Gli altri strumenti diagnostici.
CSF e genetica: indicazioni e limiti

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I biomarkers liquorali
AD Progression

Abnormal

Normal

Pre-symptomatic  EMCI  LMCI  Dementia

Time

CSF abeta42  MRI hippocampal volume  Function (ADL)
Amyloid imaging  CSF Tau
FDG PET  Cognitive performance

### Applications of CSF biomarkers in AD

<table>
<thead>
<tr>
<th>Application</th>
<th>Details</th>
<th>Time point for use</th>
<th>Possible biomarker and their role</th>
</tr>
</thead>
<tbody>
<tr>
<td>Improving the accuracy of diagnosis</td>
<td>CSF biomarkers could be used in clinical trials to improve diagnostic accuracy in trial participants, enabling patient cohorts to be enriched with cases of AD</td>
<td>Before trial initiation</td>
<td>High T-tau and P-tau and low $A\beta_{1-42}$ are indicative of AD</td>
</tr>
<tr>
<td>Stratification of AD cases</td>
<td>AD cases with CSF biomarker evidence of a disturbance in $A\beta$ metabolism might be more responsive to anti-$A\beta$ drugs than patients who do not exhibit such a disturbance</td>
<td>Post hoc analysis</td>
<td>$A\beta_{1-42}$ might be used to stratify cases in trials of anti-$A\beta$ disease-modifying drug candidates; $\tau$-tau might be used to stratify cases in trials of drugs that aim to reduce tau phosphorylation and neurofibrillary tangle pathology</td>
</tr>
<tr>
<td>Safety monitoring</td>
<td>Anti-$A\beta$ drug candidates, such as $A\beta$ immunotherapy, might elicit adverse effects, such as meningoencephalitis or vasogenic oedema</td>
<td>Baseline evaluation and assessment during trial</td>
<td>CSF cell count, IgG or IgM index and IgG or IgM oligoclonal bands are standard measures for identifying and monitoring inflammatory processes, such as meningoencephalitis, in the CNS; the CSF:serum albumin ratio is the standard measure to identify and monitor a disturbance in the blood–brain barrier, which can lead to cerebral edema</td>
</tr>
<tr>
<td>Theragnostics</td>
<td>CSF biomarkers might indicate whether a drug has an effect on the molecular pathology of AD in living patients</td>
<td>Baseline evaluation and at time points throughout the trial, including the last week of the study</td>
<td>$A\beta_{1-42}$ is the main biomarker for $A\beta$ metabolism and deposition; APP isoforms ($sA\beta_{1-42}$ and $sA\beta_{1-40}$) and BACE1 activity might be valuable in clinical trials of BACE1 inhibitors; $\tau$-tau is the main biomarker for monitoring the phosphorylation state of tau; $t$-tau might be a valuable biomarker for identifying and monitoring a downstream effect on the intensity of neuronal or axonal degeneration</td>
</tr>
</tbody>
</table>

*Table 2* Applications of CSF biomarkers in AD clinical trials

Combined Aβ42 and τ measures in the diagnosis of Alzheimer Disease

Lines indicate optimized cutoffs. The high τ/low Aβ42 quadrant contains AD patients with only a single exception, whereas the low τ/high Aβ42 quadrant contains only control individuals.

Motter et al., 1995. Ann Neurol
Combination of Aβ and Tau CSF levels

Discrimination between AD and normal elderly: accuracy of about 90%

Normal
Aβ: >600 pg/ml
Tau: <300 pg/ml

AD
Aβ: ≈200 pg/ml
Tau: ≈800 pg/ml

Hulstaert et al., 1996, 1999

\[ \frac{A\beta_{42}}{[240 + (1.18 \times T-tau)]} < 1 \]
**Phosphotau (P-Tau) as biomarker for discriminating AD from FTD**

\[
P-tau_{181} \quad \text{sensitivity and specificity} \quad >85\%
\]

P-tau_{199} → low accuracy, not useful

Usefulness for differentiating between AD and other dementias, particularly FTD

_Hampel et al., 2004_
Aβ and Tau CSF levels in MCI

137 MCI subjects
39 healthy subjects

3-year follow-up

57 (42%) developed AD
21 (15%) developed other dementias
56 (41%) stable

Sensitivity = 95%
Specificity = 83%

to predict progression from MCI to AD

Hansson et al., 2006
53.7%: MCI-AD; 15.7%: MCI-OD

Baseline CSF Aβ42 levels equally reduced in early converters vs late converters

CSF T-tau and P-tau levels significantly higher in early converters vs late converters

Aβ42/Ptau ratio predicted the development of AD within 9.2 years with a sensitivity of 88%, specificity of 90%, positive predictive value of 91%, and negative predictive value of 86%
Biomarkers

limiti
Will CSF analysis become routine in people with memory complaints?

Serge Gauthier, Lancet Neurology 2009; 8: 595-6

A worldwide multicentre comparison of assays for cerebrospinal fluid biomarkers in Alzheimer’s disease

N A Verwey¹,², W M van der Flier², K Blennow³, C Clark⁴, S Sokolow⁵, P P De Deyn⁶, D Galasko⁷, H Hampe⁸,⁹, T Hartmann¹⁰, E Kapaki¹¹, L Lannfelt¹², P D Mehta¹³, L Parnetti¹⁴, A Petzold¹⁵, T Pirttila¹⁶, L Saleh¹⁷, A Skinningsrud¹⁸, J C v Swieten¹⁹, M M Verbeek²⁰, J Wiltfang²¹, S Younkin²², P Scheltens² and M A Blankenstein¹

Amyloid beta: interlab variations
Problema?
First Law of Socio-Genetics: Celibacy is not hereditary.

Anonymous
Sporadic AD
[prevalence: ~1/10 >65 yrs.]

EOAD AD
[prevalence: ~5/10^5]
Sporadic AD

Familial AD

EOAD AD

Sporadic AD
IL CERVELLO CHE CAMBIA 2

10 Novembre 2012
Genova, Palazzo Ducale - Sala Minor Consiglio

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Items 1 - 20 of 197

1. #104310
   ALZHEIMER DISEASE 2
   Gene map locus 19q13.2

2. #104300
   ALZHEIMER DISEASE; AD
   ALZHEIMER DISEASE, FAMILIAL, 1, INCLUDED; AD, INCLUDED
   Gene map locus 20p, 19p13.2, 17q23.1, 17q23, 17q11.2, 12q11.23-q13.12, 12p13.3-p12.3, 11q23.2-q24.2, 10q24, 10q24, 7q36, 7q36, 7q36, 7q36, 4p14

3. #606889
   ALZHEIMER DISEASE 4
   Gene map locus 1q31-q42

4. #607822
   ALZHEIMER DISEASE 3
   ALZHEIMER DISEASE, FAMILIAL, 3, WITH SPASTIC PARAPARESIS AND UNUSUAL PLAQUES, INCLUDED
   Gene map locus 14q24.3
APP locus duplication causes autosomal dominant early-onset Alzheimer disease with cerebral amyloid angiopathy

Anne Rovelet-Lecrux¹, Didier Hannequin¹², Gregory Raux¹, Nathalie Le Meur³, Annie Laquerrière⁴, Anne Vital⁵, Cécile Dumannin¹, Sébastien Feuillet⁵, Alexis Brice⁶, Martine Vercelletto⁷, Frédéric Dubas⁸, Thierry Frebourg¹ & Dominique Campion¹,⁹
### Autosomal dominant AD: summary

**Thirty years of Alzheimer’s disease genetics: the implications of systematic meta-analyses**

*Lars Bertram and Rudolph E. Tanzi*

<table>
<thead>
<tr>
<th>Gene (and protein)</th>
<th>Chromosomal location</th>
<th>Total number of pathogenic mutations (affected families)</th>
<th>Relevance to AD pathogenesis</th>
</tr>
</thead>
<tbody>
<tr>
<td>APP (amyloid precursor protein)</td>
<td>21q21.3</td>
<td>29 (78)</td>
<td>Increase in Aβ production or Aβ$<em>{42}$/Aβ$</em>{40}$ ratio; mutations in the Aβ sequence or close to the β- and γ-secretase site of APP; locus duplications</td>
</tr>
<tr>
<td>PSEN1 (presenilin 1)</td>
<td>14q24.3</td>
<td>166 (362)</td>
<td>Increase in Aβ$<em>{42}$/Aβ$</em>{40}$ ratio; mutations throughout molecule; enzymatic role in γ-secretase complex</td>
</tr>
<tr>
<td>PSEN2 (presenilin 2)</td>
<td>1q31–42</td>
<td>10 (18)</td>
<td>Increase in Aβ$<em>{42}$/Aβ$</em>{40}$ ratio; mutations throughout molecule; enzymatic role in γ-secretase complex</td>
</tr>
</tbody>
</table>
take-home messages

✓ Esistono forme genetiche, mendeliane di demenza
  ✓ geni, mutazioni, effetto biologico noti
  ✓ rare
  ✓ geneticamente eterogenee

✉ È possibile effettuare un test genetico per demenza
AD family?
take-home messages

✔ È possibile effettuare un test genetico per demenza,
  ✔ se clinicamente utile
    ✔ al paziente
    ✔ alla famiglia

✔ Esistono forme genetiche, mendeliane di demenza
  ✔ geni, mutazioni, effetto biologico noti
    ✔ rare
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• È indicato effettuare una consulenza genetica
Genetic counselling is a communication process which deals with the human problems associated with the occurrence, or risk of occurrence, of a genetic disorder in a family. This process involves an attempt by one or more appropriately trained persons to help the individual or the family to

- **comprehend the medical facts**, including the diagnosis, the probable course of the disorder and the available management;

- **appreciate the way heredity contributes to the disorder and the risk of recurrence** in specified relatives;

- **understand the options for dealing with the risk of recurrence**;

- **choose the course of action** which seems appropriate to them in view of their risk and their family goals and act in accordance with the decision;

- **make the best possible adjustment** to the disorder in an affected family member and/or to the risk of recurrence of that disorder.

Predictive values of genetic testing are function of:

**Prevalence**

- **Sensitivity** = true pos. / (true pos. + false neg.)
- **Specificity** = true neg / (true neg. + false pos.)

<table>
<thead>
<tr>
<th></th>
<th>Affecteds</th>
<th>Non-affecteds</th>
<th><strong>Total</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Test +</strong></td>
<td>true pos.</td>
<td>false pos.</td>
<td>true pos. + false pos.</td>
</tr>
<tr>
<td><strong>Test -</strong></td>
<td>false neg.</td>
<td>true neg.</td>
<td>false neg. + true neg.</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>true pos.+ false neg.</td>
<td>false pos.+ true neg.</td>
<td></td>
</tr>
</tbody>
</table>

**Positive predictive value** = true pos. / (true pos. + false pos.)

**Negative predictive value** = true neg. / (true neg. + false neg.)
for a given late-onset genetic disease:

Prevalence: low
Genetic heterogeneity ~ 0
Penetrance ~ 1

<table>
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<th>Non-affecteds</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Test +</td>
<td>~1</td>
<td>~0</td>
<td>1</td>
</tr>
<tr>
<td>Test -</td>
<td>~0</td>
<td>~1</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>1</td>
<td>1</td>
<td></td>
</tr>
</tbody>
</table>

Positive predictive value = true pos. / (true pos. + false pos.) ~ 1
Negative predictive value = true neg. / (true neg. + false neg.) ~ 1
... or for this late-onset genetic disease:

Prevalence: low  
Genetic heterogeneity > 0  
Penetrance < 1

<table>
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<th>Non-affecteds</th>
<th>Total</th>
</tr>
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<tbody>
<tr>
<td>Test +</td>
<td>8</td>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td>Test -</td>
<td>92</td>
<td>898</td>
<td>990</td>
</tr>
<tr>
<td>Total</td>
<td>100</td>
<td>900</td>
<td>1000</td>
</tr>
</tbody>
</table>

Positive predictive value = true pos. / (true pos. + false pos.) = $\frac{8}{10} = 80\%$

Negative predictive value = true neg. / (true neg. + false neg.) = $\frac{92}{990} \approx 9\%$
take-home messages

✔ Esistono forme genetiche, mendeliane di demenza
  ✔ geni, mutazioni, effetto biologico noti
  ✔ rare
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✔ È possibile effettuare un test genetico per demenza,
  ✔ se clinicamente utile
  ✔ tenendo presente i valori predittivi [bassi]

สร้างสรรÈ indicato effettuare una consulenza genetica(130,423),(630,597)
Online Mendelian Inheritance in Man
Apolipoprotein E

- 3 isoforms: E2, E3, E4
- Coded by 3 alleles: ε2, ε3, ε4
- Allele frequency: 0.08, 0.75, 0.17
  - Genotype frequency: ε3/ε4 ~ 0.50
  - ε4/ε4 ~ 0.15
  - OR (95% CI): ε3/ε4 = 3.2 (2.8–3.8)
    [Bertram et al 2007]
  - OR (95% CI): ε4/ε4 = 14.9 (10.8–20.6)
At present, APOE testing is used in a research setting to identify study participants who may have an increased risk of developing AD. This knowledge helps scientists look for early brain changes in participants and compare the effectiveness of treatments for people with different APOE profiles.

Most researchers believe that the APOE test is useful for studying AD risk in large groups of people but not for determining any one person’s specific risk. Someday, perhaps, screening in otherwise healthy people may be useful if an accurate and reliable test is developed and effective ways to treat or prevent AD become available.
**take-home messages**

- Esistono forme genetiche, mendeliane di demenza
  - geni, mutazioni, effetto biologico noti
  - rare
  - geneticamente eterogenee
- È possibile effettuare un test genetico per demenza,
  - se clinicamente utile
  - tenendo presente i valori predittivi [bassi]
- È frequente l’aggregazione familiare
- La componente genetica costituisce un fattore di rischio
  - variabile all’interno della popolazione
- La genotipizzazione di ApoE non è clinicamente utile
  - non va effettuata in ambito clinico
- È indicato effettuare una consulenza genetica
FTD

- Mutations in the progranulin gene (PGRN), on chromosome 17q21, have been identified as a major cause of familial frontotemporal dementia (FTD).
- These cases have a characteristic pattern of neuropathology that is a distinct subtype of frontotemporal lobar degeneration with ubiquitinated inclusions (FTLD-U).
- There is no abnormal accumulation of PGRN protein in the brain and immunohistochemical and biochemical analysis indicates that the ubiquitinated pathological protein is TDP-43.

*The neuropathology and clinical phenotype of FTD with progranulin mutations*  
Brain weighted 650 gr. Massive atrophy of the frontal temporal lobes with relative sparing of Rolandic and calcarine regions

Severe atrophy of the caudate nucleus and of white matter