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Neuropsychological profile of amyloid-positive versus amyloid-negative amnestic Mild Cognitive Impairment

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Introduction. Patients diagnosed with amnestic mild cognitive impairment (aMCI) are at high risk of progressing to dementia. It became possible, through the use of biomarkers, to diagnose those patients with aMCI who have Alzheimer's disease. However, it is presently unfeasible that all patients undergo biomarker testing. Since neuropsychological testing is required to make a formal diagnosis of aMCI, it would be interesting if it could be used to predict the amyloid status of patients with aMCI.

Methods. Participants with aMCI, known amyloid status (Aβ+ or Aβ−) and a comprehensive neuropsychological evaluation, were selected from the Cognitive Complaints Cohort database for this study. Neuropsychological tests were compared in Aβ+ and Aβ− aMCI patients. A binary logistic regression analysis was conducted to model the probability of being amyloid positive.

Results. Of the 216 aMCI patients studied, 117 were Aβ+ and 99 were Aβ−. Aβ+ aMCI patients performed worse on several memory tests, namely Word Total Recall, Logical Memory Immediate and Delayed Free Recall, and Verbal Paired Associate Learning, as well as on Trail Making Test B, an executive function test. In a binary logistic regression model, only Logical Memory Delayed Free Recall retained significance, so that for each additional score point in this test, the probability of being amyloid positive decreased by 30.6%. The resulting model correctly classified 64.6% of the aMCI cases regarding their amyloid status.

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Conclusions. The neuropsychological assessment remains an essential step to diagnose and characterize patients with aMCI; however, neuropsychological tests have limited value to distinguish the aMCI patients who have amyloid pathology from those who might suffer from other clinical conditions.

As a consequence of the ageing of the population, the number of people affected by neurodegenerative disorders, particularly Alzheimer disease (AD), is increasing dramatically worldwide (Prince, 2015). There has been a growing interest in detecting AD as soon as possible along its insidious evolution, before the establishment of the diagnosis of dementia. The correct identification of patients with memory complaints who already have an ongoing neurodegenerative process is desirable, since it offers patients the possibility to make important life decisions, anticipate future care, start symptomatic drugs, initiate cognitive rehabilitation therapy, and eventually participate in clinical trials with putative neuroprotective drugs (de Mendonça, 2012). About 2 decades ago, the Mayo Clinic group fostered an important advance by proposing the concept of amnestic mild cognitive impairment (aMCI), as a condition characterized by subjective memory complaints, objective memory deficit, normal general cognitive performance, and maintained activities of daily living (Petersen *et al.*, 1999). Patients diagnosed with aMCI in a clinical setting have about 10% annual progression rate of conversion to dementia, usually AD (Mitchell & Shiri-Feshki, 2009). However, aMCI can have other aetiologies (Hanfelt, Peng, Goldstein, & Lah, 2018), and some aMCI patients actually remain stable for as long as a decade (Alves *et al.*, 2018).

In recent years, the use of biomarkers has allowed the possibility of diagnosing AD *in vivo* in patients who present with aMCI. These biomarkers are surrogates of pathological alterations in the brain characteristic of AD (Jack *et al.*, 2018). The presence of amyloid pathology may be determined by measuring amyloid Aβ1–42 concentrations in the cerebrospinal fluid (CSF) and/or quantifying brain deposits of Aβ with amyloid positron emission tomography (PET) (Bocchetta *et al.*, 2015).

In spite of the remarkable advance that the development of biomarkers represents both from an investigational and a clinical perspective and the rapid acceptance of these methods by reference centres (Bocchetta *et al.*, 2015), the generalization of biomarker testing to other settings has been more sluggish. Several explanations might be advanced; for instance, lumbar puncture, used to obtain CSF, is an invasive procedure with contra-indications and side effects, and amyloid PET is quite expensive and not widely available. Bearing this in mind, it would be important to discover non-invasive and affordable methods that could discriminate between amyloid-positive (Aβ+) and amyloid-negative (Aβ−) aMCI patients.

Since neuropsychological testing is not invasive and is required to make a formal diagnosis of aMCI, it would be very interesting if it could be used to identify the amyloid status in patients with aMCI (Bahar-Fuchs *et al.*, 2013). In other words, Aβ+ aMCI patients might have a particular neuropsychological profile that would distinguish them from Aβ− aMCI patients. Several studies compared global cognition, attention, executive functions, visuospatial functions, language, visual memory, and verbal memory between Aβ+ and Aβ− aMCI patients.

Since patients with AD typically have deficits in episodic memory as a consequence of early and marked hippocampal neurodegeneration, it is not surprising that Aβ+ aMCI patients consistently presented more prominent episodic memory deficits than Aβ− aMCI patients in several different studies (Bahar-Fuchs *et al.*, 2013; Huijbers *et al.*,

2015; Kandel, Avants, Gee, Arnold, & Wolk, 2015; Kim *et al*., 2018; Reijs *et al*., 2017; Tomadesso *et al*., 2018, 2019; Wolk *et al*., 2009). However, regarding attention and executive functions, different studies produced less consistent results, possibly depending on the kind of test used to measure these abilities as well as the number of patients recruited. In the Alzheimer's Disease Neuroimaging Initiative (ADNI) cohort, Aβ+ aMCI patients took longer to complete the Trail Making Tests A and B, when compared to Aβ− aMCI patients (Kandel *et al*., 2015). These results were not corroborated by other studies, that did not observe significant differences in the Trail Making Tests A and B between Aβ+ and Aβ− aMCI patients (Tomadesso *et al*., 2018, 2019; Wolk *et al*., 2009). Regarding another commonly used executive test, Verbal Semantic Fluency, Aβ+ aMCI patients had worse performance in one study (Kandel *et al*., 2015) but not in other work (Wolk *et al*., 2009).

We now reappraise neuropsychological testing in Aβ+ and Aβ− aMCI patients, particularly concerning performances on executive tests, as well as cognitive domains so far scarcely analysed, like abstract reasoning and calculation. Furthermore, we aim to test whether a statistical model involving different neuropsychological variables could be valuable to help identify the amyloid status of patients with aMCI.

Materials and methods

Participants

Participants belong to the Cognitive Complaints Cohort (CCC). The CCC was established in a prospective study to evaluate the cognitive evolution of patients with cognitive complaints and no dementia, based on a comprehensive neuropsychological evaluation and other biomarkers. Detailed information concerning CCC establishment was provided in a previous publication (Marôco *et al*., 2011). The study was approved by the local ethics committee and conducted according to the declaration of Helsinki. Informed consent was obtained from patients before any procedure.

Inclusion criteria

1. Diagnosis of amnestic MCI (aMCI). The criteria for the diagnosis of aMCI were adapted from Petersen *et al*. (1999):

   a. Presence of memory complaints;
   b. Abnormal memory function, documented by impairment in the Logical Memory A test Immediate Free Recall score. Logical Memory is a subtest of the Bateria de Lisboa para Avaliação das Demências (BLAD) (Garcia, 1984; Guerreiro, 1998) (see below). For the memory function to be considered abnormal, we set the cut-off score of the Logical Memory A Immediate Free Recall at 1 *SD* below the age and education norms. Busse, Hensel, Gühne, Angermeyer, and Riedel-Heller (2006) observed, in the cohort of the Leipzig Longitudinal Study of the Aged, that the 'MCI modified, 1.0 *SD*' criteria had the highest relative predictive power for the development of dementia;
   c. Normal general cognitive function, determined by the Mini-Mental State Examination (MMSE) (Folstein, Folstein, & McHugh, 1975) (see below) within normal values for the Portuguese population (Guerreiro, 1998);
   d. No or a minimal impairment in activities of daily living, determined by the Instrumental Activities of Daily Living Scale (IADL) (Lawton & Brody, 1969)

(see below), that is to say, no more than one item from the IADL scale was altered.
2. Known amyloid status, determined by CSF Aβ1–42 measurement and/or cortical uptake of the Pittsburgh compound B (^{11}C-PiB) on the PET scan.

Exclusion criteria

1. Presence of neurological (stroke, brain tumour, significant head trauma, epilepsy) or psychiatric disorders that may induce cognitive deficits; patients with major depression according to DSM-IV-TR (APA, 2000) or serious depressive symptoms, indicated by a score >20 in Geriatric Depression Scale (GDS30) or >10 in Geriatric Depression Scale short version (GDS15) (Barreto, Leuschner, Santos, & Sobral, 2008; Yesavage *et al.*, 1982; Yesavage & Sheikh, 1986) (see below);
2. Presence of systemic illness with cerebral impact (uncontrolled hypertension, metabolic, endocrine, toxic, and infectious diseases);
3. History of alcohol abuse or recurrent substance abuse or dependence;
4. Medication use with possible cognitive side effects;
5. Seriously reduced vision or other sensory deficits likely to interfere with assessment;
6. Presence of dementia according to DSM-IV-TR (APA, 2000);
7. Interval between neuropsychological assessment and knowledge of amyloid status longer than 12 months.

The diagnosis of aMCI was made by an experienced neurologist, after multidisciplinary consensus using all available clinical, neuropsychological, and neuroimaging information available from the diagnostic workup.

Biomarker analysis

The amyloid biomarker status was based on CSF Aβ1–42 level and/or cortical uptake on ^{11}C-PiB PET, and the aMCI patients were classified as Aβ+ or Aβ−. Both sources of amyloid status were considered interchangeable since a high agreement between Aβ1–42 concentrations in the CSF and amyloid PET scan results in aMCI and AD disease patients was confirmed by previous studies (Leuzy *et al.*, 2016).

The levels of Aβ1–42 were measured using commercially available enzyme-linked immunosorbent assays (INNOTEST® β-amyloid (1–42); Innogenetics, Ghent, Belgium) according to the established protocols on participating centres (Teunissen, Tumani, Engelborghs, & Mollenhauer, 2014). The levels of Aβ1–40 and the ratio Aβ1–42 over Aβ1–40 were not determined routinely, only in exceptional cases where a discrepancy was found between CSF and PET scan amyloid results. The expected site assay variability present in multicentre studies was acknowledged (Mattsson *et al.*, 2009), and positivity was determined using locally available cut-off values.

The cortical uptake with ^{11}C-PiB PET was performed only in one centre using the same scanner (Philips PET/CT Gemini GXL, Philips Portuguesa, Porto Salvo, Portugal), preceded by a low-dose brain computed tomography (CT) acquisition for attenuation correction (Institute of Nuclear Science Applied to Health, ICNAS, University of Coimbra). ^{11}C-PiB PET images were classified as amyloid positive or negative based on a support vector machines local classifier, which uses the voxelwise brain grey matter standardized uptake value ratio and the cerebellar grey matter as reference region (Oliveira *et al.*, 2018).

Neuropsychological assessment

The comprehensive neuropsychological assessment was carried out by the same team of trained neuropsychologists, following a standard protocol and comprised the following instruments and scales:

1. Mini-Mental State Examination (Folstein *et al.*, 1975; Guerreiro, 1998) – the MMSE is a brief screening instrument to assess global cognitive performance. The Portuguese version was applied, and normative data were >27 for individuals with more than 11 years of education and >22 for patients with 11 or less years of education (Guerreiro, 1998).
2. Battery of Lisbon for the Assessment of Dementia (Garcia, 1984; Guerreiro, 1998) – the BLAD is a comprehensive neuropsychological battery that includes some tests from the Wechsler Memory Scale (WMS; Wechsler, 1969) and has been validated for the Portuguese population. This battery includes tests for the following cognitive domains: immediate memory (Digit Span forward); verbal memory (Word Total Recall, a five words 1-min delayed recall test, in which the total score contemplates spontaneous and cued recall); logical memory (Logical Memory Immediate and Delayed Recall; for this test, the score is based on the combination of 7 literal elements and 17 meaningful elements); associate learning (Verbal Paired Associate Learning); general information (General Information, consisting of 20 questions on subjects of general knowledge); working memory (Digit Span backward); attention (Cancellation Task); verbal initiative (Verbal Semantic Fluency); verbal and non-verbal abstraction (Raven's Coloured Progressive Matrices – Ab series-B and Interpretation of Proverbs); and calculation (Basic Written Calculation);
3. Trail Making Test (part A and part B; Cavaco *et al.*, 2013; Reitan, 1958) – the TMT task measures sustained attention, visuomotor processing speed (part A), visuospatial working memory, and cognitive flexibility (part B). The part A consists of 25 circles numbered 1–25 distributed over a sheet of paper, and the patient should draw lines to connect the numbers in ascending order. In part B, there are 25 circles as well, but the circles include both numbers (1–13) and letters (A–M), and the patient has to draw lines to connect them all in an ascending pattern with the added task of alternating between the numbers and letters (i.e., 1-A-2-B-3-C).
4. Geriatric Depression Rating Scale (GDS; Barreto *et al.*, 2008; Yesavage & Sheikh, 1986; Yesavage *et al.*, 1982) – the GDS is a self-report instrument used specifically to identify depressive symptomatology in the elderly. For this study, the Portuguese versions of GDS30 and GDS15 were used (Barreto *et al.*, 2008).
5. Blessed Dementia Rating Scale is a clinical rating scale with 22 items that measures changes in performance of everyday activities (8 items), self-care habits (3 items), and changes in personality, interests, and drives (11 items). Ratings are based on information from relatives or friends and concern behaviour over the preceding 6 months.

For the present work, the neuropsychological assessment closest to the knowledge of the amyloid status was used.

Statistical analysis

For comparison of demographic and clinical data between groups, the independent samples two-tailed Student's *t*-test and the chi-squared Pearson test were used, for

numerical and nominal data, respectively. The neuropsychological assessments were standardized according to the age and education norms for the Portuguese population (Garcia, 1984; Guerreiro, 1998), and *z* scores were calculated with the equation [$z = (x\text{-mean})/SD$]. The comparison of neuropsychological results between Aβ+ and Aβ− groups was done with the independent samples two-tailed Student's *t*-test. To check whether the differences that were found between groups still held when controlling for the MMSE score, a general linear model analysis was performed considering the MMSE as a covariate. A binary logistic regression analysis was conducted to assess whether the neuropsychological tests scores could predict amyloid positivity. The tests that were significantly different between the groups entered the model. The Enter method (i.e., standard regression analysis) was used. Receiver operating characteristic (ROC) curves were obtained when appropriate. In order to control for an eventual redundancy in the tests comprising the neuropsychological battery, a principal component analysis using a rotated varimax component matrix was performed.

Statistical analyses were performed using SPSS Statistics for Windows, version 17.0 (SPSS Inc., Chicago, IL, USA) package. A probability value of $<.05$ was assumed to be statistically significant.

Results

A total of 216 patients with aMCI were enrolled from the CCC for the present study, of whom 117 were Aβ+ and 99 were Aβ−. The two groups did not differ in terms of gender, education, age of first symptoms, and time between symptoms onset and neuropsychological assessment. They did not differ in terms of the presence of depressive symptoms either. Regarding the Blessed Dementia Rating Scale scores, aMCI patients in the two groups had similar global levels of severity (Table 1).

Neuropsychological evaluation (Table 2) showed that Aβ+ aMCI patients had lower MMSE scores than Aβ− aMCI patients. MMSE values for Aβ+ aMCI patients were 26.8 (*SD* 2.2, skewness −0.3, range 23–30) and for Aβ− aMCI patients 27.6 (*SD* 2.0, skewness −0.7, range 23–30).

Aβ+ aMCI patients also performed worse on several memory tests, namely the Word Total Recall, Logical Memory Immediate and Delayed Free Recall, and Verbal Paired Associate Learning, as compared to Aβ− aMCI patients. To check whether the differences

Table 1. Demographic and clinical characterization

	Aβ+ aMCI ($n = 117$)	Aβ− aMCI ($n = 99$)	*p* value
Gender, male/female, *n* (% female)	53/64 (54.7%)	41/58 (58%)	.676[a]
Education, years, mean (*SD*)	10.6 (4.6)	9.8 (4.7)	.204[b]
Age of first symptoms, years, mean (*SD*)	64.0 (7.7)	61.8 (10.8)	.117[b]
Time between symptoms onset and neuropsychological assessment, years, mean (*SD*)	2.8 (2.5)	3.3 (2.9)	.163[b]
Presence of depressive symptoms [c],%	34.5%	42.3%	.317[a]
Blessed Dementia Rating Scale, mean (*SD*)	3.3 (2.0)	3.5 (2.0)	.439[b]

Note. [a]Chi-squared Pearson's test; [b]Independent samples Student's *t*-test; [c]Presence of depressive symptoms was considered when GDS_{15} score was higher than 5 points or when GDS_{30} score was higher than 10 points.

Table 2. Neuropsychological tests in Aβ+ and Aβ− aMCI patients (n = 216)

Cognitive domain Neuropsychological tests	Aβ+ aMCI (n = 117) Mean (SD)	Aβ− aMCI (n = 99) Mean (SD)	p value
Global cognition			
Mini-Mental State Examination	26.8 (2.2)	27.6 (2.0)	**.004**
Memory and learning			
Digit Span Forward, z score	0.51 (1.27)	0.24 (1.19)	.122
Word Total Recall, z score	−1.70 (1.53)	−0.96 (1.27)	**<.001**
Logical Memory Immediate Free Recall, z score	−1.49 (1.73)	−0.89 (1.25)	**.005**
Logical Memory Delayed Free Recall, z score	−2.21 (1.23)	−1.61 (1.25)	**.001**
Verbal Paired Associate Learning, z score	−1.56 (1.40)	−0.71 (1.31)	**<.001**
General Information, z score	−0.34 (1.34)	−0.39 (1.26)	.811
Attention and executive functions			
Digit Span Backward, z score	−0.06 (1.14)	−0.04 (1.26)	.886
Trail Making Test A time, z score	−1.49 (2.19)	−0.91 (1.70)	.054
Trail Making Test B time, z score	−2.57 (2.54)	−1.50 (2.28)	**.005**
Cancellation Task, total, z score	0.02 (1.29)	0.33 (1.64)	.139
Verbal Semantic Fluency, z score	−0.56 (1.67)	−0.44 (1.39)	.610
Abstract reasoning			
Raven's Coloured Progressive Matrices, z score	−0.39 (1.42)	−0.12 (1.30)	.151
Interpretation of Proverbs, z score	0.53 (1.62)	0.84 (1.56)	.162
Calculation			
Basic Written Calculation, z score	−0.37 (1.25)	−0.34 (1.46)	.917

Note. Bold values represent statistically significant p values (≤ .05).

on these memory tests still held when the groups were controlled for the MMSE, a general linear model analysis was performed considering the distinct neuropsychological tests as dependent variables and the MMSE score as a covariate. Aβ+ aMCI patients essentially kept poorer performances in the same tests as previously found: Word Total Recall (F = 6.181, p = .003); Logical Memory, Immediate Free Recall (F = 3.077, p = .052); Logical Memory, Delayed Free Recall (F = 7.651, p = .001); and Verbal Paired Associate Learning (F = 12.281, p < .001).

Regarding attention and executive functions, there were no differences in the Digit Span Backward, in the Trail Making Test A, in the Cancellation Task nor in the Verbal Semantic Fluency test; however, the Aβ+ aMCI patients performed significantly worse on the Trail Making Test B. Using the Trail Making Test B over A ratio, we found no significant differences between groups (p = .905). For the Aβ+ aMCI patients, the mean value of the ratio was 2.9 (SD 1.1); for the Aβ− aMCI patients, the mean was 2.9 (SD 1.2). Finally, there were no statistically significant differences between the two groups in the abstract reasoning and calculation domains.

A binary logistic regression model was built in order to predict the amyloid status of aMCI patients. In general, the tests in which there were significant differences between the two groups entered the model. Regarding Logical Memory, the Delayed Free Recall measure was chosen. Only Logical Memory Delayed Free Recall retained statistical significance to determine the amyloid status of aMCI patients. For each additional score point in the Logical Memory Delayed Free Recall z score, the odds ratio was 0.694, that is, the probability of being Aβ+ decreased by 30.6% (Table 3). The resulting model correctly classified 64.6% of the aMCI cases regarding their amyloid status. Only 17.7% of the

Table 3. Neuropsychological predictors of amyloid positivity

Neuropsychological tests	*B*	*SE*	Wald	*p* value	Exp(*B*)	95% C.I. for Exp (*B*) Lower	Upper
Word Total Recall, *z* score	−0.240	.146	2.709	.100	0.787	0.592	1.047
Logical Memory Delayed Free Recall, *z* score	−0.366	.172	4.549	**.033**	0.694	0.495	0.971
Verbal Paired Associate Learning, *z* score	−0.085	.155	0.301	.583	0.918	0.677	1.245
Trail Making Test B time, *z* score	−0.114	.080	2.058	.151	0.892	0.763	1.043

Note. Binary logistic regression analysis.
Bold values represent statistically significant *p* values (≤ .05).

variation in the dependent variable (amyloid positivity) was explained by the present model. The ability of Logical Memory Delayed Free Recall (*z* score) to discriminate between Aβ+ and Aβ− aMCI patients was checked with a ROC curve, producing an area under the curve (AUC) of 0.633.

Since there might be some redundancy in the tests comprising the neuropsychological battery that was applied, a principal component analysis was performed. The rotated varimax component matrix pointed out 5 factors. Of these, there were significant differences between Aβ+ and Aβ− aMCI patients in factor 2 (Memory factor, comprising Logical Memory Immediate Free Recall, Logical Memory Delayed Free Recall, and Verbal Paired Associate Learning; $F = 9.546$, $p = .003$) and in factor 3 (Executive factor, comprising Trail Making Test A time and Trail Making Test B, and Raven's Coloured Progressive Matrices, $F = 5.881$, $p = .017$). These results confirmed that Aβ+ and Aβ− aMCI patients essentially differed in memory test as well as in executive tests.

Discussion

The main finding of the present study is that aMCI patients who are Aβ+ have more deficits in general cognition, memory tests, and executive functions as compared to Aβ− aMCI patients. A few points deserve consideration.

In the first place, we confirmed that Aβ+ aMCI patients are more impaired in memory tests as compared to Aβ− aMCI patients, as previously reported by several studies (Bahar-Fuchs *et al.*, 2013; Huijbers *et al.*, 2015; Kandel *et al.*, 2015; Kim *et al.*, 2018; Reijs *et al.*, 2017; Tomadesso *et al.*, 2018, 2019; Wolk *et al.*, 2009). As patients with aMCI patients who are Aβ+ suffer from AD (Jack *et al.*, 2018), the observed memory deficits correspond to the typical cognitive profile of AD, reflecting the hippocampal atrophy observed early in the course of the disease. In the present work, Word Total Recall, Logical Memory (Immediate and Delayed Free Recall), and Verbal Paired Associate Learning were significantly worse in Aβ+ aMCI patients.

The second point is that we contributed to clarify the controversial issue whether Aβ+ aMCI patients are more affected in executive functions and attention, which has not been clear from previous studies. We showed that tests assessing executive functions, namely the Trail Making Test B, were more affected in Aβ+ aMCI patients. It could be argued that the worse performance on the Trail Making Test part B in Aβ+ when compared to Aβ− aMCI patients was due to impairment of visuospatial abilities in the first group. However, the observation that there were no significant differences between the Aβ+ and Aβ− aMCI

patients in the Raven Progressive Matrices, a visuospatially very demanding test, suggests that differences in the Trail Making Test part B are probably not attributable to visuospatial difficulties. The results concerning Trail Making Tests are in accordance with Kandel *et al*. (2015) reports in aMCI patients from the ADNI cohort, who also found significantly worse results in both Trail Making Tests in Aβ+ patients. We did not observe differences in Verbal Semantic Fluency between Aβ+ and Aβ− aMCI patients, similarly as reported in a previous study (Tomadesso *et al*., 2018). However, another study found that Verbal Semantic Fluency was significantly worse in Aβ+ as compared to Aβ− aMCI patients (Kandel *et al*., 2015). This last study used animal category for the task, while we used supermarket food items, which might explain the discrepancy of the results. Regarding attention, we found no significant differences between the two groups in the Cancellation Task, no previous studies having previously compared, to the best of our knowledge, Aβ+ and Aβ− aMCI patients on this test. More studies are certainly needed to further investigate how the amyloid status influences performances in different tests of executive functions and attention in patients with aMCI. It should be added that patients with aMCI who are Aβ+ showed less global cognitive performance, albeit within the normative range, assessed by the MMSE, as compared to Aβ− aMCI patients, probably reflecting the more pronounced alteration in several cognitive domains, particularly memory and executive functions, as described above.

A third point has to do with the value of neuropsychological tests to predict the patients with aMCI who have amyloid pathology. In the present study, the statistical model could only correctly classify 64.6% of the aMCI cases regarding their amyloid status. The only test that remained in the model was the Logical Memory Delayed Free Recall.

It is noteworthy that in the present study, the Logical Memory Immediate Recall score was chosen to classify patients as aMCI and the Logical Memory Delayed Recall score for analysis, in order to avoid circularity bias. However, it could be argued that both measures were rather equivalent. This did not seem to be the case, as there was no significant collinearity between these neuropsychological test variables, with a variance inflation factor value relating Logical Memory Immediate Recall and Logical Memory Delayed Recall of 1.199.

The Logical memory Delayed Recall score produced a modest AUC (0.633). In a previous study in aMCI patients, the 30-min delayed recall score of the Rey Auditory Verbal Learning Test was the best predictor of Aβ status among the psychometric tests, but it produced an AUC of only 0.67 (Kandel *et al*., 2015). Using a 16-word list, Tomadesso *et al*. (2018) calculated slightly better AUC values for the free recall (0.73) and recognition (0.74) tasks in classifying the aMCI cases according to the amyloid status. It thus seems that neuropsychological tests have a limited ability to identify the aMCI cases who are Aβ+ and those who are Aβ−, not attaining the values of 80% recommended for AD biomarkers (The Ronald and Nancy Reagan Research Institute of the Alzheimer's Association and the National Institute on Aging Working Group, 1998). Of course, these results do not exclude that neuropsychological tests could add predictive value to determine the amyloid status in conjunction with other clinical or neuroimaging biomarkers.

Finally, the intriguing question of the aetiology of aMCI cases who are Aβ− certain merits further research. Depressive symptoms were not more frequent in Aβ− than in Aβ+ aMCI patients. Patients with history of stroke or relevant cerebrovascular disease in brain imaging were excluded in the present study. It is possible that Aβ− aMCI patients might be at an initial stage of a neurodegenerative disorder other than AD, for instance frontotemporal dementia or the Lewy body dementia–Parkinson's disease continuum (Ye *et al*., 2014). To be sure, a long follow-up of these Aβ− aMCI patients might be needed.

The main strength of this study is that it was carried out in the context of a large prospective cohort, in which the participants underwent comprehensive standardized neuropsychological assessment. Several limitations of the study must be recognized. Participants were patients who attended a memory clinic or a general hospital outpatient clinic, and the findings may not applicable to different clinical settings. Certainly, only a proportion of patients with aMCI undergo a comprehensive AD biomarker workout, and these are probably different from those patients with aMCI who do not.

In conclusion, the neuropsychological assessment remains an essential step to diagnose and characterize patients with aMCI. However, neuropsychological tests have limited value to distinguish the aMCI patients who have amyloid pathology and AD, from those who might suffer from other clinical conditions.

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Conflicts of interest

All authors declare no conflict of interest.

Author contributions

Luísa Alves, M.D. (Conceptualization; Writing – original draft); Sandra Cardoso (Data curation; Investigation); Dina Silva (Data curation; Investigation); Tiago Mendes (Visualization); João Marôco (Formal analysis); Joana Nogueira (Investigation; Visualization); Marisa Lima (Investigation; Visualization); Miguel Tábuas-Pereira (Investigation; Visualization); Inês Baldeiras (Investigation; Resources; Visualization); Isabel Santana (Data curation; Investigation; Writing – review and editing); Alexandre de Mendonça (Conceptualization; Formal analysis; Investigation; Methodology; Project administration; Supervision; Writing – original draft; Writing – review and editing); Manuela Guerreiro (Conceptualization; Investigation; Methodology; Supervision).

Data availability statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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