropathology

1998: Working group on Biomarkers, Consensus report
What’s a biomarker

✓ Genetic
✓ Neuroimaging
✓ Biochemical/neurochemical
I marcatori biologici per la demenza: Why?

✓ Preclinical/early diagnosis
  ✓ Marker diagnostici
  ✓ Marker antecedenti

✓ Clinical trials:

...Must have a scientific rationale, should change with the disease progression and must be measurable and reproducible
Should be used to:
Identify appropriate dosage
Improve safety assessment
Demonstrate pharmacological activity
Identify preliminary evidence of efficacy
  ✓ Marker prognostici

Thal et al, Alz.Dis. Assoc. Disord, 2006
I marcatori biologici per la demenza: **Which?**

“Should reflect a neuropathologic characteristic of AD”

*Hampel et al., Alzheimer & Dementia, 2008*

---

McNaull et al., Gerontology 2010
I marcatori biologici per la demenza

“Should reflect a neuropathologic characteristic of AD”

Hampel et al., Alzheimer & Dementia, 2008

Querfurth H W, La Ferla FM, NEJM, 2010
Designing biomarkers on neuropathological characteristics of AD
Perrin et al., 2009
CSF markers: A\(\beta\) and \(\tau\)

<table>
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<tr>
<th></th>
<th>N</th>
<th>T-tau (ng/L)</th>
<th>P-tau(_{181}) (ng/L)</th>
<th>A(\beta)42 (ng/L)</th>
<th>A(\beta)42/P-tau(_{181}) ratio</th>
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<tr>
<td>Controls</td>
<td>39</td>
<td>326 (157)</td>
<td>61 (17)</td>
<td>700 (181)</td>
<td>12.5 (4.7)</td>
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<td>Stable MCI</td>
<td>56</td>
<td>340 (212)</td>
<td>62 (16)</td>
<td>551 (188)*</td>
<td>9.5 (3.8)*</td>
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<tr>
<td>MCI-AD</td>
<td>57</td>
<td>816 (426)*†</td>
<td>95 (29)*†</td>
<td>324 (101)*†</td>
<td>3.7 (1.6)*†</td>
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<tr>
<td>MCI-Other</td>
<td>21</td>
<td>480 (516)‡</td>
<td>60 (26)‡</td>
<td>579 (155)‡</td>
<td>10.7 (3.9)‡</td>
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<tr>
<td>MCI-VaD</td>
<td>15</td>
<td>476 (592)‡</td>
<td>60 (30)‡</td>
<td>567 (173)‡</td>
<td>10.8 (4.4)‡</td>
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<tr>
<td>MCI-DLB</td>
<td>3</td>
<td>587 (184)</td>
<td></td>
<td>64 (11)**</td>
<td>572 (121)††</td>
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<tr>
<td>MCI-FTD</td>
<td>1</td>
<td>300</td>
<td>51</td>
<td>600</td>
<td>11.8</td>
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<tr>
<td>MCI-SD</td>
<td>1</td>
<td>828</td>
<td>81</td>
<td>760</td>
<td>9.4</td>
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<tr>
<td>MCI-TBI</td>
<td>1</td>
<td>58</td>
<td>42</td>
<td>579</td>
<td>13.8</td>
</tr>
</tbody>
</table>

Research criteria for the diagnosis of Alzheimer’s disease: revising the NINCDS–ADRDA criteria


The NINCDS–ADRDA and the DSM-IV-TR criteria for Alzheimer’s disease (AD) are the prevailing diagnostic standards in research; however, they have now fallen behind the unprecedented growth of scientific knowledge. Distinctive and reliable biomarkers of AD are now available through structural MRI, molecular neuroimaging with PET, and cerebrospinal fluid analyses. This progress provides the impetus for our proposal of revised diagnostic criteria for AD. Our framework was developed to capture both the earliest stages, before full-blown dementia, as well as the full spectrum of the illness. These new criteria are centred on a clinical core of early and significant episodic memory impairment. They stipulate that there must also be at least one or more abnormal biomarkers among structural neuroimaging with MRI, molecular neuroimaging with PET, and cerebrospinal fluid analysis of amyloid β or tau proteins. The timeliness of these criteria is highlighted by the many drugs in development that are directed at changing pathogenesis, particularly at the production and clearance of amyloid β as well as at the hyperphosphorylation state of tau. Validation studies in existing and prospective cohorts are needed to advance these criteria and optimise their sensitivity, specificity, and accuracy.

Position Paper
Platelet biomarkers

Di Luca et al., Arch Neurol 1998

Padovani et al., Neurology 2001
Platelet APPr in MCI subjects

Borroni et al., Arch Neurol. 2002
APPr scores according to the follow-up diagnosis

Borroni et al., Arch of Neurology 2002
Amyloid cascade in platelets of AD patients

Colciaghi et al., Neurology, 2004
Beyond Diagnosis: What Biomarkers Are Teaching Us about the "Bio"logy of Alzheimer Disease

Rabinovici et al, Ann Neurol, 2010
I marcatori biologici per la demenza

The sensitivity of the ideal biomarker to detect AD should be at least 85%

Its specificity to differentiate AD patients from controls of the same age and from patients with other forms of dementia should be at least 75%

Blennow et al., 2008
Combining biological markers: 

towards preclinical diagnosis of AD

<table>
<thead>
<tr>
<th>Markers</th>
<th>Authors</th>
<th>Marker Combination</th>
<th>Accuracy</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>An amyloid cascade</em></td>
<td>Di Luca M, 2005</td>
<td>Platelet APP forms, ADAM10, BACE</td>
<td>ACC 97.0%</td>
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<tr>
<td><em>Plasma Abeta</em></td>
<td>Breteler M, 2006</td>
<td>Plasma Abeta 40 and Plasma Abeta 42</td>
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<tr>
<td><em>CSF</em></td>
<td>Andreasen N, 1999</td>
<td>CSF Abeta 42 and CSF Tau</td>
<td>SEN 88%</td>
</tr>
<tr>
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<td>Riemenschneider M, 2002</td>
<td>CSF Abeta 42 and CSF Tau</td>
<td>SEN 90.0%, SPE 90.0%</td>
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<td>Hansson O, 2006</td>
<td>CSF Abeta 42 and CSF Tau</td>
<td>SEN 95.0%, SPE 83.0%</td>
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<td>Parnetti L, 2006</td>
<td>CSF Abeta 42, CSF Tau, and CSF phosho Tau</td>
<td>SEN 90% SPE 88%</td>
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<tr>
<td><em>NPS and neuroimaging</em></td>
<td>De Leon MJ, 2006</td>
<td>CSF phosphor Tau and CSF isoprostane</td>
<td>ACC 90.0%</td>
</tr>
<tr>
<td></td>
<td>Huang C, 2003</td>
<td>NPS (Mini-Mental State Examination, Block Design, Word Recognition test) and SPECT hypoperfusion</td>
<td>ACC 84.0%</td>
</tr>
<tr>
<td></td>
<td>Borroni B, 2006</td>
<td>NPS (Short Story, Rey Figure Recall, Rey List Delayed Recall) and SPECT hypoperfusion</td>
<td>SEN 77.8%, SPE 77.8%</td>
</tr>
<tr>
<td></td>
<td>Anchisi D, 2005</td>
<td>NPS (California Verbal Learning Test) and PET</td>
<td>SEN 85.7%, SPE 97.1%</td>
</tr>
<tr>
<td><em>Biomarkers and neuroimaging</em></td>
<td>Okamura N, 2002</td>
<td>CSF Tau and SPECT hypoperfusion</td>
<td>SEN 88.5%, SPE 90.0%</td>
</tr>
<tr>
<td></td>
<td>Borroni B, 2005</td>
<td>APP forms and SPECT hypoperfusion</td>
<td>SEN 95.2%, SPE 75.0%</td>
</tr>
<tr>
<td></td>
<td>Bouwman FH, 2006</td>
<td>CSF Abeta 42, CSF Tau, MRI (medial temporal atrophy)</td>
<td>SEN 94.0%, SPE 70.0%</td>
</tr>
</tbody>
</table>

Borroni et al., CMC 2007
I marcatori biologici per la demenza

“Should reflect a neuropathologic characteristic of AD”
Hampel et al., Alzheimer & Dementia, 2008

Molecular pathogenesis of neurodegenerative diseases, such as Alzheimer’s disease (AD), is the result of a complex interplay of several crossing pathways, involving primary and secondary events

Bossy-Wetzel et al., 2004; Marcello et al, 2007
La complessità della patogenesi implica che debba esserci più di un biomarker?
Oxidative stress and mitochondrial failure in AD

Figure 4. Oxidative Stress and Mitochondrial Failure.
Oxidative stress markers

Pratico’ et al, 2002

Isoprostanes

Thal L.J., 2006

Pratico’ et al, 2002
Inflammation in AD

Querfurth H W, La Ferla FM, NEJM, 2010
**Table 1. Summary of concentrations of cyto-/chemokines in the blood and CSF of AD patients**

<table>
<thead>
<tr>
<th>Marker</th>
<th>CSF level</th>
<th>Blood level</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Cytokine</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL-1</td>
<td>no difference [8, 9]</td>
<td>no difference [8–12]</td>
</tr>
<tr>
<td></td>
<td>increased [10–12, 38]</td>
<td>increased [13]</td>
</tr>
<tr>
<td>IL-4</td>
<td>no difference</td>
<td>no difference [16, 17]</td>
</tr>
<tr>
<td>IL-6</td>
<td>[20, 22, 25, 35, 38]</td>
<td>increased [10, 24, 25]</td>
</tr>
<tr>
<td></td>
<td>increased [8, 19]</td>
<td>increased [12, 13, 19, 21–23]</td>
</tr>
<tr>
<td>IL-10</td>
<td>no difference [30]</td>
<td>no difference [28–30]</td>
</tr>
<tr>
<td>IL-12</td>
<td>no difference [30]</td>
<td>increased [31, 32]</td>
</tr>
<tr>
<td>IL-16</td>
<td></td>
<td>increased [36]</td>
</tr>
<tr>
<td>IL-18</td>
<td></td>
<td>increased [36]</td>
</tr>
<tr>
<td>TNF</td>
<td>no difference [10, 38]</td>
<td>decreased [39, 40]</td>
</tr>
<tr>
<td></td>
<td>increased [12]</td>
<td>no difference [10, 12, 14, 25]</td>
</tr>
<tr>
<td>TGF-β</td>
<td>increased [30]</td>
<td>increased [37, 41–43]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>increased [36]</td>
</tr>
<tr>
<td><strong>Chemokine</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MCP</td>
<td>no difference [19, 46]</td>
<td>increased [19, 44, 45]</td>
</tr>
<tr>
<td>IL-8</td>
<td>no difference [27]</td>
<td>increased [44]</td>
</tr>
<tr>
<td>MIF</td>
<td></td>
<td>increased [47]</td>
</tr>
<tr>
<td>MIG</td>
<td></td>
<td>increased [47]</td>
</tr>
<tr>
<td>Fractalkine</td>
<td></td>
<td>increased [48]</td>
</tr>
</tbody>
</table>

Lee et al., *Dement Geriatr Cogn Disord* 2009;28:281–287
Biomarkers, both chemical and imaging, are indicators of specific changes that characterize AD in vivo. Evidence suggests that these AD biomarkers do not reach abnormal levels or peak simultaneously but do so in an ordered manner. Measurement of these biomarkers in longitudinal observational studies is now commonplace, enabling investigators to establish the correct ordering of the relevant biomarkers and their relationships to clinical symptoms.

Perrin et al., Nature, 2009
Figure 1 | Biomarkers and Alzheimer's disease: proposed changes in biomarkers in relation to the time course of pathological and clinical stages.
Hypothetical model of dynamic biomarkers of AD pathology

New biomarkers that will allow us to answer the following key questions about treating AD patients.

Who should we treat?
Who actually has the pathologic process of AD
What should we treat them with?
Because etiopathogenesis of AD is heterogeneous it is important to evaluate what treatment is most effective for different sub-groups of patients
When in the course of the disease should we treat them?
Different treatments can be more effective at specific disease stages,
How well is the treatment working?
A biomarker is defined as a biological signature objectively measured and evaluated as an indicator of a normal biological process, a pathogenic process, or a pharmacological response to a therapeutic intervention, and

*That adds value to treatment development*
SCIENTIFIC COMMENTARY

Biomarkers for Alzheimer’s disease: ready for the next step
New technological approaches for CSF markers

Measurement of thirteen biological markers in CSF of patients with Alzheimer's disease and other dementias.

- Abeta/Ptau
- 5 Growth Factors: HGF, GDNF, VEGF, BDNF, FGF-2
- Cytokines/chemokynes: TNF-alpha, TGF-beta1, MIP-1alpha

The results the suitability of the ratio P-tau181/Abeta42 for the diagnosis of AD, while CSF levels of NGF and MCP-1 are less specific and reliable for AD.

Blasko I, et al., Dement Geriatr Cogn Disord. 2006
Expression patterns of Alzheimer disease (AD) signature proteins discriminate between plasma samples from patients with AD and controls

Proteomic mutiplex method:
120 chemokynes, cytochines, growth factors signaling proteins in plasma of 40 controls vs 40 AD patients

The amyloid cascade: central in AD pathogenesis

Alzheimer Disease is a synaptopathy;

How basic research can help patients’ management: biomarkers and new targets