

Why Is Amyloid- β PET Requested After Performing CSF Biomarkers?

Juhan Reimand^{a,b,c,*}, Colin Groot^a, Charlotte E. Teunissen^d, Albert D. Windhorst^e, Ronald Boellaard^e, Frederik Barkhof^{e,f}, Sergei Nazarenko^b, Wiesje M. van der Flier^{a,g}, Bart N.M. van Berckel^e, Philip Scheltens^a, Rik Ossenkoppele^{a,h} and Femke Bouwman^a

^a*Department of Neurology & Alzheimer Center Amsterdam, Amsterdam Neuroscience, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, the Netherlands*

^b*Department of Health Technologies, Tallinn University of Technology, Tallinn, Estonia*

^c*Radiology Centre, North Estonia Medical Centre, Tallinn, Estonia*

^d*Neurochemistry Laboratory, Department of Clinical Chemistry, Amsterdam Neuroscience, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, The Netherlands*

^e*Department of Radiology & Nuclear Medicine, Amsterdam Neuroscience, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, The Netherlands*

^f*Centre for Medical Image Computing, Medical Physics and Biomedical Engineering, UCL, United Kingdom*

^g*Department of Epidemiology & Biostatistics, Amsterdam Neuroscience, Vrije Universiteit Amsterdam, Amsterdam UMC, Amsterdam, the Netherlands*

^h*Clinical Memory Research Unit, Lund University, Lund, Sweden*

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Abstract.

Background: Amyloid- β positron emission tomography (PET) and cerebrospinal fluid (CSF) A β ₄₂ are considered interchangeable for clinical diagnosis of Alzheimer's disease.

Objective: To explore the clinical reasoning for requesting additional amyloid- β PET after performing CSF biomarkers.

Methods: We retrospectively identified 72 memory clinic patients who underwent amyloid- β PET after CSF biomarkers analysis for clinical diagnostic evaluation between 2011 and 2019. We performed patient chart reviews to identify factors which led to additional amyloid- β PET. Additionally, we assessed accordance with appropriate-use-criteria (AUC) for amyloid- β PET.

Results: Mean patient age was 62.0 (SD = 8.1) and mean Mini-Mental State Exam score was 23.6 (SD = 3.8). CSF analysis conflicting with the clinical diagnosis was the most frequent reason for requesting an amyloid- β PET scan ($n = 53$, 74%), followed by incongruent MRI ($n = 16$, 22%), unusual clinical presentation ($n = 11$, 15%) and young age ($n = 8$, 11%). An amyloid- β PET scan was rarely ($n = 5$, 7%) requested in patients with a CSF A β +/ τ + status. Fifteen (47%) patients with a post-PET diagnosis of AD had a predominantly non-amnesic presentation. In $n = 11$ (15%) cases, the reason that the clinician requested amyloid- β was not covered by AUC. This happened most often ($n = 7$) when previous CSF analysis did not support current clinical diagnosis, which led to requesting amyloid- β PET.

Conclusion: In this single-center study, the main reason for requesting an amyloid- β PET scan after performing CSF biomarkers was the occurrence of a mismatch between the primary clinical diagnosis and CSF A β / τ results.

Keywords: Alzheimer's disease, amyloid, cerebrospinal fluid, positron emission tomography, tau proteins

*Correspondence to: Juhan Reimand, MD, Alzheimer Center Amsterdam, VU University Medical Center, P.O. Box 7057, 1007 MB, Amsterdam, The Netherlands. Tel.: +31 20 4443506;

Fax: +31 20 4448529; E-mails: j.reimand@amsterdamumc.nl and jreimand@gmail.com

INTRODUCTION

Two methods are currently employed in the clinic to capture *in vivo* amyloid- β pathology, a pathological hallmark of Alzheimer's disease (AD) [1, 2]. A β_{42} levels in cerebrospinal fluid (CSF) reflect the soluble amyloid- β pool that has been shown to correlate with amyloid- β depositions in the brain [3]. Alternatively, amyloid- β positron emission tomography (PET) can be employed to directly visualize parenchymal fibrillary amyloid- β depositions [4]. Although CSF and PET yield conflicting results in 10–20% of patients [5–7], they are nonetheless considered interchangeable for clinical use [8].

In our center, all patients are offered CSF biomarker analysis. However, despite the availability of CSF biomarkers, occasionally amyloid- β PET-scans are requested. The diagnostic value of amyloid- β PET to a standard dementia screening has been established in many studies [9–11], but few studies have included subgroups of people with available CSF biomarkers. Reported reasons for performing amyloid- β PET in such cases included incongruent CSF biomarkers in patients with suspicion of AD or atypical clinical presentation [12, 13]. We aimed to elucidate this practice by exploring the clinical reasoning for requesting amyloid- β PET after CSF biomarkers were disclosed, and to characterize the population that received amyloid- β PET after CSF examination.

Additionally, appropriate use criteria (AUC) for amyloid- β PET have been published to support the implementation of clinical amyloid- β PET, advocating use in three groups most likely to benefit: patients with an atypical clinical presentation or mixed etiology, persistent unexplained mild cognitive impairment (MCI), and unexplained dementia in young patients [14]. As a secondary goal, we aimed to compare our clinical practice against current amyloid- β PET AUC.

METHODS

Patient inclusion

We identified 209 cases from the Amsterdam Dementia Cohort with a [^{11}C]-Pittsburgh Compound B (PIB) PET scan between 2011 and 2019 (Fig. 1). We excluded $n = 85$ cases who underwent PIB-PET for research purposes, $n = 4$ cases with CSF analysis performed after amyloid- β PET, and $n = 2$ cases with unknown CSF disclosure. Although lumbar puncture

(LP) is offered to all patients visiting our center, it was not performed for $n = 46$ patients, most often because LP was either not successful ($n = 14$, 30%), not possible ($n = 13$, 28%), or refused by the patient ($n = 12$, 26%). Finally, we included 72 cases with a clinically requested amyloid- β PET scan performed after CSF biomarkers examination for our analysis.

Cerebrospinal fluid

CSF was obtained by LP, using a 25-gauge needle and a syringe [15]. Samples were collected in polypropylene collection tubes and centrifuged at 1800 g for 10 min at 4°C. Thereafter, samples were frozen at -20°C until routine biomarker analysis. Manual analyses of A β_{42} , total tau, and phosphorylated tau (p-tau) were performed using sandwich ELISAs (Innotest assays: β -amyloid1-42, tTAU-Ag and PhosphoTAU-181p; Fujirebio) in the Neurochemistry Laboratory of the Department of Clinical Chemistry of Amsterdam UMC. In a few cases, CSF analysis was performed using automated assays for A β_{42} ($n = 9$), t-tau ($n = 1$), and p-tau ($n = 1$), due to change in routine methods (Elecsys CSF, Roche Diagnostics GmbH) [16]. Additionally, in $n = 12$ cases analyses were performed in the Department of Laboratory Medicine in Radboud UMC prior to referral to our center.

The clinical cut-off values for CSF A β_{42} have repeatedly been changed over the years due to the gradual upward drift of median CSF A β_{42} values observed in our cohort, possibly due to changes in ELISA kits and/or calibration data that are influenced by the presence of A β_{42} aggregates [17]. In order to pool all available CSF values (both local and external) in relation to different cut-offs, we created standardized values by calculating, per patient, the percentage of the CSF value relative to its concurrent cut-off. For example, a value of 150 would represent a normal CSF A β_{42} being 50% higher than the cut-off, whereas a value of 80 represented a pathologically decreased CSF A β_{42} being 20% below the cut-off value.

Positron emission tomography

Amyloid- β PET is not part of the standard diagnostic process in the Amsterdam Dementia Cohort, therefore most of the amyloid- β PET scans in our center are performed for research purposes. We only included scans with [^{11}C]-PIB, as clinically requested PET scans in our center are routinely per-

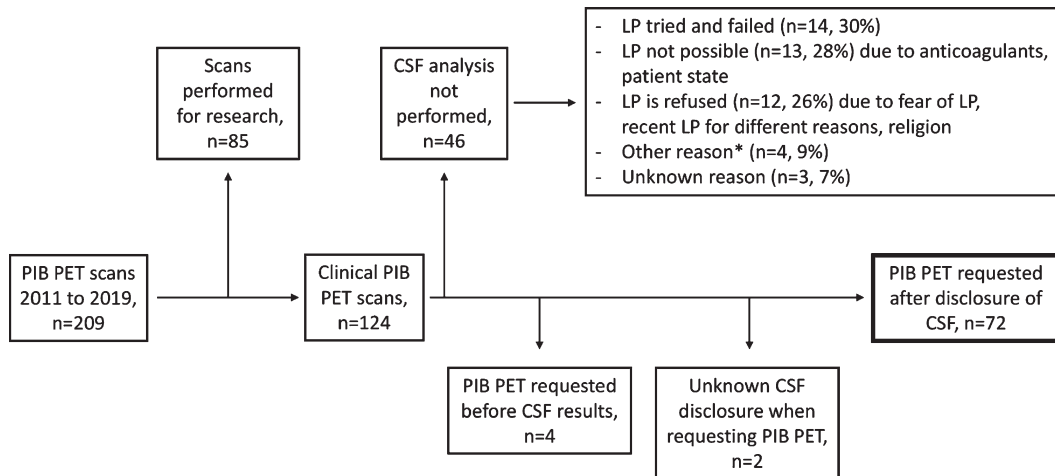


Fig. 1. Flowchart for patient inclusion. *Other reasons include normal pressure hydrocephalus, increased certainty received from amyloid- β PET imaging, and imaging having a greater influence on convincing patients. CSF, cerebrospinal fluid; LP, lumbar puncture; PIB PET, positron emission tomography with ^{11}C -Pittsburgh compound B.

formed using [^{11}C]-PIB as the radiotracer. These scans were performed using the following PET scanners: ECAT EXACT HR+ scanner (Siemens Healthcare, Germany) and Gemini TF PET/CT or Ingenuity TF PET-CT (Philips Medical Systems, the Netherlands). PET scans were performed within a median of 140 [IQR = 67, 260] days after LP. PET scans were rated as positive or negative based on visual read by an expert nuclear medicine physician [11]. Although intra-rater agreement was not available for this sample, in previous work using [^{11}C]-PIB PET, our nuclear medicine physician showed excellent (Fleiss $k = 0.88$) and good to moderate (Fleiss $k = 0.59$ and 0.68) inter-reader agreement for standardized uptake value (SUV), SUV ratio and non-displaceable binding potential images, respectively [18].

Magnetic resonance imaging (MRI)

MRI was performed as described previously [19]. The scans were visually assessed by a neuroradiologist on three different image planes for posterior cortical atrophy [20], medial temporal atrophy [21], and global cortical atrophy [22], which were thereafter age-normalized [23]. The extent of white matter hyperintensities was rated according to the Fazekas scale [24]. Additionally, the scans were assessed for the existence of lacunes and microbleeds. An external scan was used in $n = 19$ cases, and MRI was not available in $n = 2$ cases ($n = 1$ with available computed tomography).

Neuropsychological testing

Patients underwent extensive neuropsychological testing as part of their diagnostic process. We used Mini-Mental State Examination (MMSE) scores to measure global cognition. Additionally, we derived z-scores of various neuropsychological tests using the mean and standard deviation values from a group of healthy controls ($n = 360$), whose mean age was 57.8 (standard deviation [SD] = 8.3) and mean MMSE was 28.2 (SD = 1.9). Thereafter, we compiled composite scores for five cognitive domains (memory, language, attention, executive, and visuospatial) [25].

Reasons for requesting amyloid- β PET

Our main objective was to explore the clinical reasoning for requesting an amyloid- β PET after disclosure of CSF biomarkers. Therefore, J.R. and F.Bo. performed patient chart reviews to retrieve the clinical reasoning for requesting the amyloid- β PET scan. Patients were divided into two groups (AD versus non-AD) based on the most likely etiological diagnosis prior to performing a PIB PET scan. For both diagnostic groups we listed characteristics that were recorded as not compatible with the current etiological diagnosis, therefore leading to additional amyloid- β PET. Listed reasons included incongruent findings from biomarkers (CSF, imaging, EEG) or patient history and presentation, as well as other supporting factors such as age, patient wish and implementation of a new CSF assay. For example,

in the AD group we labelled CSF as a reason for additional amyloid- β PET when a normal CSF analysis or a CSF analysis with isolated low A β_{42} or high tau/p-tau did not support the clinical diagnosis. Similarly, we labelled MRI findings as a reason for additional amyloid- β PET in the non-AD group when a normal MRI or pronounced hippocampal atrophy decreased confidence in the current clinical diagnosis.

Accordance to amyloid- β PET appropriate use criteria

Previously published appropriate use criteria support amyloid- β imaging in case of 1) progressive unexplained MCI, 2) possible AD with atypical or etiologically mixed presentation, and 3) progressive dementia at an early age, usually defined as below the age of 65 [14]. Based on examining patient charts, we determined for each case accordance with the PET appropriate use criteria.

Patient population

For all patients, we determined an initial available etiological diagnosis as the first diagnosis, the first available diagnosis after amyloid- β PET as the last diagnosis, and diagnostic change as the difference between the two. In $n = 23$ (32%) cases, CSF analysis was performed prior to referral to our center ($n = 14$ with A β +/ τ - or A β -/ τ + based on CSF) and $n = 3$ (4%) patients had undergone a previous amyloid- β PET scan.

We present our patient population by the binarized status based on CSF A β_{42} and total tau. We chose to use CSF total tau instead of p-tau in order to closely resemble clinical decision-making. Our data showed that in case of dubious diagnosis, increased levels of either CSF total tau or p-tau facilitated further diagnostics and there were more patients with an isolated increase of CSF tau ($n = 8$) than p-tau ($n = 3$). CSF tau and p-tau status were identical in $n = 61$ (85%) of patients.

Statistical analysis

Statistical analysis was performed using R software (Version 3.4.4) [26–29]. We compared patient features using Chi-squared tests, two samples t -tests, Wilcoxon Rank-Sum tests and linear regression models. Cognitive scores were compared while adjusting for age, sex, and education.

RESULTS

Demographics

We included $n = 5$ (7%) patients with subjective cognitive decline (SCD), $n = 3$ (4%) whose symptoms were mainly associated with a psychiatric condition, $n = 16$ (22%) with MCI, and $n = 48$ (67%) with dementia. The average age in our patient cohort was 62.0 (standard deviation [SD] = 8.1), $n = 46$ (64%) of patients were male and average MMSE was 23.6 (SD = 3.8) (Table 1). Most patients where a clinical PIB-scan was requested were A β -/ τ - ($n = 25$, 35%), A β -/ τ + ($n = 23$, 32%), or A β +/ τ - ($n = 19$, 26%), while only a minority 5 (7%) had an A β +/ τ + status based on CSF (Fig. 2). In total, $n = 34$ (47%) patients were amyloid-positive based on PET (compared to $n = 24$ (33%) based on CSF A β_{42}). Amyloid- β CSF and PET status were discordant in $n = 32$ (44%) cases. Amyloid- β PET positivity was lower in the A β -/ τ - group ($n = 6$, 24%) compared to the A β -/ τ + ($n = 15$, 65%; $p = 0.01$) and A β +/ τ - ($n = 11$, 58%; $p = 0.048$) groups. We found no significant differences in cognitive scores and MRI measures between the groups.

Reasons for amyloid- β PET after CSF

To explore the clinical reasoning for amyloid- β PET, patients were divided into two groups (AD, $n = 41$ and non-AD, $n = 31$) based on the most likely etiological diagnosis prior to performing a PIB PET scan. More than one reason for amyloid- β PET was reported for $n = 33$ (46%) cases. Conflicting information from CSF analysis (either not supporting the current diagnosis or with discordant A β /tau status) was the most frequent reason for requesting an amyloid- β PET scan ($n = 53/72$; 74%), being more prevalent in patients with the main suspected etiological diagnosis of AD ($n = 36/41$, 88%) than in patients with a non-AD suspected etiological diagnosis ($n = 17/31$, 55%, $p = 0.004$) (Fig. 3). Other factors contributing to the request of a clinical amyloid- β PET scan after CSF included MRI not supporting the clinical diagnosis ($n = 16/72$, 22%), unusual clinical presentation ($n = 11/72$, 15%) and young age ($n = 8/72$, 11%). In some cases ($n = 5/72$, 7%, all with A β +/ τ -), inexperience in interpreting the results of a new CSF A β_{42} assay (Elecsys CSF, Roche Diagnostics GmbH) contributed to diagnostic uncertainty leading to PIB PET scan. A reason for requesting a

Table 1
Patient population stratified by binarized CSF Aβ₄₂ and tau status

	TOTAL	Normal CSF Aβ ⁻ /tau ⁻	Conflicting CSF Aβ and tau		AD-like CSF Aβ ⁺ /tau ⁺
			Aβ ⁻ /tau ⁺	Aβ ⁺ /tau ⁻	
n (%)	72	25 (35)	23 (32)	19 (26%)	5 (7)
Sex, male (%)	46 (64)	17 (68)	14 (61)	12 (63)	3 (60)
Age (mean (SD))	62.0 (8.1)	63.9 (6.3)	62.9 (9.5)	60.5 (6.4)	54.5 (11.2)
Education (median [IQR])	5 [5, 6]	5 [5, 6]	5 [5, 6]	5 [5, 6]	5 [4, 6]
APOE ε4 carriership (%)	33 (55)	14 (64)	11 (55)	8 (50)	0/2 (0)
Change in diagnosis (%)	37 (51)	20 (80)^{B,C,D}	11 (48)^A	5 (26)^A	1 (20)^A
CSF as a reason for amyloid-β PET (%)	53 (74)	15 (60)	18 (78)	17 (89)	3 (60)
Amyloid-β PET positivity (%)	34 (47)	6 (24)^{B,C}	15 (65)^A	11 (58)^A	2 (40)
PET according to AUC (%)	61 (85)	22 (88)	19 (83)	15 (79)	5 (100)
CSF-PET time difference, days (median [IQR])	140 [67, 260]	140 [75, 261]	162 [78, 304]	124 [61, 204]	109 [34, 260]
MRI as a reason for amyloid-β PET (%)	16 (22)	6 (24)	3 (13)	5 (26)	2 (40)
MTA positivity (%)	32 (46)	14 (56)	8 (40)	8 (42)	2 (40)
PCA positivity (%)	26 (54)	10 (59)	8 (67)	7 (47)	1 (25)
GCA positivity (%)	22 (46)	8 (47)	5 (42)	8 (53)	1 (25)
Fazekas positivity (%)	9 (13)	3 (12)	5 (23)	1 (5)	0 (0)
Lacune positivity (%)	4 (6)	1 (4)	1 (5)	2 (11)	0 (0)
Microbleed positivity (%)	5 (8)	0 (0)	3 (14)	2 (11)	0 (0)
MMSE (mean (SD))	23.6 (3.8)	22.8 (3.6)	23.9 (4.1)	24.4 (3.9)	23.7 (2.5)
Memory z-score (mean (SD))	-3.20 (2.87)	-3.41 (2.22)	-3.19 (4.04)	-2.98 (2.30)	-2.74 (1.14)
Language z-score (mean (SD))	-1.13 (1.39)	-1.30 (1.84)	-1.09 (1.27)	-0.87 (0.79)	-1.81 (1.47)
Attention z-score (mean (SD))	-1.10 (1.09)	-1.25 (1.01)	-0.95 (1.11)	-0.92 (1.14)	-2.57 (0.08)
Executive z-score (mean (SD))	-1.56 (1.41)	-1.82 (1.32)	-1.61 (1.69)	-1.05 (1.13)	-2.07 (1.17)
Visuospatial z-score (mean (SD))	-0.90 (1.42)	-1.02 (1.98)	-0.75 (1.00)	-0.77 (0.78)	-2.07 (1.00)

Education is staged by Verhage classification (1–7) [32]. MRI scans were regarded 1) medial temporal atrophy (MTA)-positive if the left-right averaged MTA ≥1 for a patient under the age of 65, or ≥1.5 for patient age between 65 and 75, 2) posterior cortical atrophy (PCA)-positive if the left-right averaged PCA ≥1 for a patient under the age of 65, and 3) global cortical atrophy (GCA)-positive if the GCA ≥1 for a patient under the age of 65 [23]. Cognitive domain z-scores were derived using the mean and standard deviation values from a group of healthy controls. A, B, C, D indicate difference (*p* < 0.05) from other groups: A) difference from Aβ⁻/tau⁻; B) difference from Aβ⁻/tau⁺; C) difference from Aβ⁺/tau⁻; D) difference from Aβ⁺/tau⁺.

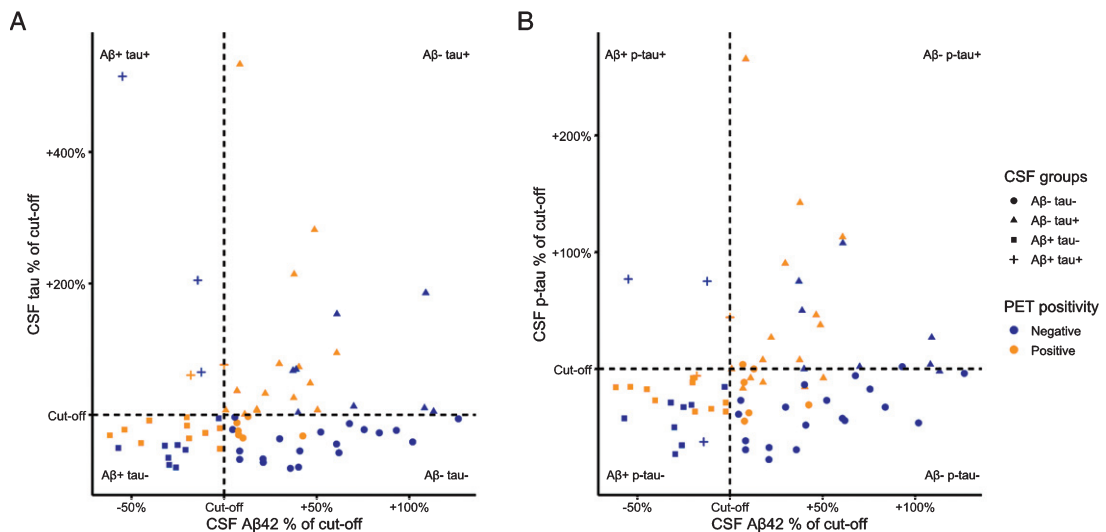


Fig. 2. CSF Aβ₄₂, and tau/p-tau values relative to their cut-offs. We present standardized CSF values, created by calculating the percentage of the CSF value relative to its concurrent cut-off. Values of <100% represent pathologically decreased CSF Aβ₄₂ and values of >100% indicate pathologically increased CSF tau (A) and p-tau (B).

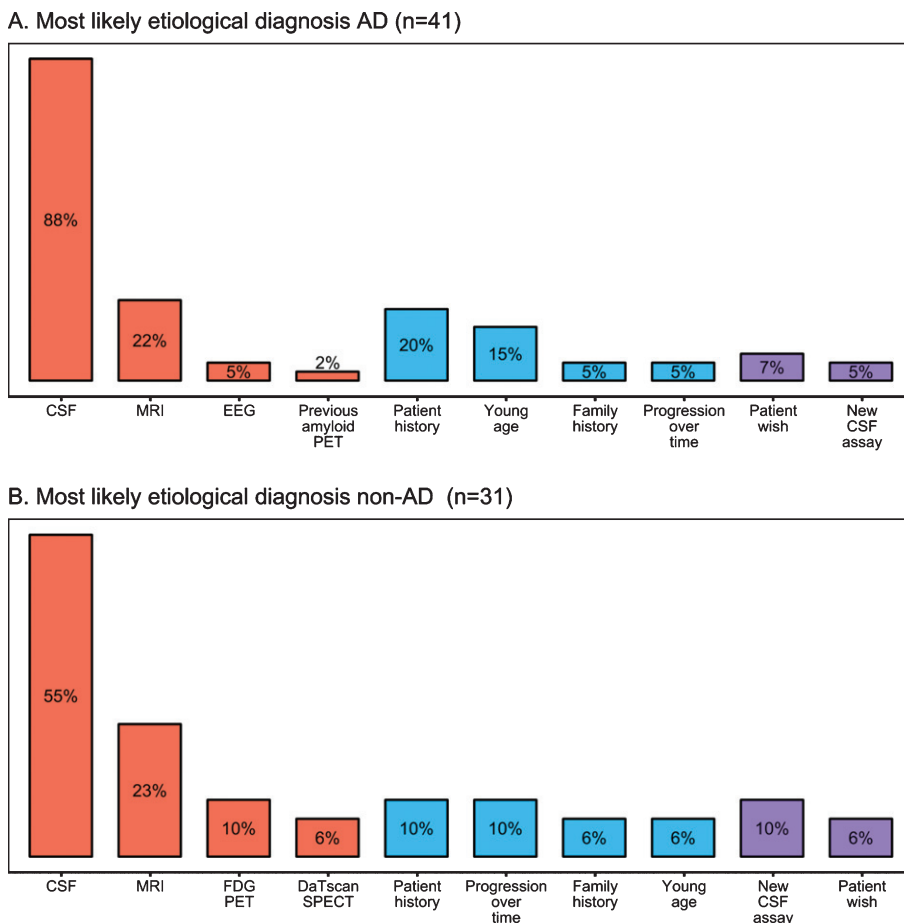


Fig. 3. Clinical reasons for requesting additional amyloid- β PET after CSF. Patients are grouped based on most likely diagnosis prior to an amyloid- β PET scan. For both diagnosis groups we list characteristics that were recorded as being not compatible with the current main diagnosis, therefore leading to additional amyloid- β PET imaging. Reasons for the amyloid- β PET scan are grouped as biomarkers (red), patient history and presentation (blue), and external (purple). More than one reason is possible per patient. CSF, cerebrospinal fluid; MRI, magnetic resonance imaging; EEG, electroencephalography; PET, positron emission tomography; FDG, [18 F]fluorodeoxyglucose; SPECT, single-photon emission computed tomography.

PET scan was not recorded in the patient chart for $n = 2/72$ (3%) cases.

Accordance to amyloid- β PET AUC

In most cases ($n = 61$, 85%), amyloid- β PET scans were performed in compliance with the AUC. Our clinical practice was not covered by the AUC in $n = 7$ ($n = 3$ amyloid-negative based on PET) as the clinical findings were suspicious of amnesic AD, but conflicting information from CSF ($n = 6$) or a previously negative amyloid- β PET combined with normal CSF ($n = 1$) led to an amyloid- β PET scan. Finally, an amyloid- β PET scan was requested for $n = 3$ patients without objective cognitive decline, who had decreased A β_{42} in the CSF analysis, lower-

ing diagnostic confidence; and for $n = 1$ patient with a known PSEN1 mutation to define the stage of pathological disease progression.

Change in diagnosis

Of the $n = 42$ patients with AD as their initial etiological diagnosis, $n = 15$ (36%) had a predominant non-amnesic presentation (either language-AD ($n = 6$), visuospatial ($n = 5$), behavioral/dysexecutive ($n = 3$), or corticobasal syndrome ($n = 1$)) (Fig. 4). Likewise, in patients with a final diagnosis of AD, about half ($n = 15/32$, 44%) had a non-amnesic presentation. Twenty-two patients (52%) with an initial clinical diagnosis of AD had amyloid- β positivity based on PET and $n = 11$ (26%) based on CSF A β_{42} .

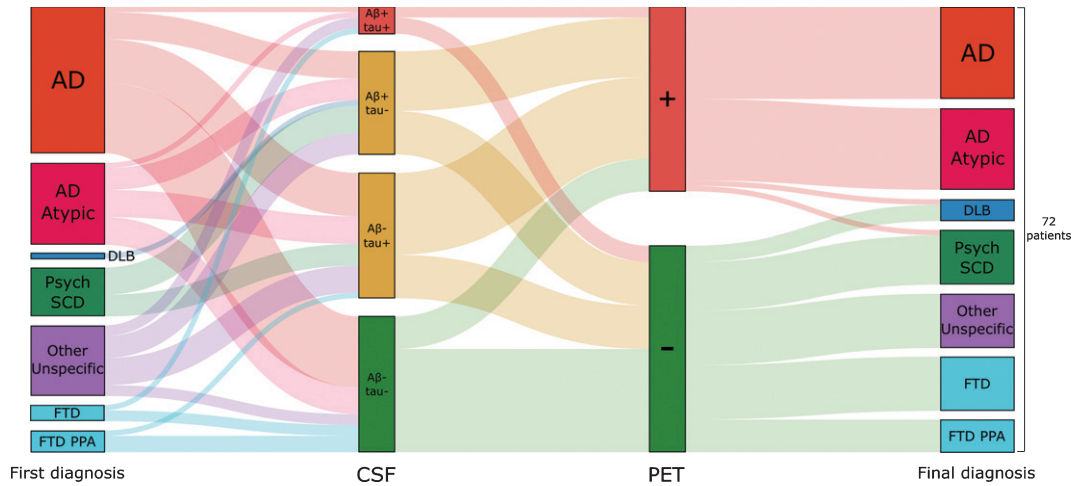


Fig. 4. Etiological diagnosis in relation to CSF A β /tau status and amyloid- β PET. A Sankey diagram showing 1) the distribution of baseline diagnoses to groups based on CSF A β /tau status, 2) the percentage of amyloid- β PET positivity by CSF A β /tau groups, and 3) the correlation of final diagnosis to amyloid- β PET positivity. DLB, dementia with Lewy bodies; Psych, psychiatric disorder; SCD, subjective cognitive decline; FTD, frontotemporal dementia; PPA, primary progressive aphasia.

Of the patients with AD as the final etiological diagnosis, $n = 32$ (100%) had amyloid- β pathology based on PET and 13 (41%) based on CSF A β_{42} .

Overall, change in diagnosis occurred in $n = 37$ (51%) of cases. Diagnosis changed more often in the A β -/tau- group ($n = 20$, 80%) compared to the A β -/tau+ ($n = 11$, 48%; $p = 0.04$), A β +/tau- ($n = 5$, 26%; $p < 0.01$), and A β +/tau+ ($n = 1$, 20%; $p = 0.03$) groups.

Final diagnoses of the five cases in the CSF A β +/tau+ group were 1) early-onset AD (age: 48 years) with a negative family history, 2) autoimmune encephalitis, 3) corticobasal syndrome due to AD, 4) logopenic variant primary progressive aphasia with a previously negative amyloid- β PET scan, and 5) a suspected genetic variant of frontotemporal dementia. To further illustrate the diagnostic process, we present a small case series including one patient from each of the four CSF A β /tau groups (Fig. 5).

DISCUSSION

We investigated the clinical reasoning behind requesting an amyloid- β PET scan after disclosure of CSF biomarkers in a clinical cohort. Our main finding was that in most cases CSF biomarkers conflicting with the clinical diagnosis contributed to diagnostic uncertainty, which led to the request of an amyloid- β PET scan to support the clinical diagnostic process. This was reinforced by the observation that an

additional amyloid- β PET scan was rarely requested in patients with a CSF A β +/tau+ status. Second, we found that an amyloid- β PET was requested in patients, that were relatively young, often had an atypical presentation of AD and often showed a change in diagnosis. Third, we observed that although amyloid- β PET scans were usually requested according to the AUC, our clinical practice was not wholly covered by these criteria, as it was often driven by inconclusive CSF biomarker results. Our results support previous work that CSF biomarkers that conflict with the clinical diagnosis often lead to additional amyloid- β PET [12, 13].

In our cohort, an amyloid- β PET was most often requested due to inconclusive results from the CSF biomarkers. This occurred in cases when diagnostic confidence was low due to inconclusive CSF A β /tau status, or when a clinical diagnosis of AD was contradicted by a non-pathologic CSF analysis, which is largely in agreement with previous findings [13]. In fact, there were few patients with CSF A β +/tau+ status in our cohort, indicating that patients with a clinical suspicion of AD supported by low A β_{42} and high tau in the CSF analysis usually do not need further confirmation with amyloid- β PET. This is also evident in the current guidelines for both clinical practice [8] and research [1], which advocate CSF and PET as parallel options to support the diagnosis of AD. However, our results show that in complicated cases clinicians valued the information from an additional amyloid- β PET.

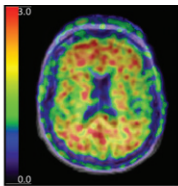
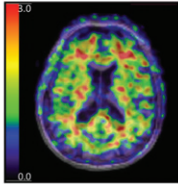
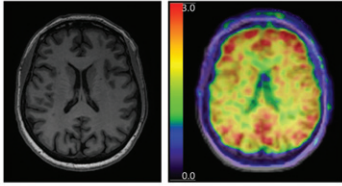
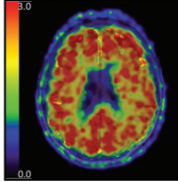
First Diagnosis	CSF (% of cut-off)	Clinical history & Rationale for amyloid- β PET	Amyloid- β PET scan	Last Diagnosis
AD	<p>N N N</p> <p>143% 68% 69%</p> <p>Aβ₄₂ tau p-tau</p>	<ul style="list-style-type: none"> - 72 y/o woman with MCI / beginning dementia (memory, behaviour) - Family history: five siblings and father with dementia - Clinically suspicious for familial AD, but conflicting CSF leads to PET - Clinical rationale not covered by the amyloid-β PET AUC <p><i>Image: [11C]PIB PET scan showing diffuse uptake in grey matter, in accordance with a positive scan.</i></p>		AD
AD	<p>N ↑ ↑</p> <p>122% 133% 127%</p> <p>Aβ₄₂ tau p-tau</p>	<ul style="list-style-type: none"> - 72 y/o man with progressive memory loss clinically fitting AD - MRI showed evident hippocampal atrophy with MTA grade 3/3, and parietal atrophy (PCA grade 2/2; not shown) - Conflicting CSF with normal Aβ₄₂ led to PIB PET (negative) - Final diagnosis was an atypical presentation of frontotemporal dementia - Clinical rationale not covered by the PET AUC <p><i>Image: [11C]PIB PET showing no evident tracer uptake in grey matter, in accordance with a negative scan.</i></p>		FTD
AD	<p>↓ N N</p> <p>90% 73% 66%</p> <p>Aβ₄₂ tau p-tau</p>	<ul style="list-style-type: none"> - 54 y/o woman with progressive memory loss - Clinically MCI most probably due to AD - CSF analysis was performed externally 2 months prior to referral - Conflicting CSF with normal tau values, young age, and a normal MRI scan led to requesting additional PIB PET <p><i>Left: MRI T1 sequence, showing no evident atrophy; Right: [11C]PIB PET scan showing diffuse uptake in grey matter, in accordance with a positive scan.</i></p>		AD
AD atypical	<p>↓ ↑ ↑</p> <p>100% 176% 144%</p> <p>Aβ₄₂ tau p-tau</p>	<ul style="list-style-type: none"> - 60 y/o man with dementia - Myoclonus, a possible alien limb syndrome and neglect on the left side - Patient was diagnosed with corticobasal syndrome most likely due to AD - Previously, DLB had been suspected due to hallucinations and fluctuations during an infection; Datscan was performed and was rated as normal (not shown) - A marginally decreased CSF Aβ₄₂ and unclear clinical findings led to requesting additional amyloid-β PET <p><i>Image: [11C]PIB PET scan showing diffuse uptake in grey matter, in accordance with a positive scan.</i></p>		AD atypical

Fig. 5. Four case reports illustrating the clinical reasoning for requesting an additional amyloid- β PET.

Although clinical diagnosis conflicting with CSF biomarkers contributed to requesting amyloid- β PET scans in most patients, overall clinical rationale was more complex. This was illustrated by a variety of other factors, often in combination with each other, that decreased clinical diagnostic confidence and led to additional amyloid- β PET imaging. Most prominently, incongruent imaging (MRI, FDG PET, DaTscan SPECT) findings or an unusual clinical presentation contributed to decreased confidence in the clinical diagnosis despite CSF findings. The added value of amyloid- β PET imaging, in particular in atypical clinical presentations of AD, has also been shown previously [12, 30]. Our results also support younger patient age being a factor for requesting an amyloid- β PET scan, as indicated in the PET AUC [14]. This is related to younger patients more often having a non-amnesic clinical presentation, in addition to the diagnosis of AD being rare and potentially having a higher impact at a younger age. Finally, we observed that a clinician's decision can also be influ-

enced by external reasons, such as patient wish or decreased confidence in CSF results due to the initiation of a new CSF assay. It is also possible that clinicians as well as patients might also be inclined to have more confidence in PET imaging due to the visual aspects of a PET scan, and that clinicians' biases and prior experiences might play a role when deciding whether to use additional amyloid- β PET diagnostics.

Amyloid- β PET was usually, but not always, requested in accordance with the PET appropriate use criteria [14]. Some differences between clinical practice and the AUC were not unexpected as the AUC were designed to build an initial framework for clinical amyloid- β PET, and were also published during the time course of our study. When clinical practice was not covered by the AUC, a PIB scan was requested in patients with no objective cognitive decline due to decreased A β ₄₂ values in the CSF, or due to decreased diagnostic confidence arising from inconclusive or normal CSF biomarker (or prior

amyloid- β PET) results in patients with a clinical syndrome suggestive of AD. Although the recently published AUC for CSF also include performing CSF analysis in SCD [31], neither the AUC for CSF nor the AUC for PET describe the diagnostic setting, where information about amyloid- β status is already available. As amyloid- β biomarkers are increasingly integrated into clinical practice, the number of such cases is likely to increase over time. The value of an additional amyloid- β PET is likely highest in patients with a CSF analysis conflicting with the clinical diagnosis of AD, as a negative amyloid- β PET scan can refute the diagnosis [11]. In cases with a non-AD diagnosis combined with a decreased A β ₄₂ and/or an increased tau in the CSF, the added value of an amyloid- β PET scan is hindered by the possibility of amyloid- β as a secondary pathology, especially in older populations. Therefore, our results combined with previous work from other centers [12, 13] suggest a group of patients (i.e., clinically diagnosed with AD without an AD-like CSF biomarker signature) might benefit from being included in updated amyloid PET AUC. However, these findings must be confirmed by larger prospective multi-center studies.

The main strength of the present study is the description of the clinical practice in a tertiary memory clinic, where both CSF biomarkers are regularly used for clinical practice and there is good access to amyloid- β PET if needed. By excluding cases where an amyloid- β PET scan was performed due to involvement in research, we were able to minimize the bias caused by research and to concentrate on the clinical decision-making process. In addition, our study has some limitations. Composition of our sample resulted in some inherent biases, caused by the infrastructure of our memory clinic. For example, all patients in our center are offered CSF biomarkers analysis, many patients (often with prior CSF analysis available) are referred to us due to a diagnostic dilemma, and referred patients are generally relatively young. Additionally, all patients are assessed by five neurologists in our center, who might share similar views on the application of biomarkers. While these sample characteristics might reduce overall generalizability, we believe our findings are likely to be generalizable to other memory clinic settings where CSF analysis is commonly used and represent a relevant clinical question. Additionally, due to the retrospective nature of the study, some of the data were retrieved from patient charts. In cases of incomplete or ambiguous descriptions, some degree of subjective judgement on the part of the investigators was

unavoidable. Our center has also been involved in several amyloid- β PET studies [9–11], which recruited patients from clinical practice. Therefore, our cohort may have missed cases where amyloid- β PET was deemed clinically useful from that period of time. Finally, due to the longitudinal upward drift of the median CSF A β ₄₂ values in our centers, the cut-off values for CSF A β ₄₂ in our center have been changing over time [17]. Therefore, it is feasible that the CSF A β ₄₂ cut-offs did not always best represent the underlying amyloid- β status.

To conclude, we presented data from a single memory clinic where CSF biomarkers are commonly used. During the period of our study, the main reason for requesting an amyloid- β PET scan was the occurrence of a mismatch between the primary clinical diagnosis and CSF A β /tau results. Future work is necessary to confirm similar clinical reasoning in other cohorts and to consider whether such practice should be represented in guidelines.

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