CSF and Blood biomarkers for AD and MCI

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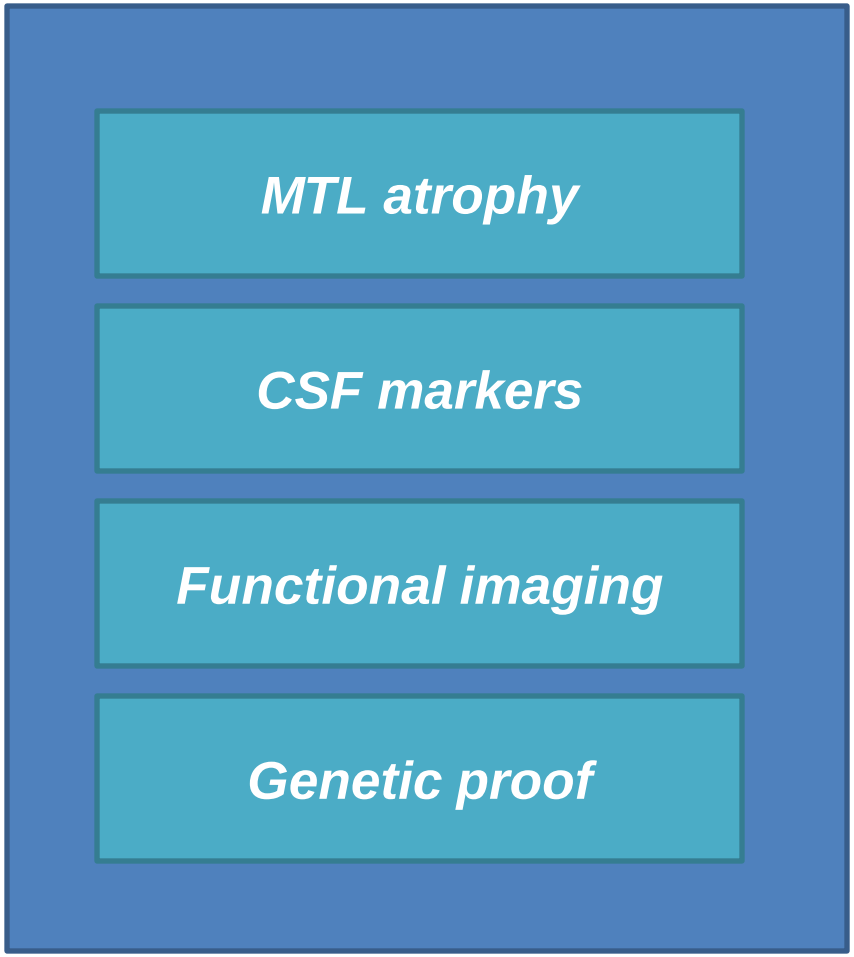
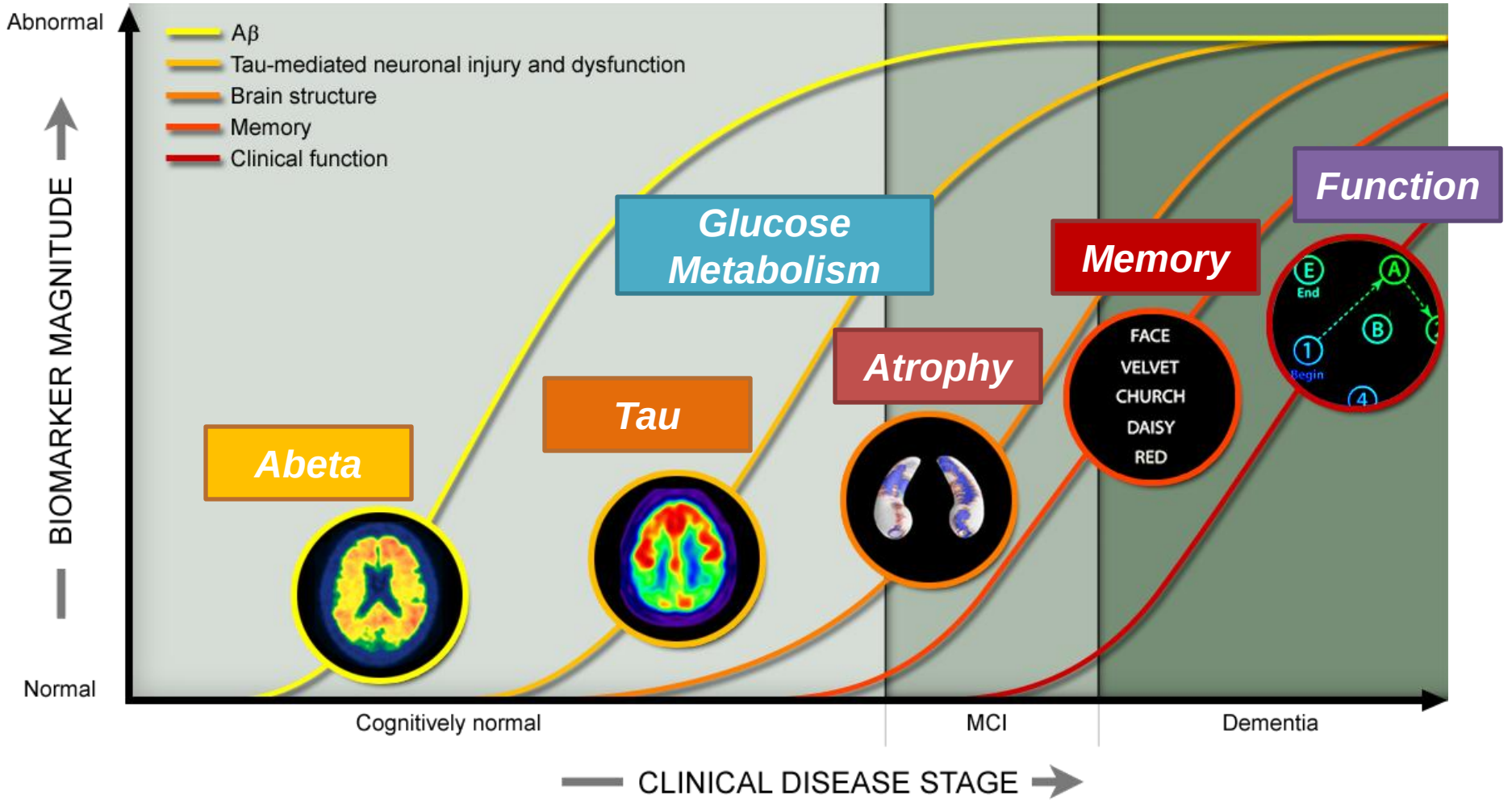
Biological markers for AD

- ***Imaging marker***
  - ***Structural /Functional***
- ***Molecular marker***
  - ***CSF/Blood/Urine/Genetic***
- ***Abeta-deposition marker***
  - ***CSF Abeta / PIB-PET***
- ***Neurodegeneration marker***
  - ***CSF tau/ FDG-PET/ st-MRI***



cf. Behavioral markers for AD

Biomarkers are pathogenesis-specific & temporally ordered



Biomarker의 중요성

- CSF marker의 중요성
  - Brain의 direct interface
  - AD pathology의 hallmarks를 측정
    - Abeta metabolism or deposition
    - Tau metabolism and phosphorylation
    - 질환의 temporal status반영할 가능성
- Peripheral marker의 중요성
  - Sampling이 용이
  - 임상 적용 가능성; 실현성은 낮고, 가용성은 높음
- 왜 필요한가?
  - AD 진단 자체를 명확히
    - 정상노화와의 감별
    - 다른 치매와의 감별
  - 초기 환자군 식별 (preclinical AD)
    - 치료 혹은 임상시험 대상자 선별
    - Disease-modifying effect 적용 혹은 증명

누가 더 '치매'라고 생각되는가?

CASE 1

- *"2년 전부터 영~ 기억이 안좋으시다"*
- ***CSF Abeta = 172.3 ng/L***
- ***CSF T-tau = 620.4 ng/L***

CASE 2

- *"2년 전부터 영~ 기억이 안좋으시다"*
- ***MMSE = 16***
- ***CDR = 1.0***

질문1: AD 진단에 도움이 되는가?

- 치매 여부를 판단하는 것 (?)
  - **clinical vs. biological entity**

- 다른 치매가 아니라 **AD dementia**라는 것을 진단하는 것 (?)

  - **Probable AD dementia with/without bio-evidence**
  - **Possible AD dementia with/without bio-evidence**
  - **Dementia-unlikely AD dementia with/without bio-evidence for AD**

  → **bio-evidence** 의 수준에 따라 **clinical diagnosis**의 신뢰성이 높아지는 구조 **(NIA-AA)**

- *그동안 **AD** 환자에 대한 **CSF marker** 분석의 **sensitivity** 와 **specificity** 는 약 **75-85%** 정도로 상당히 유용할 것처럼 보인다.*
  - ***Abeta 78-100% sensitivity, 47-81% specificity***
  - ***T-tau 70%, 92%***
  - ***P-tau 77%, 87%***
  - ***Abeta/tau ratio 71%, 83% (Hampel, 2004)***

- *다른 치매와의 감별에서는 이러한 유용성이 떨어진다.*
  - ***LBD;***
    - ***Abeta37,38,40 slightly higher in AD → Abeta42/Abeta37 ?***
    - ***alpha-synuclein, conflicting results***
  - ***FTD;***
    - ***Abeta42,** 정상보다 낮고, **AD**보다는 높다*
    - ***Tau,** 정상보다 높고, **AD**보다는 낮다 → **T(or P)-tau/Abeta42** 유용*
  - ***CJD***
    - ***Abeta42** 낮고, **T-tau**는 매우 높고, **P-tau**는 정상 수준*

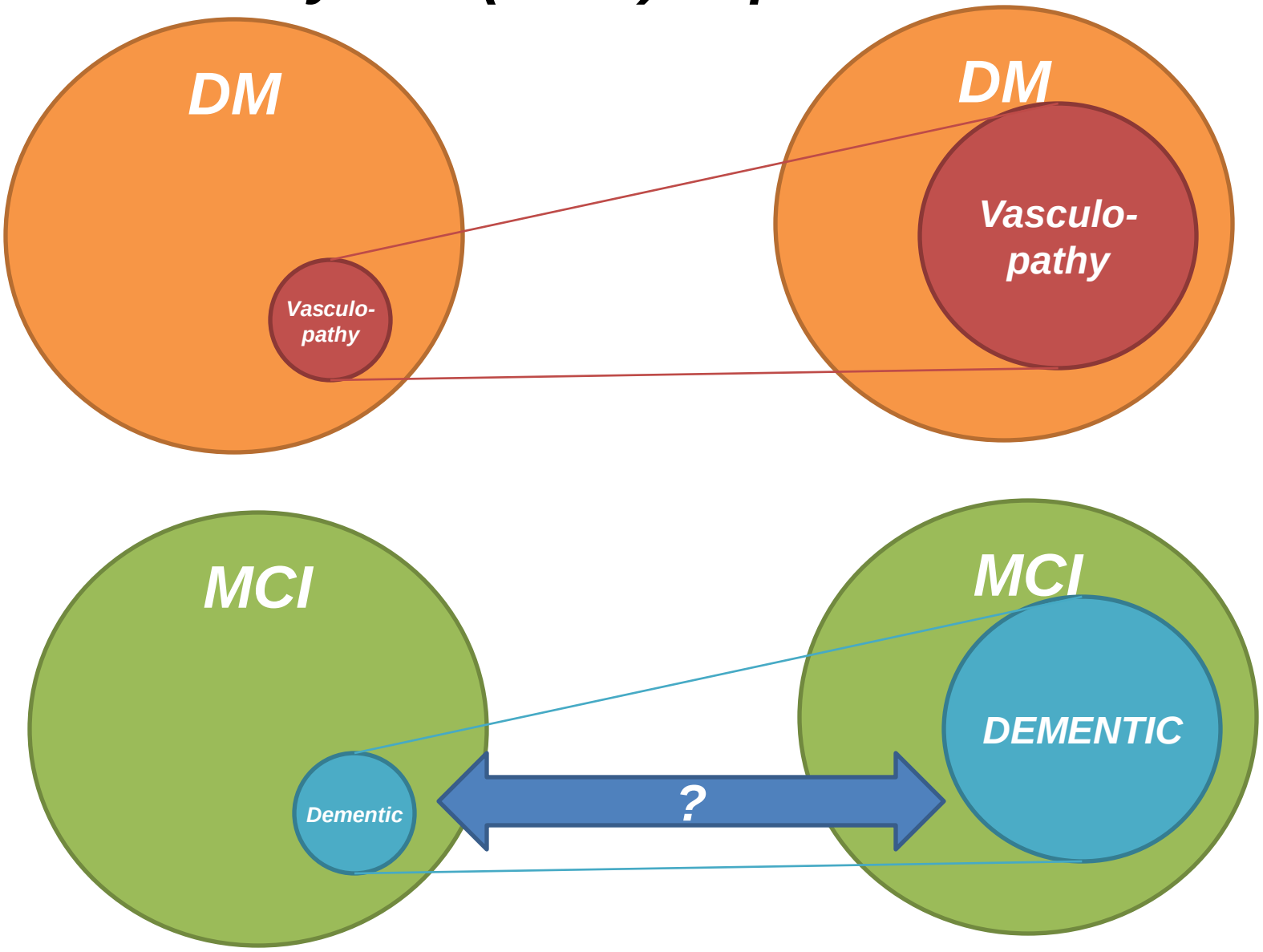
Differential Diagnosis among cognitive disorders with CSF biomarkers

인지장애	Aβ42	T-tau	P-tau	CSF AD profile	참고
AD	↓	↑	↑	↓	Aβ42/tau
MCI	↓	↑	↑	↓	
Normal Aging	↔	↔	↔	↔	
Depression	↔	↔	↔	↔	
FTD	↔ 또는 ↓	↔ 또는 ↑	↔	↓ 또는 ↑	tau/Aβ42
LBD	↔ 또는 ↓	↔ 또는 ↑	↔ 또는 ↑	↔ 또는 ↓	Aβ42/Aβ38(37)
VaD	↔ 또는 ↓	↔ 또는 ↑	↔	↔ 또는 ↓	
CJD	↔ 또는 ↓	↑ ↑ ↑	↔ 또는 ↑	↓ ↓	T-tau/P-tau

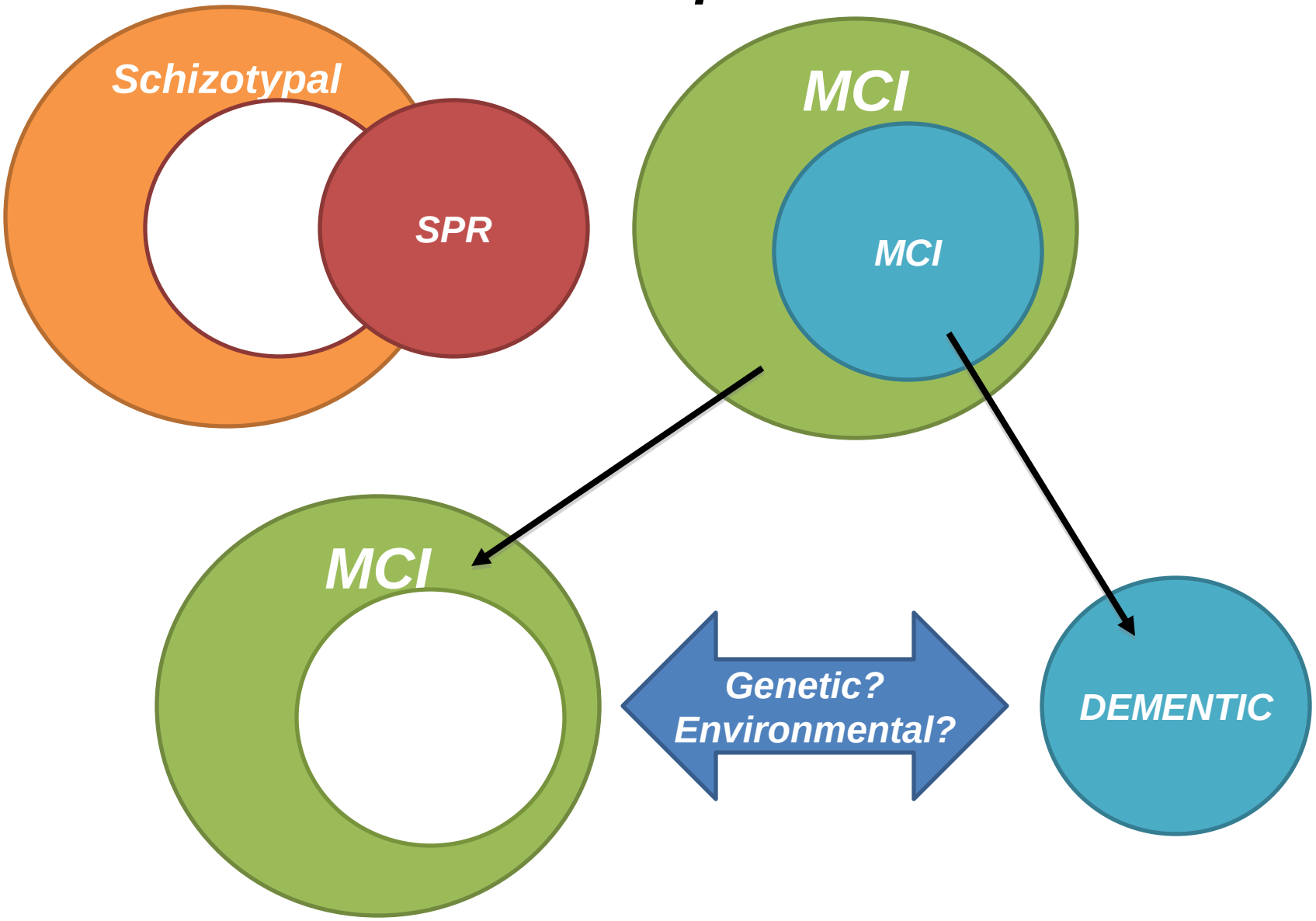
질문2: MCI 진단에 도움이 되는가?

- **MCI state를 진단하는 것 → clinical**
  - **Cognitive status in continuum from normal aging to dementia**
  - **Preclinical AD identification**
- **MCI trait를 진단하는 것 → bio-evidence**
  - **MCI due to AD vs. stable MCI 를 구분 (AD conversion prediction)**
  - **MCI due to AD vs. MCI due to other causes 를 구분 (MCI subtype)**

Conversion from MCI to AD is only time(state)-dependent?



Conversion from MCI to AD is also trait-dependent?



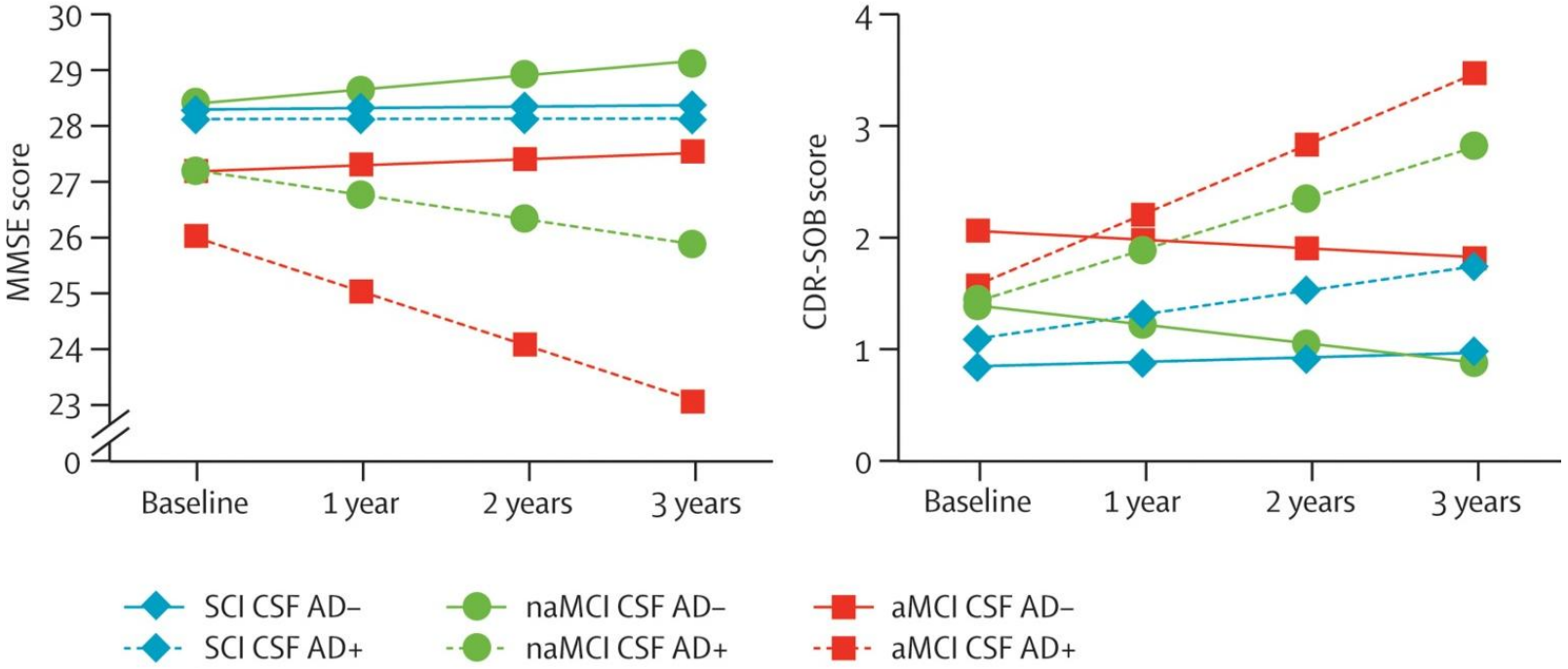
CSF reflects cognitive status
(as a temporal marker?)

CSF AD profile = ratio $A\beta_{42}$:(240+[1·18*T-tau]) < 1

	Controls (n=89)	Subjective cognitive impairment (n=60)	Non-amnestic MCI (n=37)	Amnestic MCI (n=71)
Age (years)	67·1 (6·4)	66·0 (7·9)	70·0 (7·7)§,¶	70·0 (7·7)*¶
Years in education	..	11·8 (4·1)	10·7 (3·7)	10·4 (3·3)
Women	48 (54%)	29 (48%)	17 (46%)	34 (48%)
MMSE score	29·3 (0·9)	28·8 (1·2)	27·6 (2·2)‡	25·9 (2·8)‡**‡‡
CDR-SOB	..	0·7 (0·7)	1·3 (0·9)¶	1·7 (1·2)**
Depression	..	7 (13%)	2 (6%)	2 (3%)
Delayed recall (z score)	..	0·46 (0·94)	−0·49 (0·77)**	−1·97 (0·74)**§§
Carrier of APOE ε4	..	29 (53%)	11 (36%)	37 (53%)§§
$A\beta_{42}$ (pg/mL)¶¶	703 (194)	653 (268)	583 (272)	493 (254)‡‖‡‡
T-tau (pg/mL)¶¶	329 (133)	360 (200)	401 (278)	539 (375)‡††
P-tau (pg/mL)	53 (20)	62 (27)§	67 (33)	84 (54)‡
CSF AD profile	28 (31%)	31 (52%)†	25 (68%)†	56 (79%)‡‖

Visser et al. Lancet Neurol 2009; 8: 619–27

'CSF AD profile' also identifies 'stable MCI'
(as a trait marker?)



Visser et al. Lancet Neurol 2009; 8: 619–27

Table 3. Concentrations of Aβ42, Total Tau (T-Tau), and Phosphorylated Tau (P-Tau) in Cerebrospinal Fluid Obtained at Enrollment[a]

Group	No. of Patients	Aβ42, ng/L	T-Tau, ng/L	P-Tau, ng/L
Controls	304	675 (182-1897)	280 (42-915)	51 (16-156)
AD	529	370 (85-1354)[b]	559 (85-2782)[b]	82 (17-279)[b]
MCI				
All	750	467 (96-1420)[b,c]	380 (31-2483)[b,c]	61 (15-183)[b,c]
Stable	420	589 (121-1420)[b,c]	298 (31-1580)[c]	54 (15-163)[c]
Incipient AD	271	356 (96-1075)[b,d]	582 (83-2174)[b,d]	81 (15-183)[b,d]
All other MCI	59	487 (158-857)[b,c,e,f]	275 (40-2483)[c,e]	47 (22-163)[c,e]
Vascular dementia	28	512 (190-825)[b,c,e]	319 (86-2483)[c,e]	51 (24-163)[c,e]
Dementia with Lewy bodies	14	427 (199-654)[b,g,f]	329 (40-1010)[g,h]	55 (25-125)[g,h]
Frontotemporal dementia	7	600 (366-857)[g,h]	275 (237-347)[c,e]	45 (41-58)[c,h]
Other	10	585 (158-760)	149 (58-828)[c,e]	39 (22-81)[c,e]
Stable MCI plus all other MCI cases	479	579 (121-1420)[b,c,e]	294 (31-2483)[c,e]	53 (15-163)[c,e]

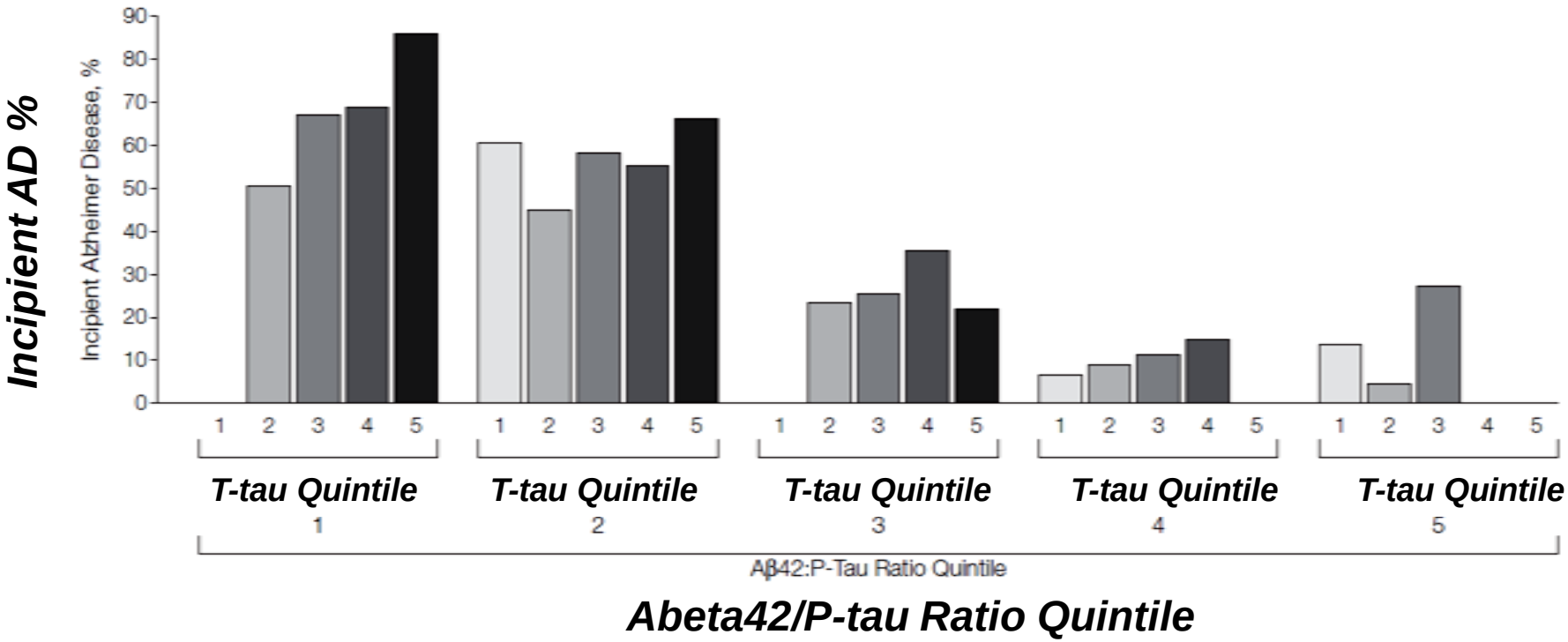
Abbreviations: AD, Alzheimer disease; MCI, mild cognitive impairment.
[a] Data presented as median (range), data from normalization model.
[b] $P<.001$ vs controls.
[c] $P<.001$ vs AD.
[d] $P<.001$ vs stable MCI.
[e] $P<.001$ vs incipient AD.
[f] $P<.01$ vs stable MCI.
[g] $P<.01$ vs AD.
[h] $P<.01$ vs incipient AD.

Mattsson et al. JAMA, 2009

CSF markers predict AD conversion

개별 site에서의 예측력은 상당히 높은 것으로 보고되었으나 (60-100%),
Multi-center study에서는 민감도와 특이도가 83%, 72%로 낮게 보고되었다.

Figure 1. Percentage of Patients With MCI Who Developed Alzheimer Disease by Quintiles of CSF T-Tau and CSF Aβ42/P-Tau Ratio

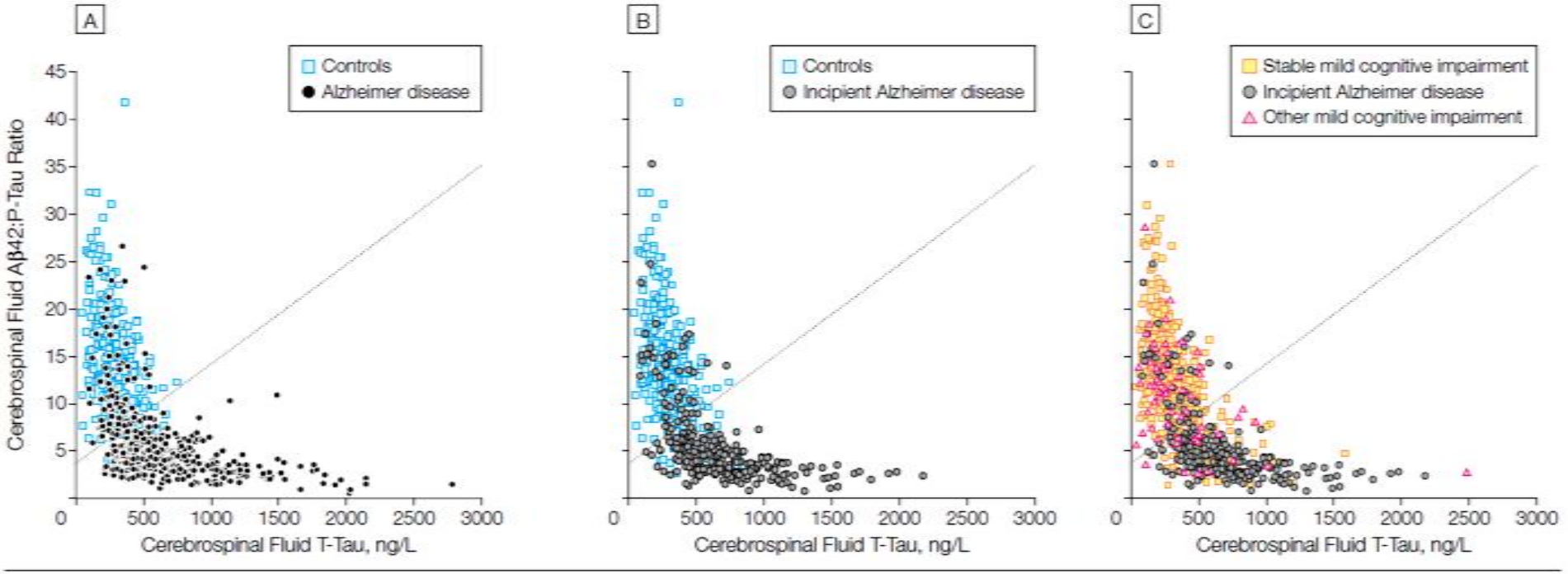


Incipient AD %

Abeta42/P-tau Ratio Quintile

Mattsson et al. JAMA, 2009

CSF markers may identify ‘stable’ MCI



$y = 3.6940 + 0105x$

Mattsson et al. JAMA, 2009

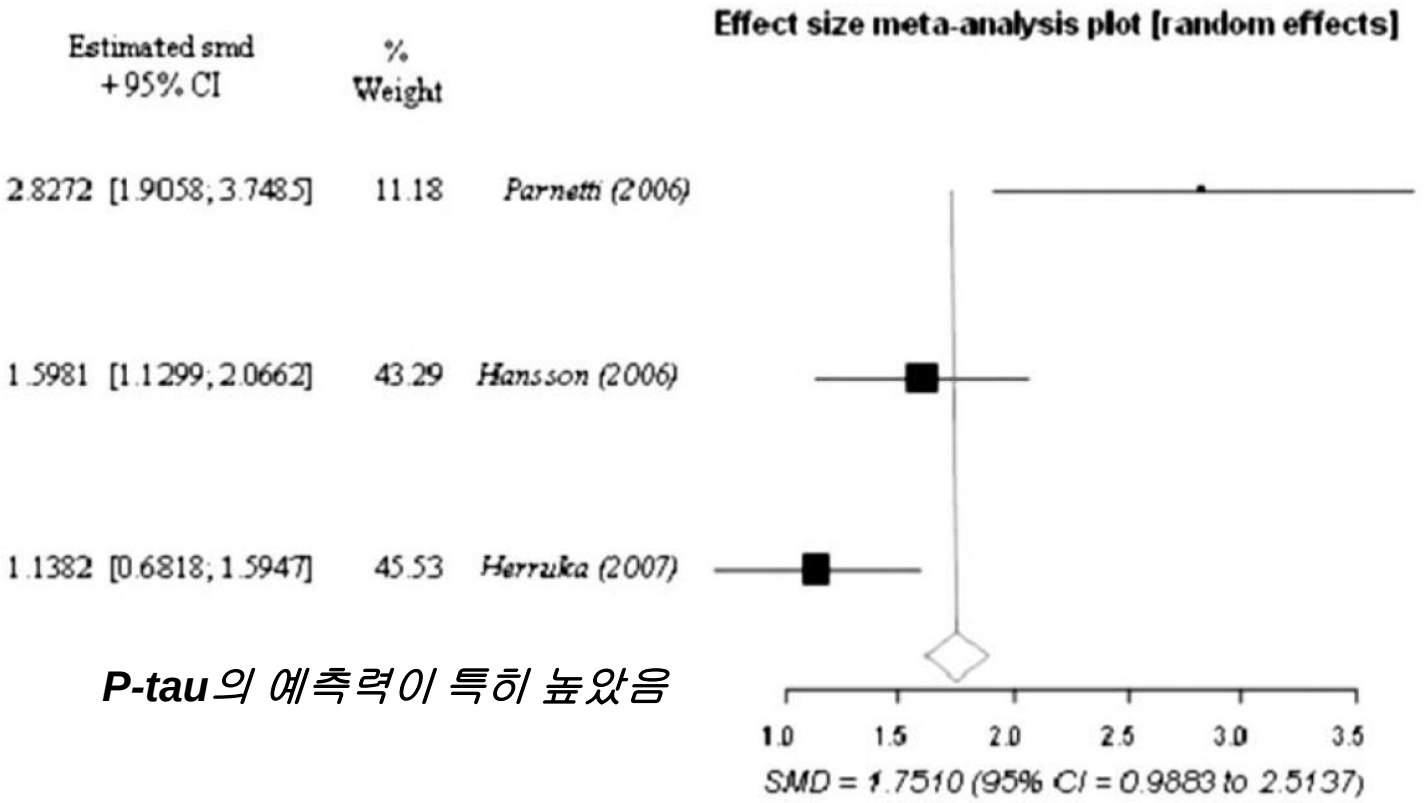
MCI criteria incorporating biomarkers

Albert et al. 2011

Diagnostic Category	Biomarker probability of AD etiology	Abeta (PET or CSF)	Neuronal injury (Tau, FDG, sMRI)
MCI Core clinical criteria	Uninformative	Conflicting/ indeterminant/ untested	Conflicting/ indeterminant/ untested
MCI due to AD -intermediate likelihood	Intermediate	Positive Untested	Untested Positive
MCI due to AD -high likelihood	Highest	Positive	Positive
MCI- unlikely due to AD	Lowest	Negative	Negative

메타분석 결과

- ***CSF marker*가 *AD converter* 를 판정하는데 분명히 유용**
- *오차가 커서 임상 실제적용에는 한계*



***P-tau*의 예측력이 특히 높았음**

Diniz et al. World J Bio Psych, 2008

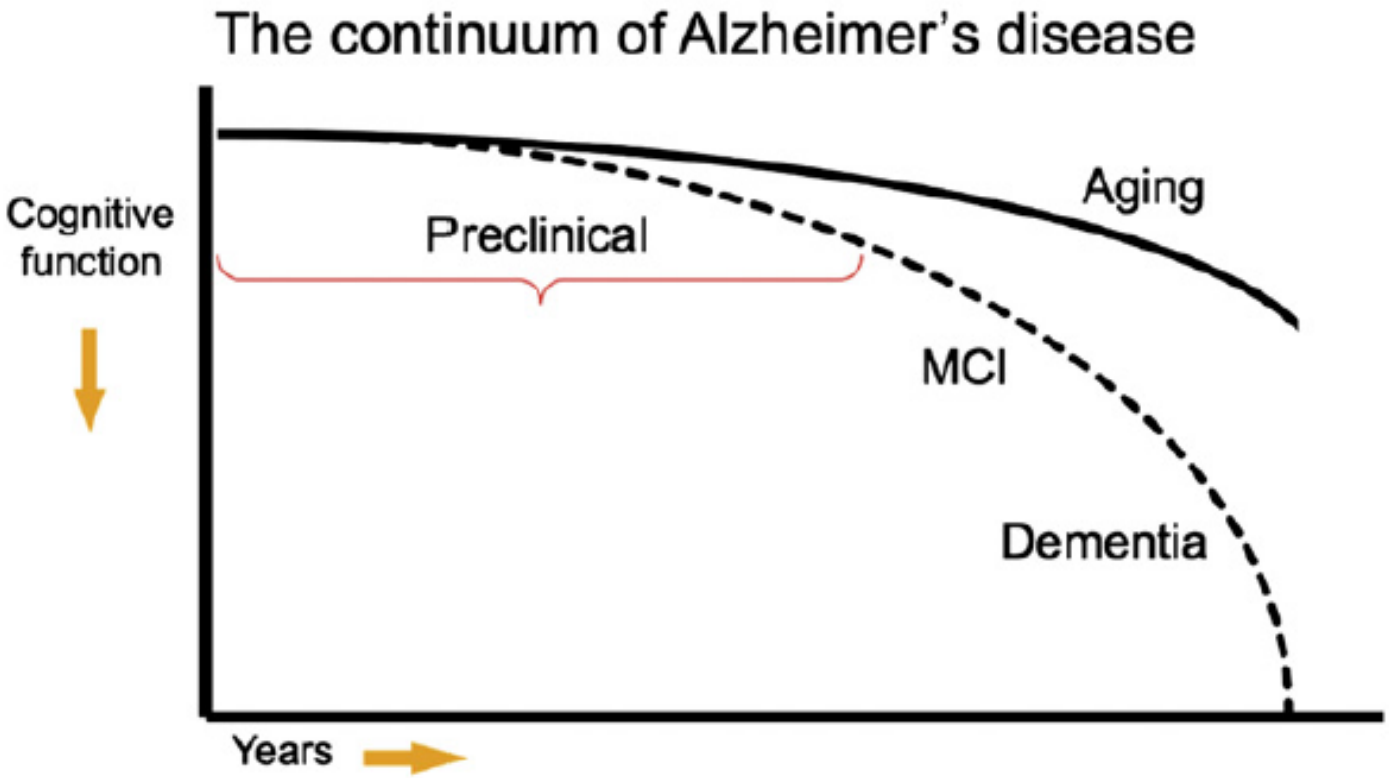
질문3: Pre-clinical AD 판명에 유용한가?

- *정상인에서도 향후* ***AD*** *발생을 예측하는 것에 유용했다는 보고들이 있음* ***(Vemuri, 2010, Gustafson, 2007, Stomrud, 2007)***

- *아직은 전향적 연구가 부족*

- *특히* ***data****의* ***variance*** *가 크기 때문에 개인별로 적용하는 것에는 한계가 큰 상태*

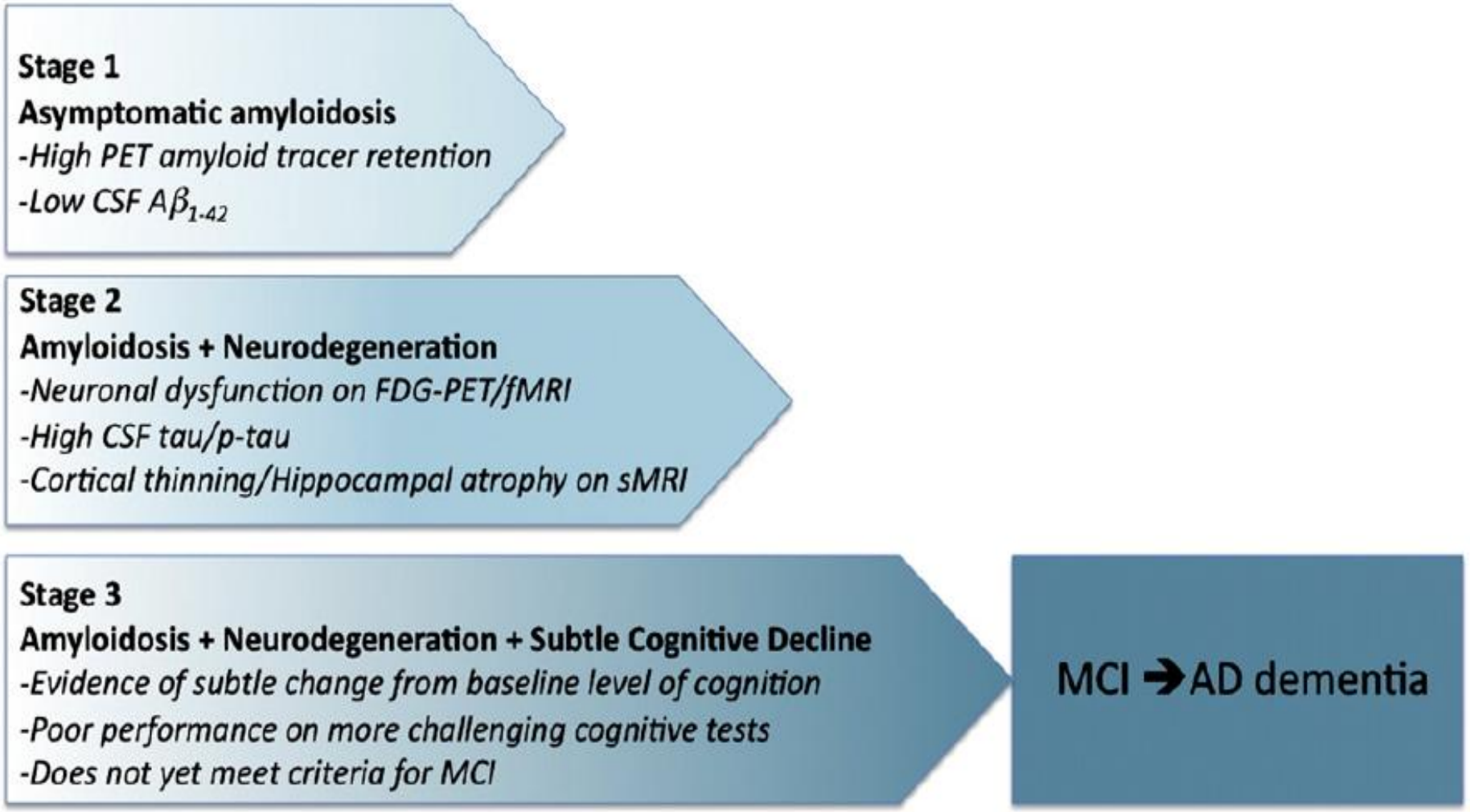
Concept of preclinical AD



Sperling et al, Alzheimer's & dementia, 2011 From NIA-AA

Proposed staging framework for preclinical AD

Sperling et al, Alzheimer's & dementia, 2011 From NIA-AA



질문4: 진행예측에 도움이 되는가?

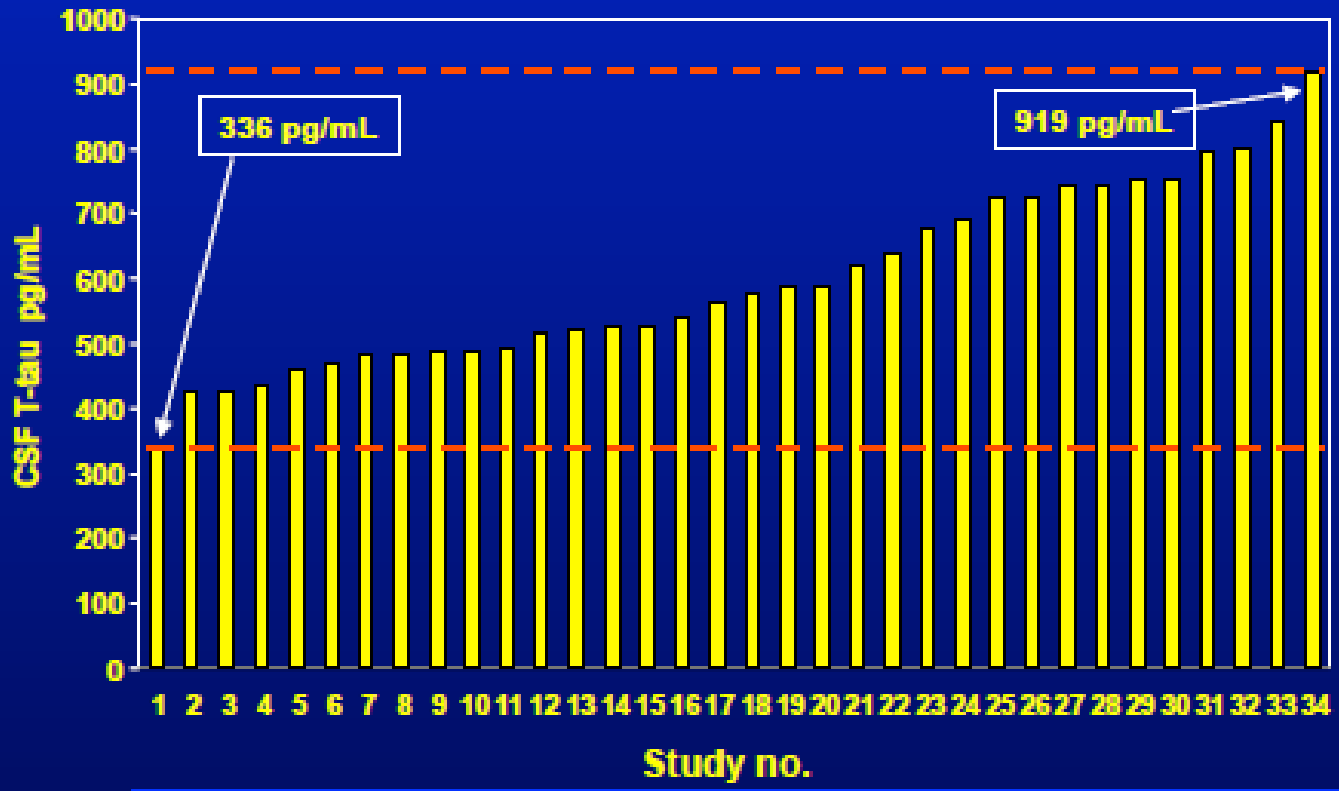
- **AD** 내에서 **disease progress**를 예측하기 위한 **sensitivity**는 낮은 것으로 보고되었음 **(Sunderland, 1999; Vemuri, 2010)**

- **MCI** 의 **progression (to AD)**는 상기 내용에서처럼 상당히 유용한 정보를 주는 것으로 보임

- 그러나 한계가 있다.
  - 다른 **marker**와의 **combination**에서 **power**적다는 것이 발표되고 있고,
  - 특히 개인별 레벨에서 적용 하기에는 **inter-individual, inter-center variation**이 큰 것이 한계점 **(**개인오차 **+** 실험오차**)**

Since there is significant variation in CSF biomarker levels between studies, there is an urgent need to standardize and validate AD biomarkers

Study design:	**All studies on CSF T-tau with >25 AD cases**
	Innogenetics T-tau ELISA
	34 studies, 2600 AD cases
Comparison of:	**mean level of CSF T-tau**



Blennow K, NYAS 2006

→ Need for standardization: CSF sampling / handling procedures

Laboratory procedures

External control program

Univariate results: Prediction of conversion

Baseline	Hazard ratio	p-value
ApoE ε4 allele	1.94	p = 0.10
FDG-PET imaging	2.94	p = 0.02
Hippocampal volume	2.49	p = 0.04
CSF markers ($p\text{-}tau_{181}$/Aß)	3.99	p = 0.03
Episodic memory	4.68	p = 0.01

→ Conversion to AD

Jagust, PET core http://www.adni-info.org

Multivariate results: Prediction of conversion

Baseline	Hazard ratio	p-value
FDG-PET imaging	2.95	p = 0.02
Episodic memory	5.08	p = 0.01

Conversion to AD

Jagust, PET core http://www.adni-info.org

ORIGINAL ARTICLE

Utility of Combinations of Biomarkers, Cognitive Markers, and Risk Factors to Predict Conversion From Mild Cognitive Impairment to Alzheimer Disease in Patients in the Alzheimer's Disease Neuroimaging Initiative

Jesus J. Gomar, PhD; Maria T. Bobes-Bascaran, MA; Concepcion Conejero-Goldberg, MD, PhD; Peter Davies, PhD; Terry E. Goldberg, PhD; for the Alzheimer's Disease Neuroimaging Initiative

Context: Biomarkers have become increasingly important in understanding neurodegenerative processes associated with Alzheimer disease. Markers include regional brain volumes, cerebrospinal fluid measures of pathological Aβ1-42 and total tau, cognitive measures, and individual risk factors.

Objective: To determine the discriminative utility of different classes of biomarkers and cognitive markers by examining their ability to predict a change in diagnostic status from mild cognitive impairment to Alzheimer disease.

Design: Longitudinal study.

Participants: We analyzed the Alzheimer's Disease Neuroimaging Initiative database to study patients with mild cognitive impairment who converted to Alzheimer disease (n=116) and those who did not convert (n=204) within a 2-year period. We determined the predictive utility of 25 variables from all classes of markers, biomarkers, and risk factors in a series of logistic regression models and effect size analyses.

Setting: The Alzheimer's Disease Neuroimaging Initiative public database.

Outcome Measures: Primary outcome measures were odds ratios, pseudo-R^2s, and effect sizes.

Results: In comprehensive stepwise logistic regression models that thus included variables from all classes of markers, the following baseline variables predicted conversion within a 2-year period: 2 measures of delayed verbal memory and middle temporal lobe cortical thickness. In an effect size analysis that examined rates of decline, change scores for biomarkers were modest for 2 years, but a change in an everyday functional activities measure (Functional Assessment Questionnaire) was considerably larger. Decline in scores on the Functional Assessment Questionnaire and Trail Making Test, part B, accounted for approximately 50% of the predictive variance in conversion from mild cognitive impairment to Alzheimer disease.

Conclusions: Cognitive markers at baseline were more robust predictors of conversion than most biomarkers. Longitudinal analyses suggested that conversion appeared to be driven less by changes in the neurobiologic trajectory of the disease than by a sharp decline in functional ability and, to a lesser extent, by declines in executive function.

Arch Gen Psychiatry. 2011;68(9):961-969

Gomar, et al. Arch Gen Psychiatry 2011

Table 5. Clustered Logistic Regression Models of Conversion During 2 Years

Variable	OR (95% CI)	ΔR^2	*P* Value
Demographic characteristics and *APOE* ($\chi^2 = 14.17/P < .001$; AUC = 0.61)			
APOE	2.51 (1.55-4.09)	.06	<.001
Cognitive markers ($\chi^2 = 106.15/P < .001$; AUC = 0.80)			
ADAS memory	1.07 (1.01-1.14)	.18	<.001
Logical Memory delay	1.01 (0.96-1.06)	.06	<.001
Clock Drawing test	0.95 (0.85-1.07)	.05	<.001
AVLT delay	0.80 (0.70-0.91)	.04	<.001
Trails A	0.99 (0.98-0.99)	.03	<.001
Brain volumetric measures ($\chi^2 = 50.15/P < .001$; $R^2 = 0.27$; AUC = 0.77)			
Left middle temporal lobe	0.02 (0.01-0.09)	.18	<.001
Left hippocampus	0.022 (0.006-0.087)	.09	<.001
CSF biomarkers ($\chi^2 = 12.36/P < .001$; AUC = 0.64)			
Tau/Aβ1-42 ratio	0.03 (0.03-0.22)	.11	<.001
"Winners" model, ie, including only previous significant measures ($\chi^2 = 29.45/P < .001$; AUC = 0.80)			
Logical Memory delayed total	0.80 (0.67-0.95)	.18	<.001
Left middle temporal lobe thickness	0.04 (0.01-0.27)	.10	<.001
AVLT delayed	0.77 (0.64-0.92)	.06	.02

Abbreviations: ADAS, Alzheimer Disease Assessment Scale; *APOE*, apolipoprotein; AUC, area under the curve; AVLT, Auditory Verbal Learning Test; CI, confidence interval; CSF, cerebrospinal fluid; OR, odds ratio.

Gomar, et al. Arch Gen Psychiatry 2011

CSF marker 결론

- Brain과 맞닿은 시료로 hallmark pathology를 측정한다
  - PET 보다 저렴하다
  - Disease-modifying drug 개발에 이용가능
  - MCI progress (conversion) 예측가능

- Limitations
  - Acceptability of patients
  - Inter- intra-regional & inter-individual variance
    - Need for standardization of sampling and measurement
  - 독립적 유용성 외에 다른 검사에 부가적으로 필요한가?
    - cutoff를 말하기 힘들다면 cognitive marker를 능가하는 유용성은 아직 회의적

Peripheral biomarkers

- ***AD pathology related***
  - ***Abeta***
  - ***Tau***
  - ***Oxidative stress***
  - ***Mitochondrial function (blood cell 이용)***
  - ***Inflammatory markers (cytokines)***

  → ***Hypothesis-proving***

- ***Explorative***
  - ***Application of omics tool***

  → ***Hypothesis-generating***

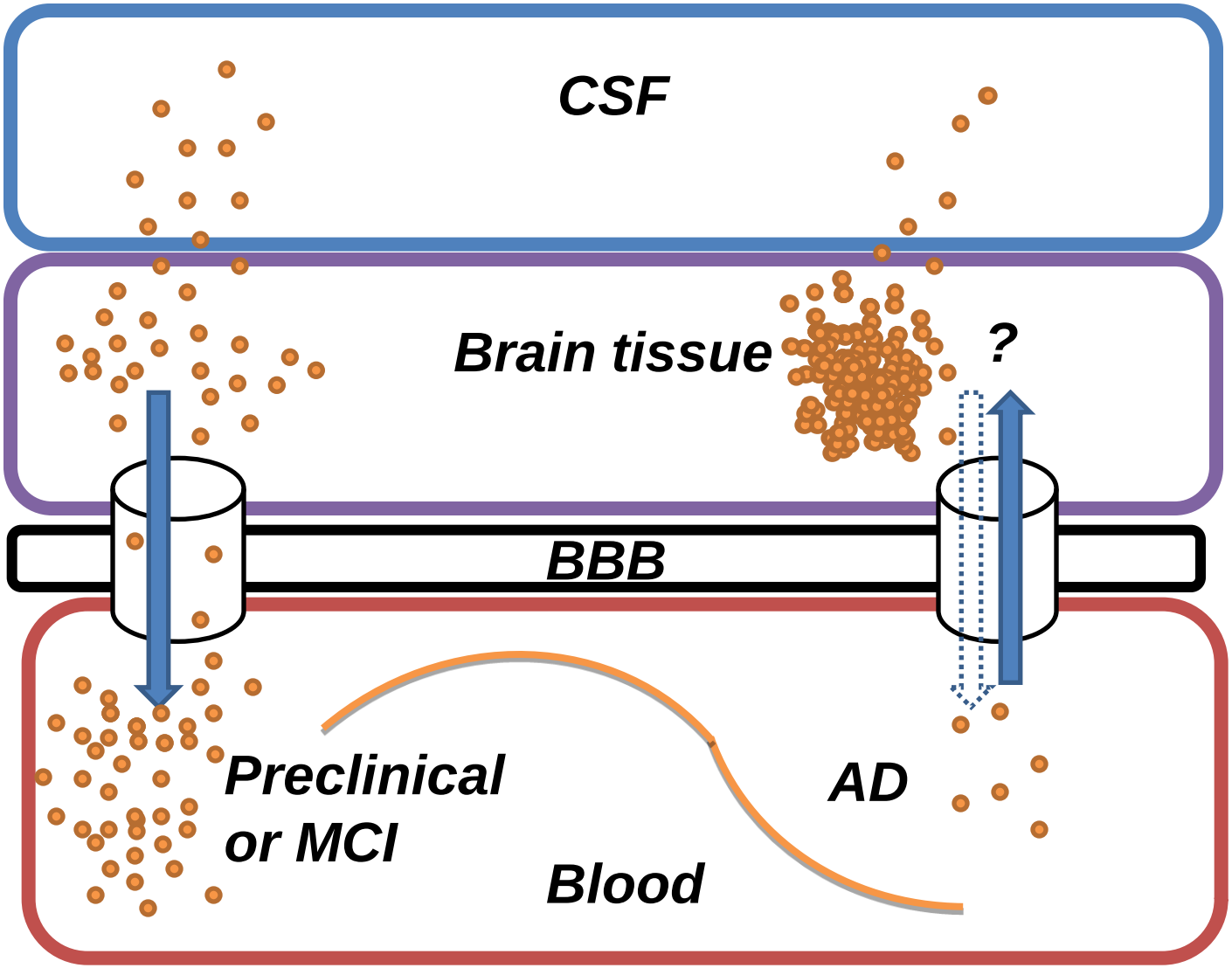
Plasma Abeta

- ***Plasma tau*는 *detection limit*이하**

- *따라서 주로* ***Abeta****가 연구됨 (아래는 주요 이슈들)*

  - ***Sink theory of Abeta:*** *시점에 따라 결과가 다름*
  - ***Abeta****의 종류: 종류에 따라* ***CSF****와 다른 특징*
  - ***Abeta****의* ***binding: albumin****등에* ***binding form****으로 존재*
  - ***Abeta****의* ***generation site: brain*** *외의 말초기관도 생산*
  - ***Detection method****의 어려움:* ***CSF detection*** *보다 어려움*

Sink Theory of Abeta



측정당시 높은 ***plasma Abeta42*** *농도가*
향후 치매발병위험을 ***3*** *배 높였다.*

Table 2. Relation of initial Aβ peptide levels to incidence of AD

Aβ levels	No. at risk	AD, *n* (%)	Hazard rate, model A†	Hazard rate, model B‡
Quartile of Aβ 40				
9.0–34.9	281	21 (6.7)	1.0 (reference)	1.0 (reference)
35–73.35	281	31 (10.6)	1.3 (0.7–2.4)	1.3 (0.6–2.7)
72.5–113.2	282	25 (9.3)	1.0 (0.5–2.1)	1.0 (0.5–2.1)
113.6–588.1	279	27 (10.9)	1.4 (0.7–2.9)	0.9 (0.4–2.1)
Quartile of Aβ 42				
9.0–18.8	282	15 (5.1)	1.0 (reference)	1.0 (reference)
18.85–33.4	281	27 (9.2)	1.9 (0.9–3.8)	2.1 (0.9–4.6)
33.45–49.25	281	31 (11)	2.2 (1.1–4.7)*	2.3 (1.0–5.4)*
49.3–198.7	281	31 (12.1)	3.4 (1.6–7.6)**	3.5 (1.4–8.6)**
Quartiles of Aβ 42/Aβ 40 ratio				
0.07–0.3530	281	23 (8.2)	1.0 (reference)	1.0 (reference)
0.35–0.51	281	24 (8.5)	1.1 (0.6–1.9)	1.2 (0.6–2.2)
0.51–0.75	281	30 (10.7)	1.2 (0.7–2.0)	1.6 (0.8–2.9)
0.75–7.40	279	27 (9.6)	0.9 (0.5–1.7)	0.9 (0.5–1.7)

*, $P < 0.05$; **, $P < 0.01$.

†Cox proportional hazards model and 95% confidence interval, with Aβ 42 and Aβ 40 in the model, unadjusted.

‡Cox proportional hazards model and 95% confidence interval, adjusted for age at baseline, cohort membership, sex, ethnicity, education, BMI, and the presence of the APOE ε 4 allele.

Schupf et al., PNAS, 2008

인지기능이 손상을 보이기 시작하면서부터는
***plasma Abeta42** 농도가 감소하는 것이 치매발병과 연관되었다.*

Table 3. Relation of change Aβ peptide levels to incidence of AD

Change in Aβ levels	No. at risk	AD, *n* (%)	Odds ratio, model A†	Odds ratio, model B‡
Change as a continuous variable				
Change in Aβ 40	1123	104 (9.3)	1.003 (0.99–1.01)	1.002 (0.99–1.01)
Change in Aβ 42	1125	104 (9.2)	0.98 (0.97–0.99)*	0.98 (0.97–0.99)*
Change in Aβ 42/Aβ 40 ratio	1123	104 (9.3)	0.77 (0.55–1.08)	0.7 (0.47–1.01)
Change in Aβ 40 by group				
Increasing	626	59 (9.4)	1.0 (reference)	1.0 (reference)
No change	434	41 (9.4)	0.9 (0.6–1.4)	0.7 (0.4–1.3)
Decreasing	58	4 (6.9)	0.6 (0.2–1.7)	0.5 (0.2–1.7)
Change in Aβ 42 by group				
Increasing	493	39 (7.9)	1.0 (reference)	1.0 (reference)
No change	502	41 (8.2)	1.1 (0.7–1.8)	1.5 (0.7–2.0)
Decreasing	130	24 (17.6)	2.8 (1.6–5.1)***	2.6 (1.3–5.1)**
Change in Aβ 42/Aβ 40 ratio by group				
Increasing	93	3 (2.9)	1.0 (reference)	1.0 (reference)
No change	692	65 (9.4)	3.1 (1.0–10.1)	3.2 (0.9–11)
Decreasing	333	36 (10.8)	3.6 (1.1–12.1)*	3.4 (1.0–11.8)*

*, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$.

†Logistic regression model, with Aβ 42 and Aβ 40 in the model, unadjusted.

‡Logistic regression model, adjusted for age at baseline, cohort membership, sex, ethnicity, education, BMI, and the presence of the APOE ε 4 allele.

Schupf et al., PNAS, 2008

Plasma Abeta42/Abeta40 ratio

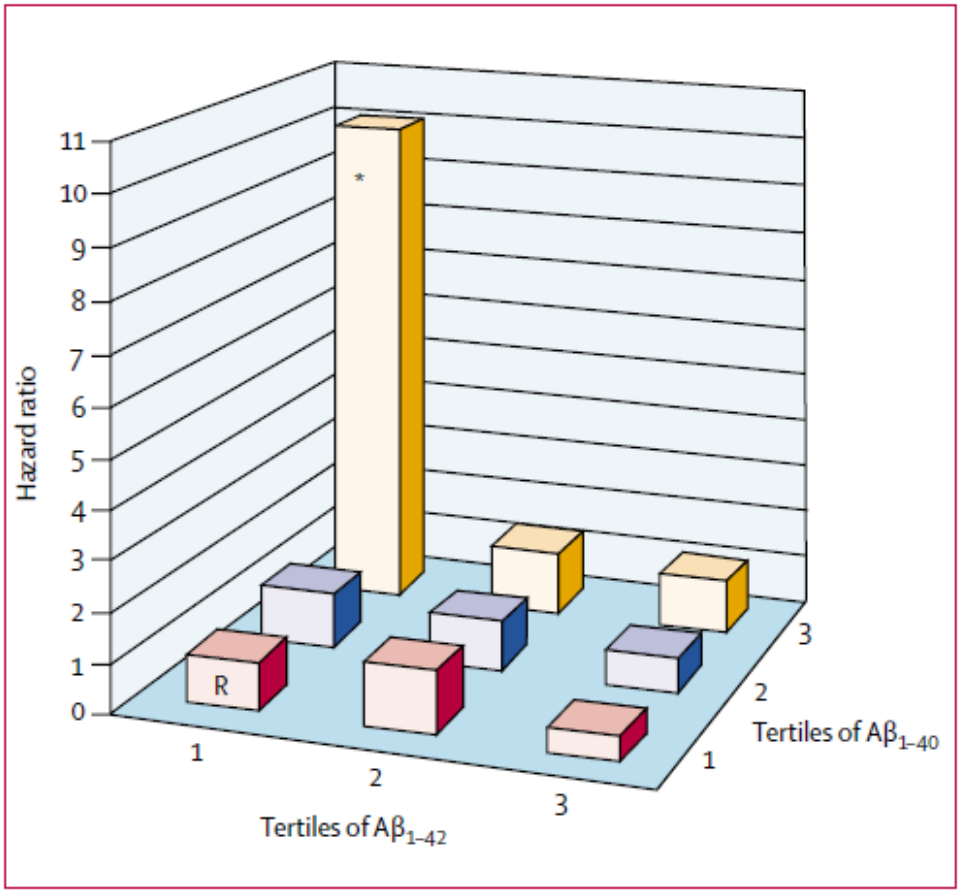
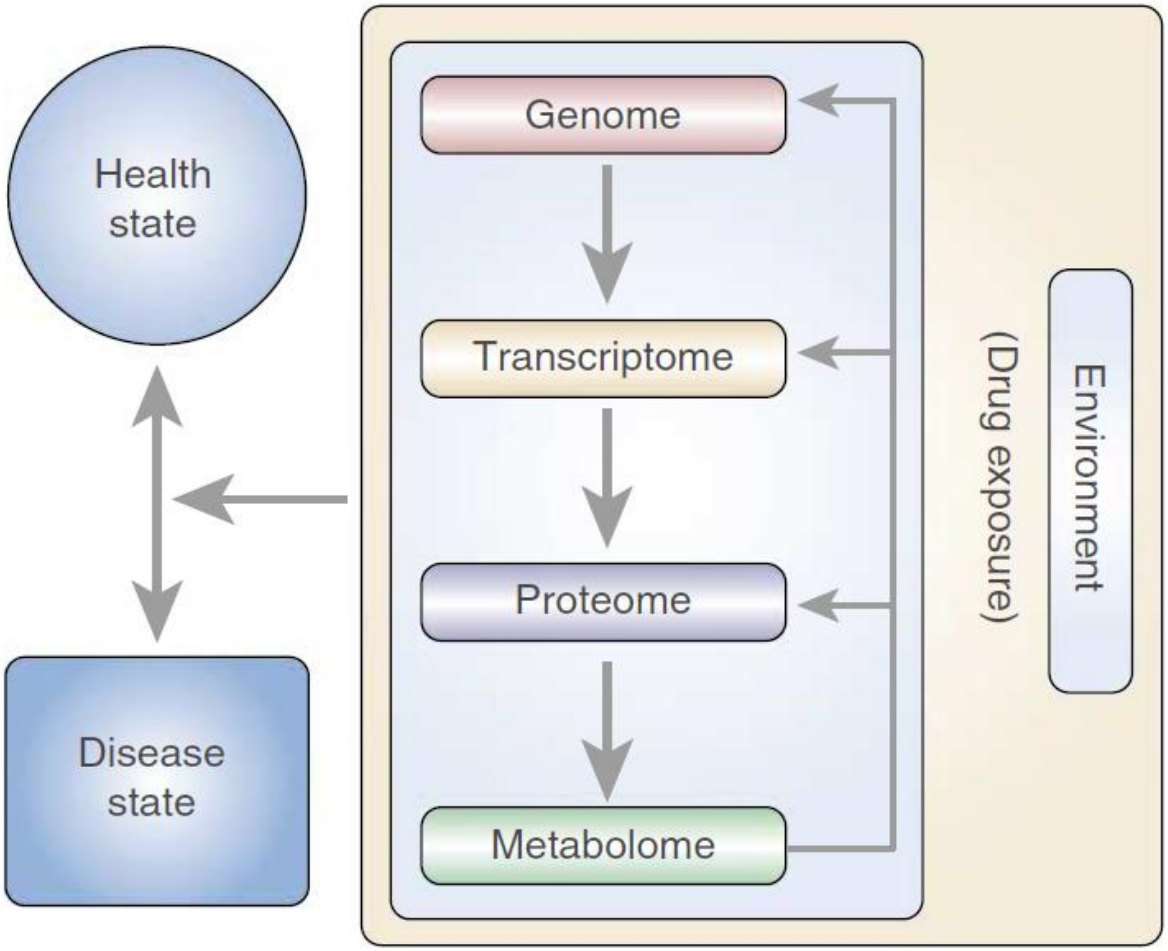


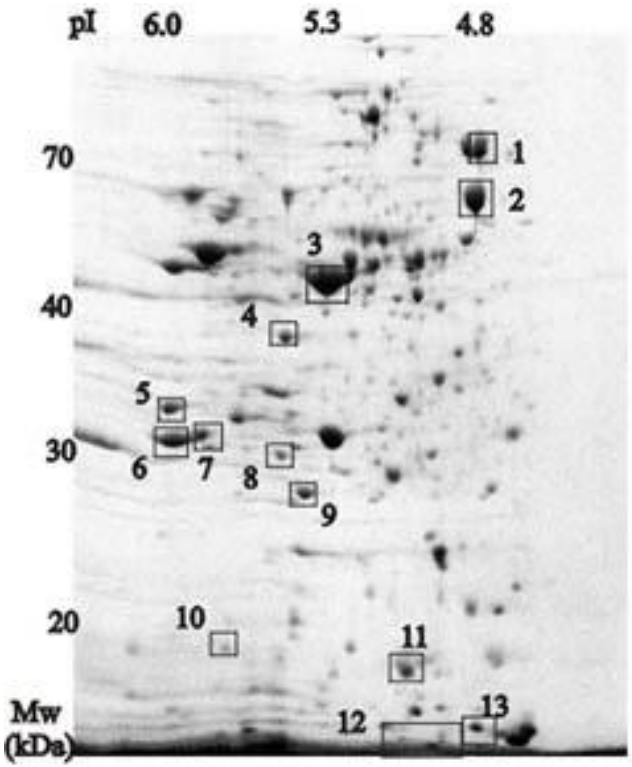
Figure: Hazard ratios for dementia by concentrations of $A\beta_{1-40}$ and $A\beta_{1-42}$
R=reference category (low levels of both of $A\beta_{1-42}$ and $A\beta_{1-40}$). *p<0·0001.

van Oijen et al. Lancet Neurol 2006; 5: 655–60

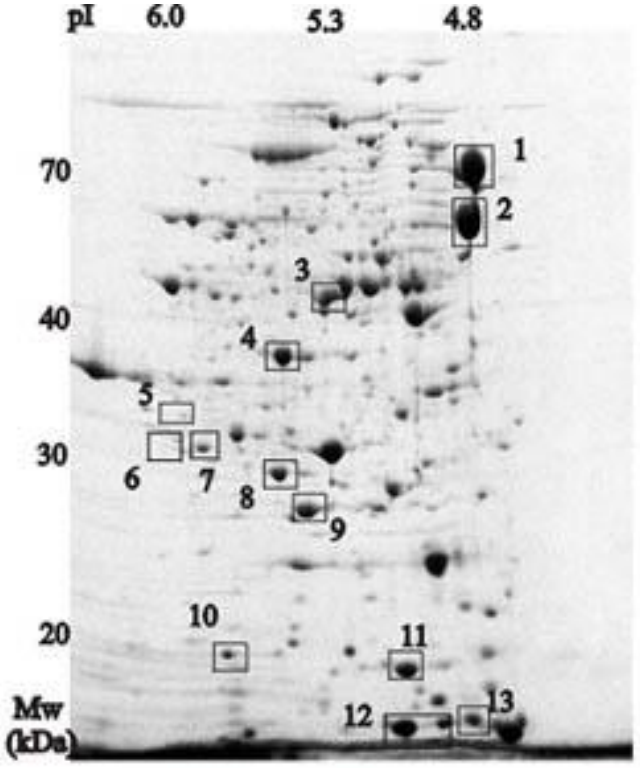
Methodology for searching new peripheral markers



Proteomics



XL1-Blue/pJC4 cultured in LB medium
(No PHB accumulated)



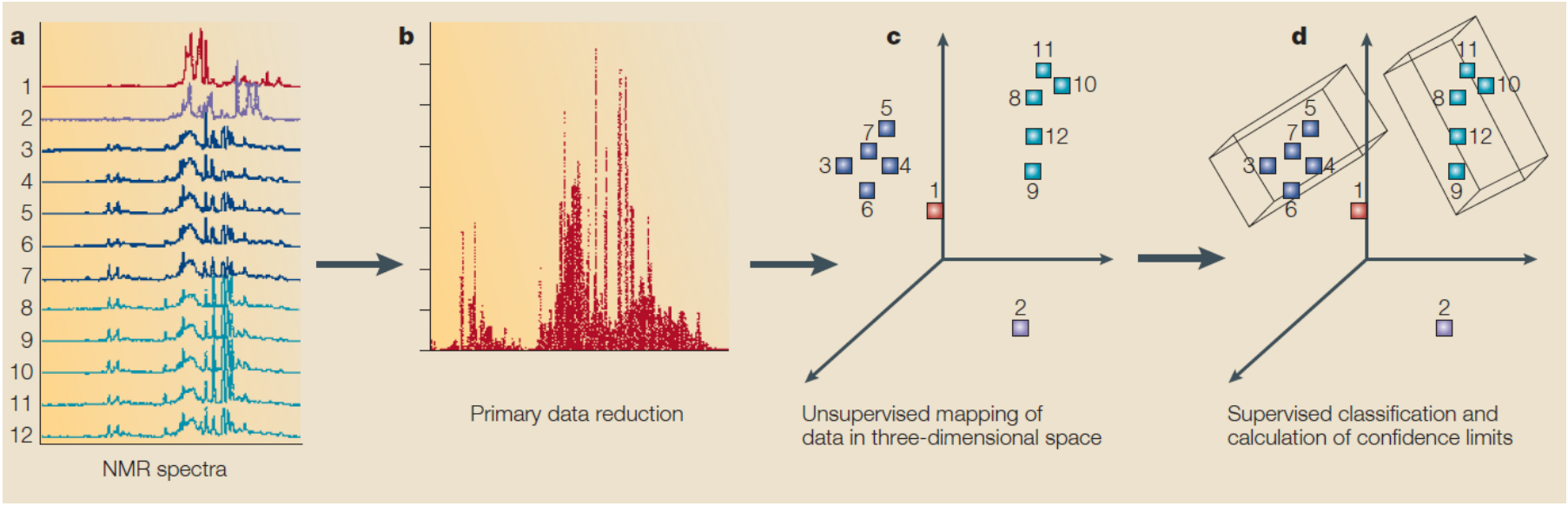
XL1-Blue/pJC4 cultured in LB + Glucose 20g/L
(68% PHB accumulated)

http://mbel.kaist.ac.kr/lab/research/proteomics_en.html

Table 2 Proteins identified in plasma from Alzheimer's disease and age-matched controls

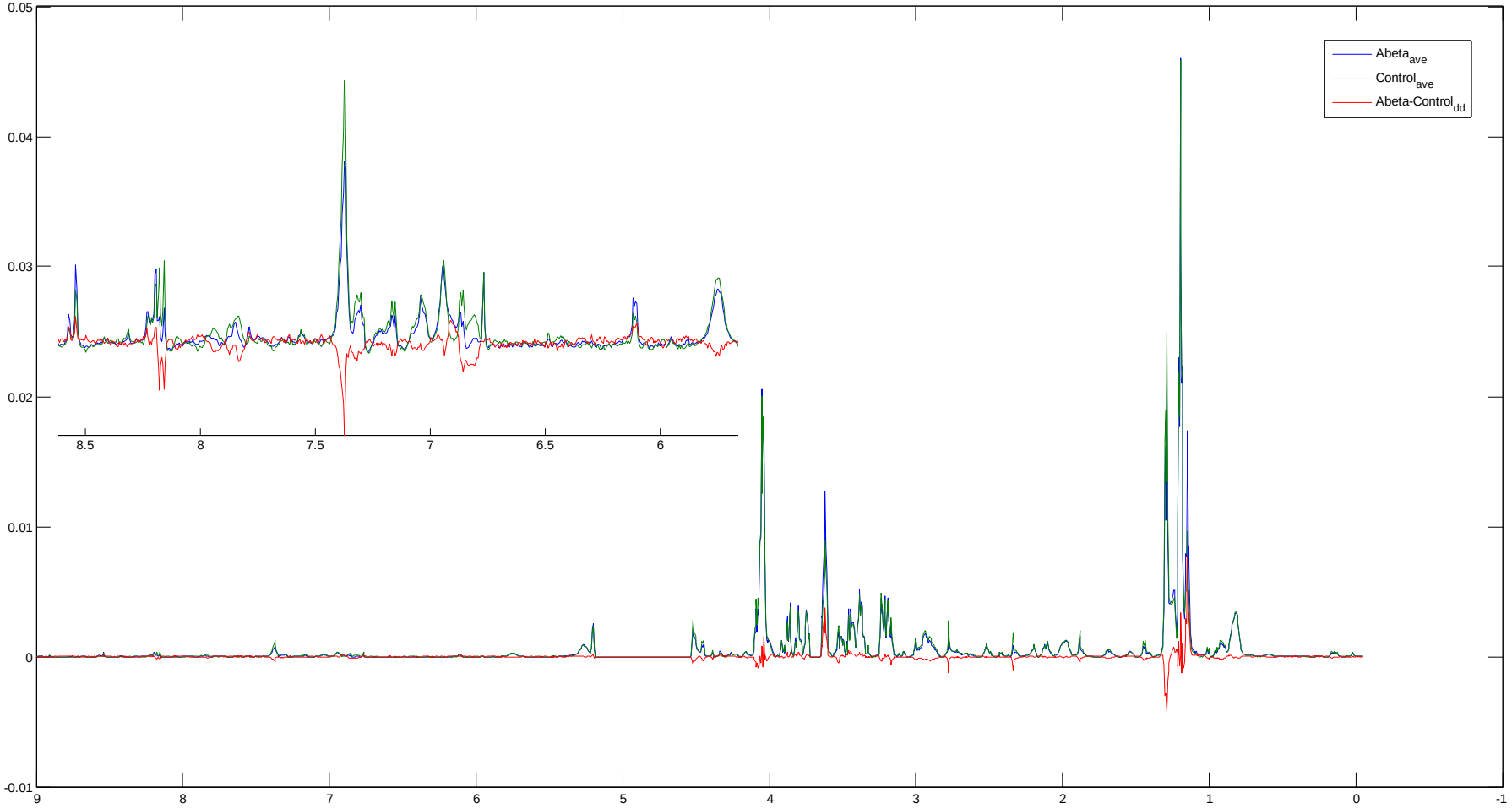
Spot No.	Rank	*P*-value	Fold change	State change	Protein ID	Accession no.
196	1	0.0003	1.78	↑ AD	Desmoplakin	P15924
					Ig kappa chain C region	P01834
					Ig kappa chain V-II region TEW	P01617
					Serum amyloid P-component precursor (SAP)	P02743
171	2	0.0013	2.11	↑ AD	Ig kappa chain C region	P01834
					Serum albumin precursor	P02768
					Galectin-7	P47929
2	3	0.0014	13.75	↑ AD	Complement factor H (CFH) precursor	P08603
					Serum albumin precursor	P02768
					α-$_2$-macroglobulin (α-$_2$M) precursor	P01023
					Ceruloplasmin precursor	P00450
184	4	0.0054	2.43	↑ AD	Ig lambda chain C regions	P01842
					Ig lambda chain V-III region LOI	P80748
					Serum albumin precursor	P02768
					CFH related protein 2 precursor	P36980
177	5	0.0054	1.92	↑ AD	Ig lambda chain C regions	P01842
					Serum albumin precursor	P02768
					Ig lambda chain V-III region LOI	P80748
					Ig kappa chain C region	P01834
4	6	0.0060	8.83	↑ AD	α2-macroglobulin (α-$_2$M) precursor	P01023
170	7	0.0117	1.98	↑ AD	Ig lambda chain C regions	P01842
					Ig kappa chain C region	P01834
					Serum albumin precursor	P02768
					Ig lambda chain V-II region TRO	P01707
					Ig kappa chain V-I region Lay	P01605
					Ig kappa chain V-IV region Len	P01625
13	8	0.0155	4.23	↓ AD	Inter-alpha-trypsin inhibitor heavy chain H4 precursor	Q14624
					Ceruloplasmin precursor	P00450
165	9	0.0183	1.58	↓ AD	Serum albumin precursor	P02768
164	10	0.0206	2.03	↓ AD	Complement C4 precursor	P01028
					Ig gamma-1 chain C region	P01857
14	11	0.0250	10.82	↓ AD	Serum albumin precursor	P02768
					Histone H2B.a/g/h/k/l	P62807
126	12	0.0290	1.6	↓ AD	CD5 antigen-like precursor	O43866
					Serum albumin precursor	P02768
					Ig mu chain C region	P01871
176	13	0.0291	1.75	↑ AD	Ig lambda chain C regions	P01842
					Serum albumin precursor	P02768
					Ig lambda chain V-III region LOI	P80748
123	14	0.0314	1.36	↑ AD	Serum albumin precursor	P02768
1	15	0.0347	3.32	↑ AD	α-$_2$M precursor	P01023
					Ig alpha-1 chain C region	P01876

Metabolomics



Difference Spectrum

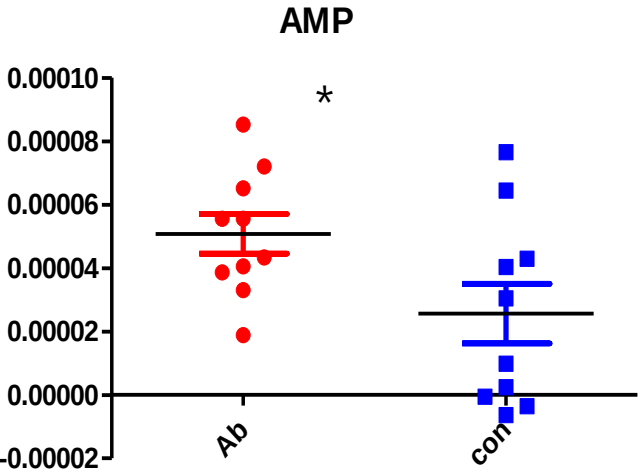
between the mean spectrum of Aβs and the mean spectrum of Controls

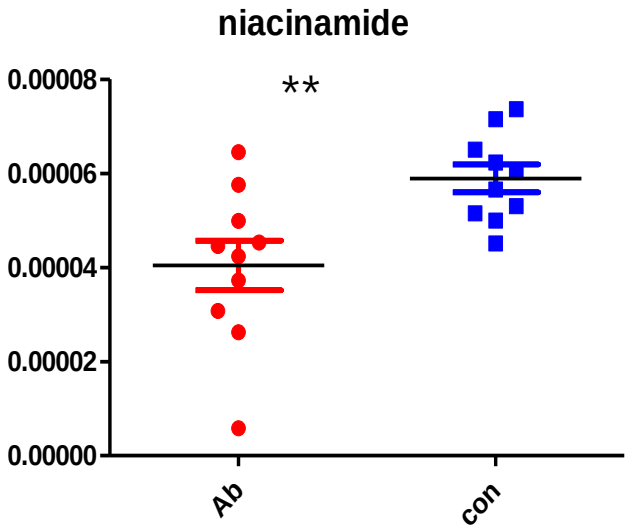


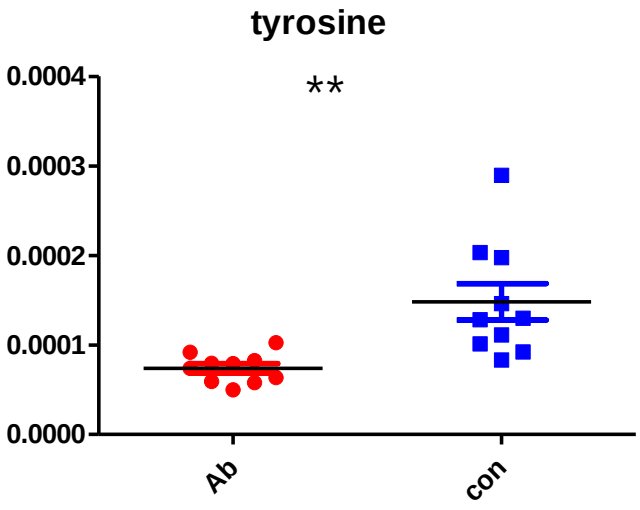
Kim et al. Unpublished data

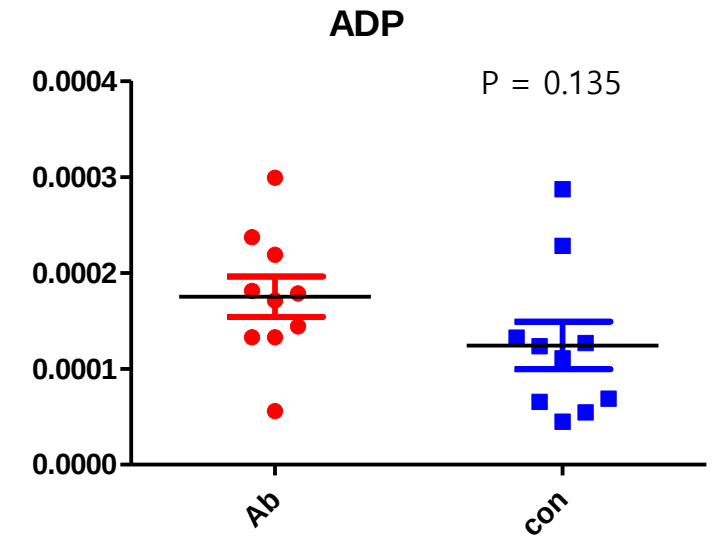
t-tests of the metabolites which are considered to play important role to separate Aβ group and Control group

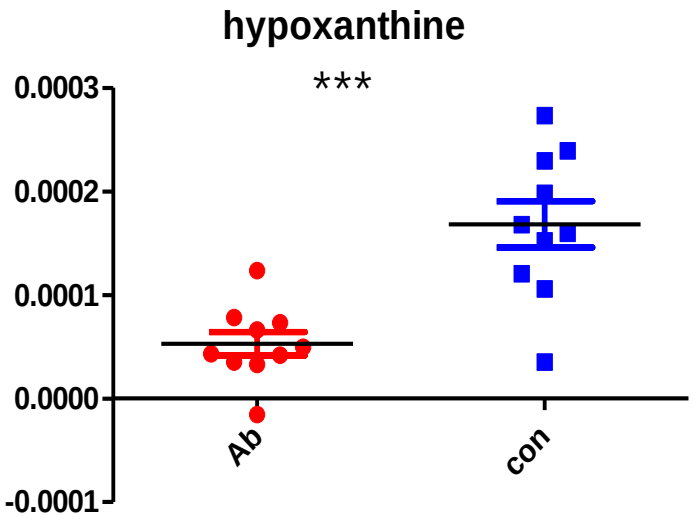
Metabolites at down-field







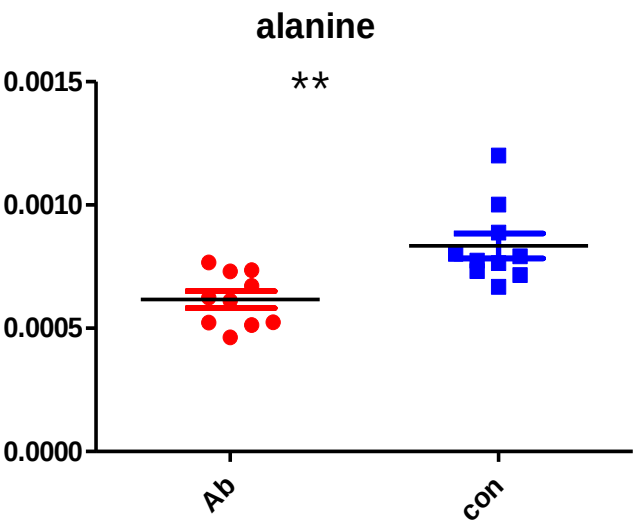


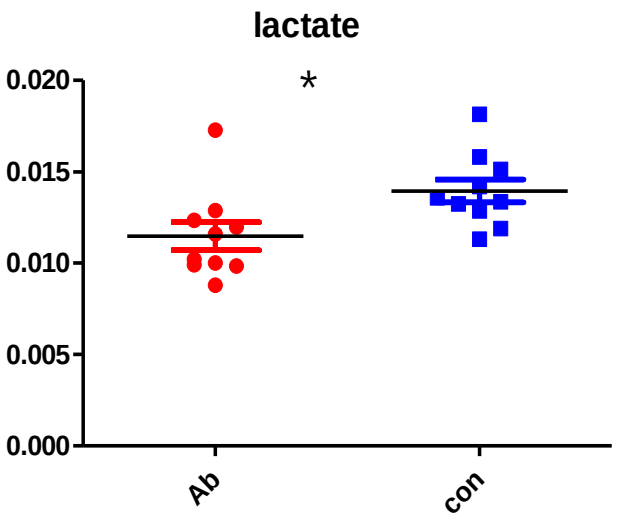


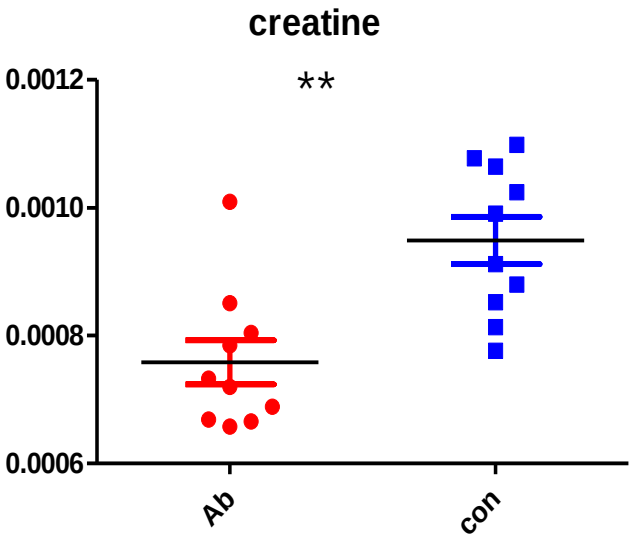
Kim et al. Unpublished data

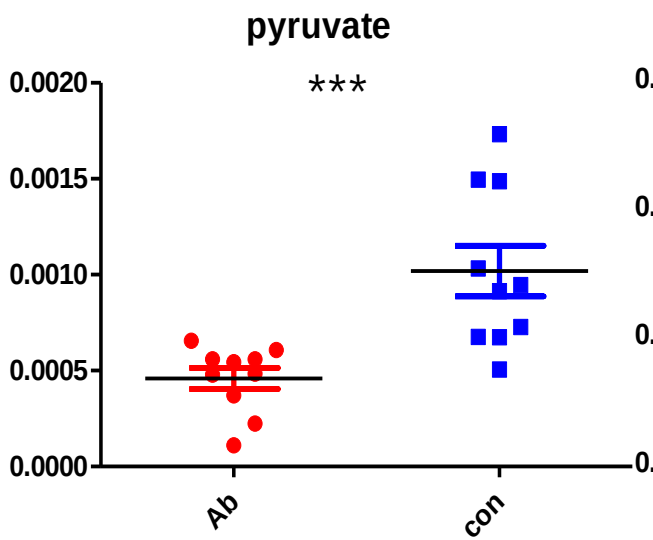
t-tests of the metabolites which are considered to play important role to separate Aβ group and Control group

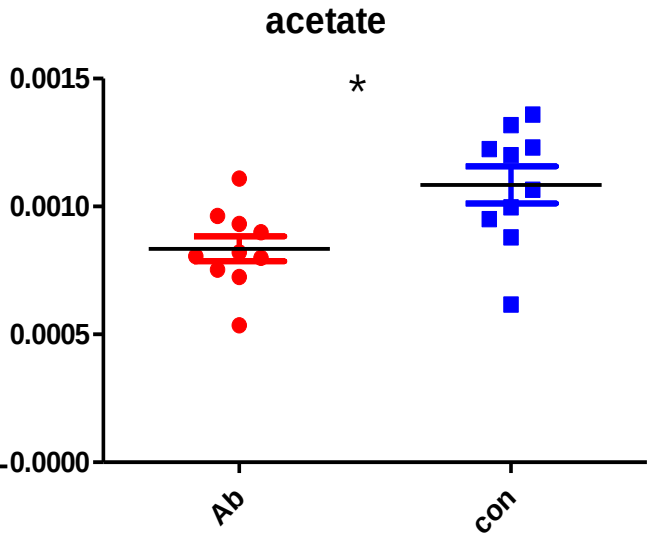
Metabolites at up-field

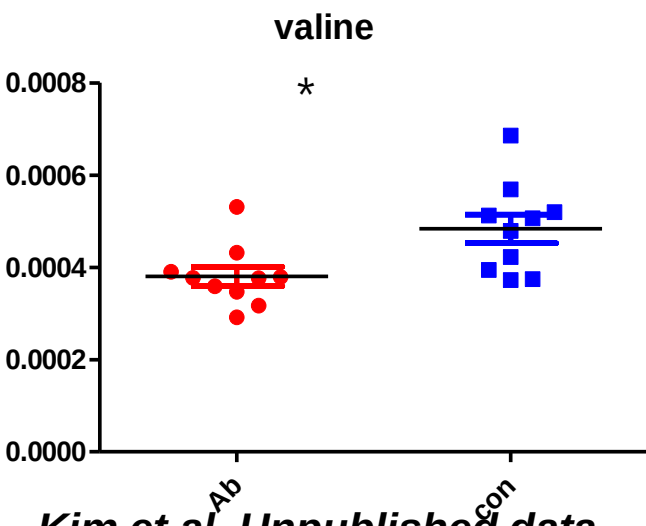




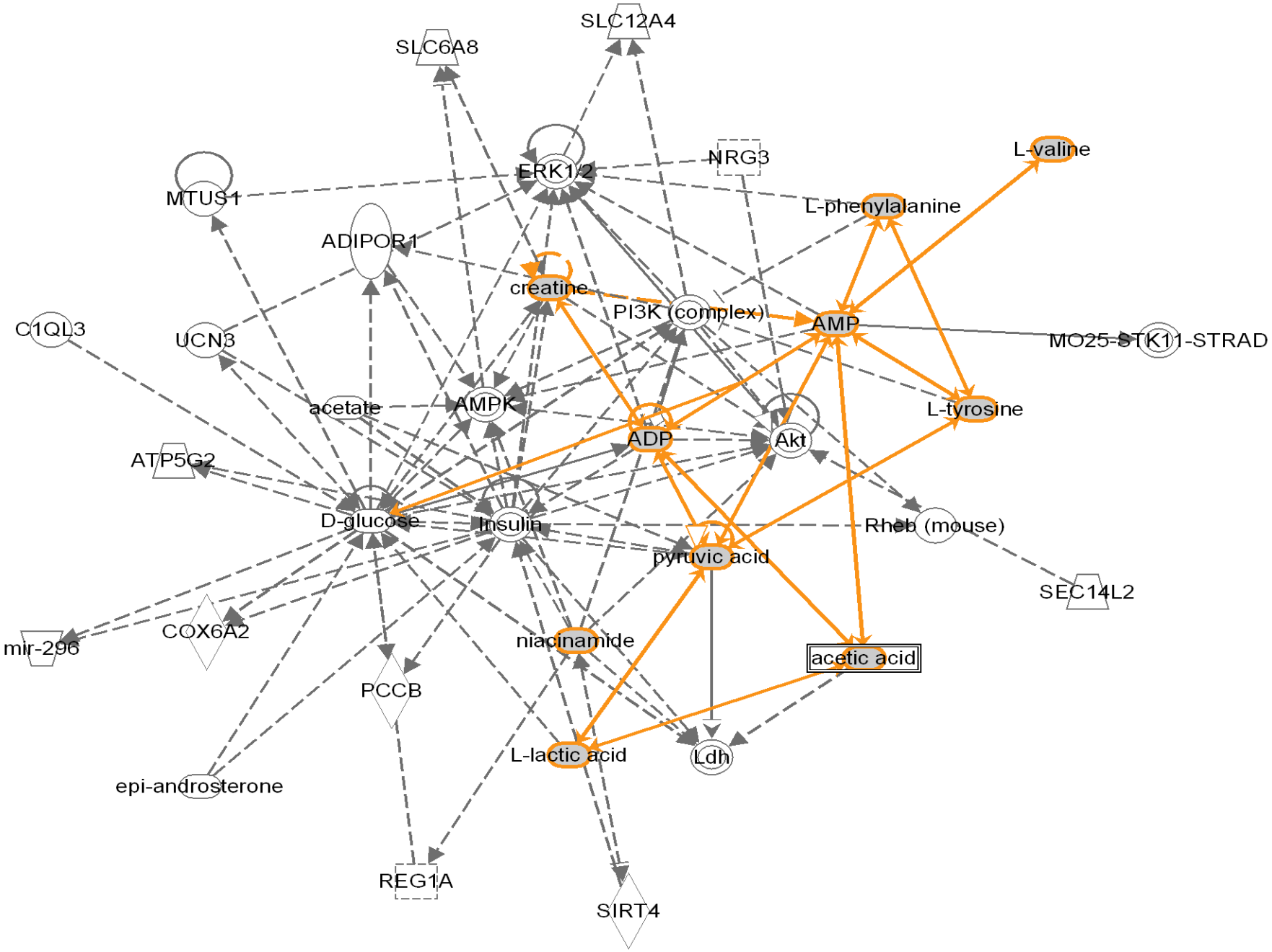








Kim et al. Unpublished data


SLC12A4
SLC6A8
NRG3
ERK1/2
L-valine
MTUS1
L-phenylalanine
ADIPOR1
creatine
PI3K (complex)
AMP
C1QL3
UCN3
MO25-STK11-STRAD
acetate
AMPK
L-tyrosine
ADP
Akt
ATP5G2
D-glucose
Insulin
Rheb (mouse)
pyruvic acid
SEC14L2
mir-296
COX6A2
niacinamide
acetic acid
PCCB
L-lactic acid
Ldh
epi-androsterone
REG1A
SIRT4

ORIGINAL ARTICLE

Association of Plasma Clusterin Concentration With Severity, Pathology, and Progression in Alzheimer Disease

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Context: Blood-based analytes may be indicators of pathological processes in Alzheimer disease (AD).

Objective: To identify plasma proteins associated with AD pathology using a combined proteomic and neuroimaging approach.

Design: Discovery-phase proteomics to identify plasma proteins associated with correlates of AD pathology. Confirmation and validation using immunodetection in a replication set and an animal model.

Setting: A multicenter European study (AddNeuroMed) and the Baltimore Longitudinal Study of Aging.

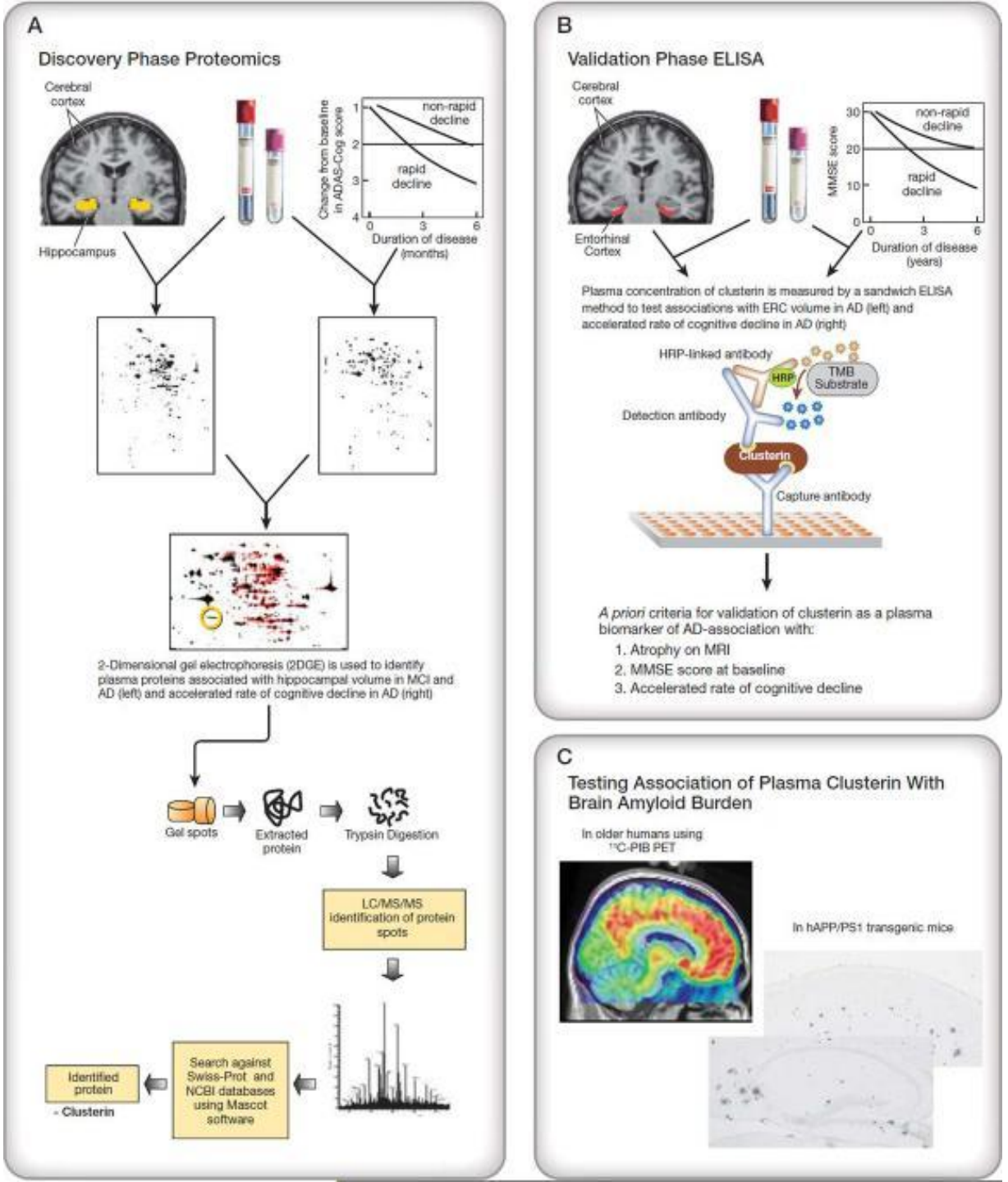
Participants: Patients with AD, subjects with mild cognitive impairment, and healthy controls with standardized clinical assessments and structural neuroimaging.

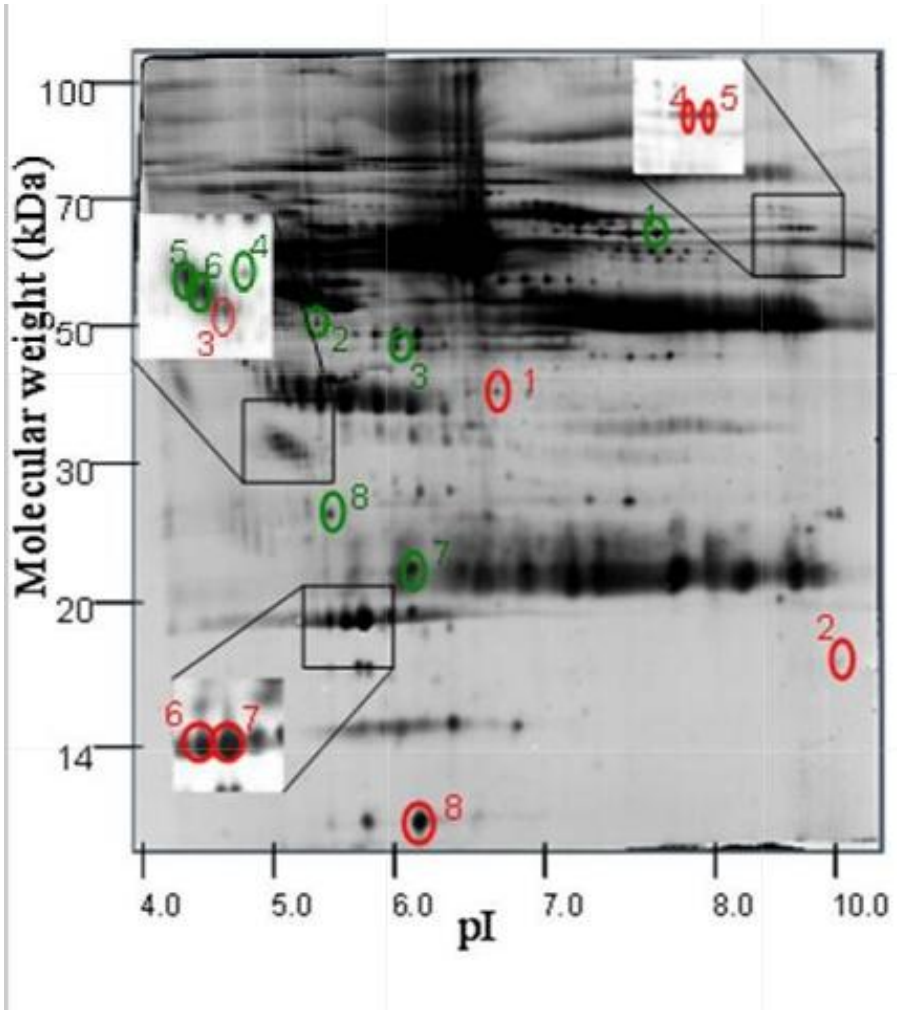
Main Outcome Measures: Association of plasma proteins with brain atrophy, disease severity, and rate of clinical progression. Extension studies in humans and transgenic mice tested the association between plasma proteins and brain amyloid.

Results: Clusterin/apolipoprotein J was associated with atrophy of the entorhinal cortex, baseline disease severity, and rapid clinical progression in AD. Increased plasma concentration of clusterin was predictive of greater fibrillar amyloid-β burden in the medial temporal lobe. Subjects with AD had increased clusterin messenger RNA in blood, but there was no effect of single-nucleotide polymorphisms in the gene encoding clusterin with gene or protein expression. *APP/PS1* transgenic mice showed increased plasma clusterin, age-dependent increase in brain clusterin, as well as amyloid and clusterin colocalization in plaques.

Conclusions: These results demonstrate an important role of clusterin in the pathogenesis of AD and suggest that alterations in amyloid chaperone proteins may be a biologically relevant peripheral signature of AD.

Arch Gen Psychiatry. 2010;67(7):739-748

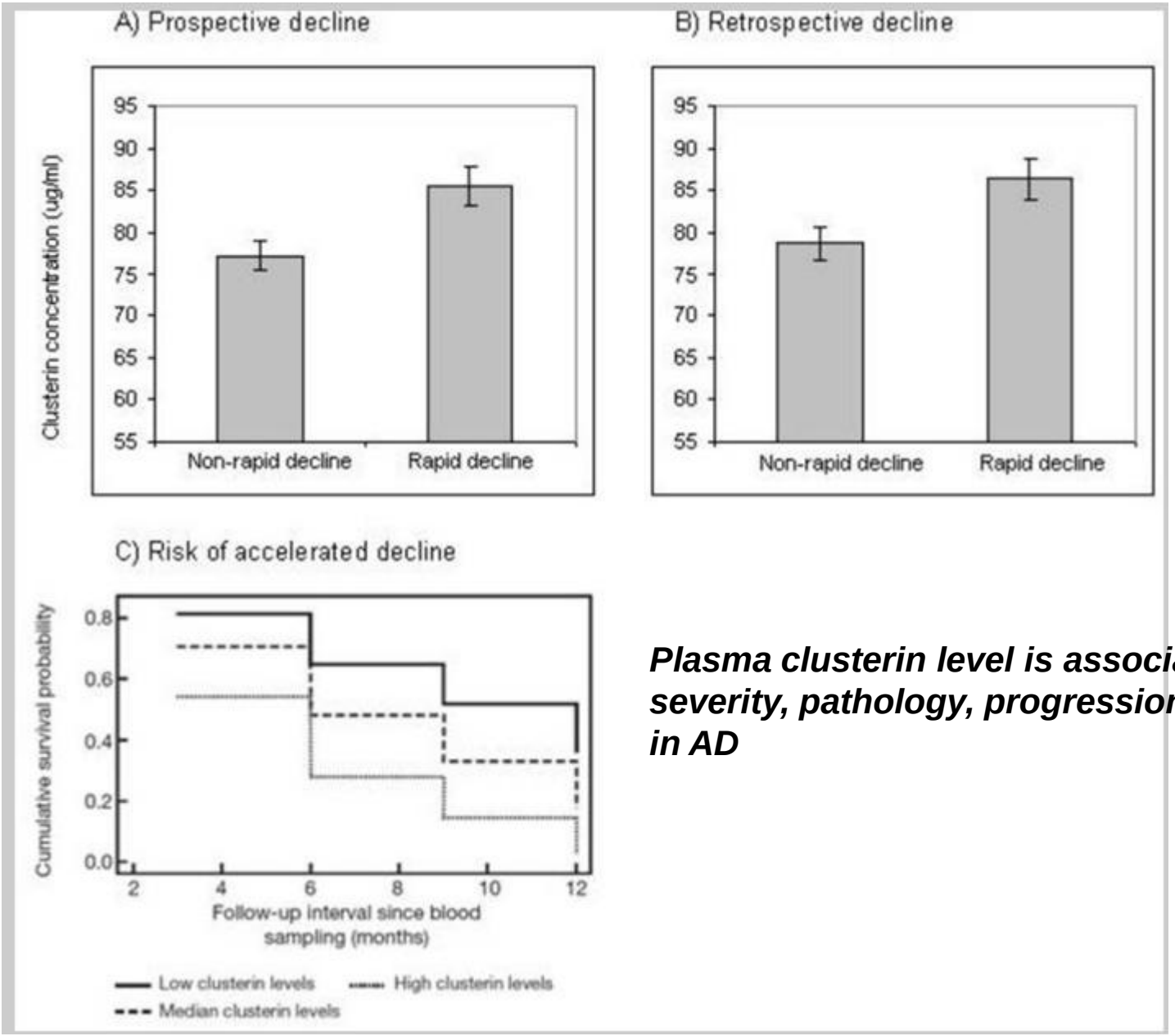

A
Discovery Phase Proteomics
Cerebral cortex
Hippocampus
Change from baseline in ADAS-Cog score
non-rapid decline
rapid decline
Duration of disease (months)
2-Dimensional gel electrophoresis (2DGE) is used to identify plasma proteins associated with hippocampal volume in MCI and AD (left) and accelerated rate of cognitive decline in AD (right)
Gel spots
Extracted protein
Trypsin Digestion
LC/MS/MS identification of protein spots
Search against Swiss-Prot and NCBI databases using Mascot software
Identified protein
- Clusterin
B
Validation Phase ELISA
Cerebral cortex
Entorhinal Cortex
MMSE score
non-rapid decline
rapid decline
Duration of disease (years)
Plasma concentration of clusterin is measured by a sandwich ELISA method to test associations with ERC volume in AD (left) and accelerated rate of cognitive decline in AD (right)
HRP-linked antibody
HRP
TMB Substrate
Detection antibody
Clusterin
Capture antibody
A priori criteria for validation of clusterin as a plasma biomarker of AD-association with:
1. Atrophy on MRI
2. MMSE score at baseline
3. Accelerated rate of cognitive decline
C
Testing Association of Plasma Clusterin With Brain Amyloid Burden
In older humans using ^{11}C-PIB PET
In hAPP/PS1 transgenic mice



Protein ID ○	Accession Number
[1]Complement C3	P01024
[2]γ Fibrinogen	P02679
[3]Serum albumin	P02768
[4]Complement factor-I	P05156
[5]Clusterin	P10909
[6]Clusterin	P10909
[7]Serum amyloid-P	P02743
[8]α-1-macroglobulin	P02760

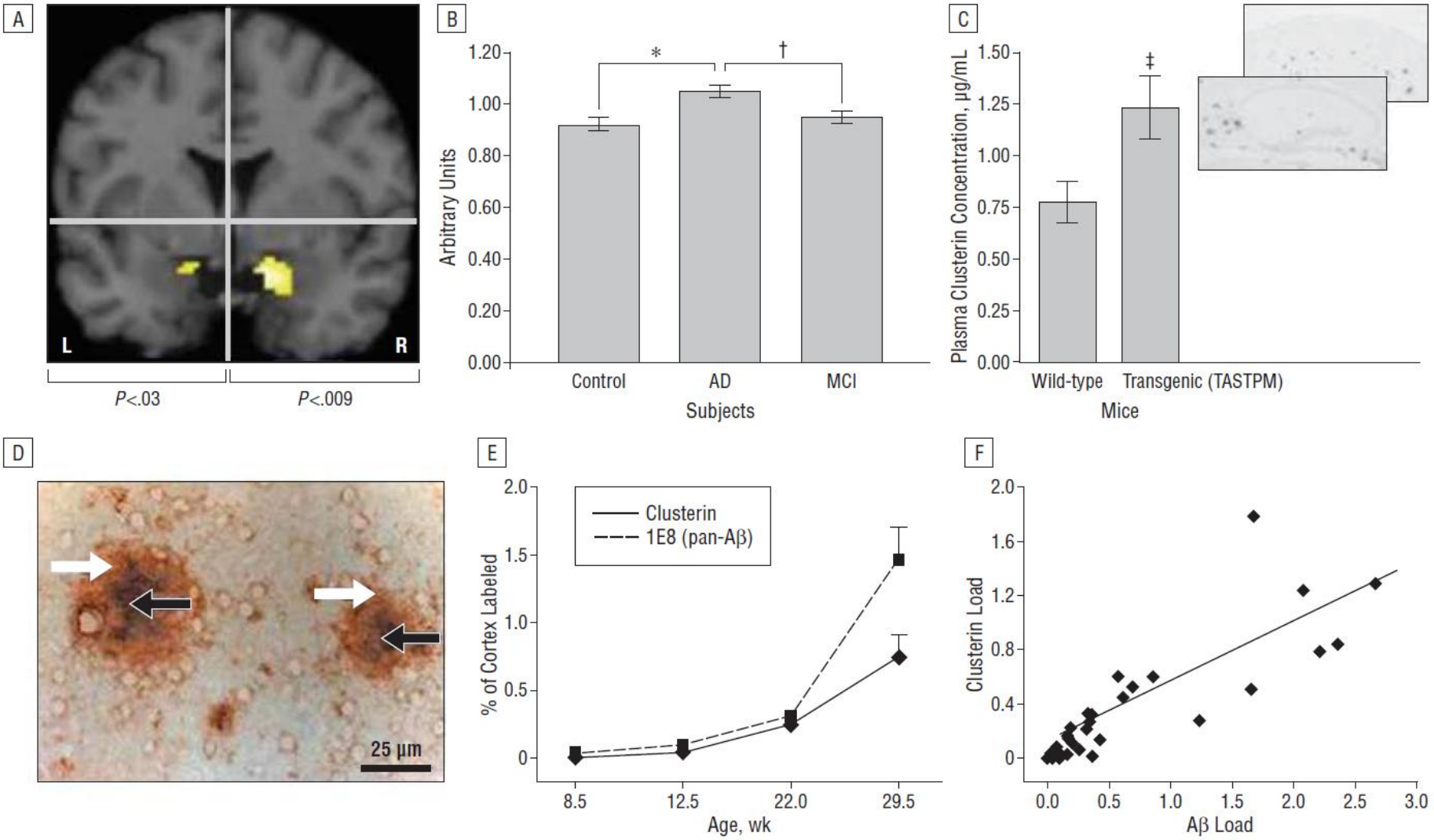
Protein ID ○	Accession Number
[1]Complement C4a	P0C0L4
[2]Complement component C8	P07360
[3]Clusterin	P10909
[4]Complement C4a	P0C0L4
[5]Complement C4a	P0C0L4
[6]Apolipoprotein-A1	P02647
[7]Apolipoprotein-A1	P02647
[8]Transthyretin	P02766

Thambisetty et al. Arch Gen Psych, 2010.



Plasma clusterin level is associated with severity, pathology, progression in AD

Thambisetty et al. Arch Gen Psych, 2010.



Thambisetty et al. Arch Gen Psych, 2010.

Wrap-up

- ***CSF study**의 유용성은 상당한 수준으로 인정된다.*
  - *그룹별 경향은 분명히 차이를 보여줄 수 있으나, 개인별 임상 유용성(정보력)에는 한계가 있다 (high variance).*
  - *실제 임상 적용은 거부감으로 한계가 있다.*
  - *가장 현실적인 것은 한 개인 내에서 신약사용 후 병리변화를 보는 것이다 (disease-modifying drug); 특히 at-risk MCI population*

- ***Peripheral marker**는 두 가지로 분류된다.*
  - *핵심 병리 관련 물질(Abeta, tau) 혹은 omics novel marker 발굴*
  - *한계가 많으나 계속 도전할만 하다 (clusterin의 재발견)*

- *감사합니다.*