Individualized prognosis of cognitive decline and dementia in mild cognitive impairment based on plasma biomarker combinations

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Abstract

We developed models for individualized risk prediction of cognitive decline in mild cognitive impairment (MCI) using plasma biomarkers of β-amyloid (Aβ), tau and neurodegeneration. A total of 573 patients with MCI from the Swedish BioFINDER study and the Alzheimer's Disease Neuroimaging Initiative (ADNI) were included in the study. The primary outcomes were longitudinal cognition and conversion to Alzheimer's disease (AD) dementia. A model combining tau phosphorylated at threonine 181 (P-tau181) and neurofilament light (NfL), but not Aβ42/Aβ40, had the best prognosis performance of all models (area under the curve = 0.88 for 4-year conversion to AD in BioFINDER, validated in ADNI), was stronger than a basic model of age, sex, education and baseline cognition, and performed similarly to cerebrospinal fluid biomarkers. A publicly available online tool for individualized prognosis in MCI based on our combined plasma biomarker models is introduced. Combination of plasma biomarkers may be of high value to identify individuals with MCI who will progress to AD dementia in clinical trials and in clinical practice.

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Research in context

Evidence before this study: A research framework for Alzheimer's disease (AD) has suggested that biomarkers for β-amyloid (Aβ, A), tau pathology (T) and neurodegeneration (N) can be used to identify AD even in early clinical disease stages. This has been corroborated by many studies using cerebrospinal fluid (CSF) biomarkers. Over the last few years, several studies have also shown that blood-based biomarkers for these processes, including Aβ42/Aβ40, phosphorylated tau (P-tau) and neurofilament light (NfL) are altered in AD and correlate with AD pathology. However, few studies have tested optimal combinations of these biomarkers in blood, especially to predict longitudinal disease progression, and comparisons with bench-marked methods such as CSF biomarkers are lacking. We searched PubMed (on June 1st 2020) for papers on blood-based biomarkers for Aβ, P-tau and neurodegeneration in the prodromal stage of AD (patients with mild cognitive impairment, MCI). The search was restricted to original papers published during the last 10 years. The search query "(((alzheimer's OR alzheimer) AND (prodromal OR MCI OR "mild cognitive impairment") AND (plasma OR serum OR blood) AND (amyloid OR beta-amyloid OR abeta) AND (tau OR P-tau) AND (neurodegeneration OR neurofilament)) NOT (review[Publication Type])) AND (("2010/06/01"[Date - Completion] : "3000"[Date - Completion]))" resulted in 26 papers. None of these papers established and validated prediction models for subject-level individualized prediction of cognitive or clinical change, and none of them compared the predictive accuracy of plasma ATN biomarkers vs CSF ATN biomarker. It is also rare with analyses that test additive effects of biomarkers over baseline cognition.

Added value of this study: We demonstrate that blood-based measures of Aβ, P-tau and NfL have high performance to predict longitudinal disease progression with cognitive decline and conversion to dementia in patients with prodromal AD. The combination of P-tau and NfL

had particularly high performance for individualized prediction and was non-inferior to more complex models (including Aβ), as well as to CSF-based biomarker models. The models were validated between two independent cohorts.

Implications of all the available evidence: Blood-based biomarkers may be used to predict future disease progression in individual patients with MCI. This may reduce the need for CSF analyses or other complex or expensive biomarker investigations and improve the treatment and care of this patient population.

Introduction

About 50 million people live with dementia globally, and the prevalence is increasing fast.[1] Fifty to seventy percent of all dementia cases are caused by Alzheimer's disease (AD).[2] The ability to correctly identify AD as a cause of cognitive impairment is essential to ensure optimal patient management, including access to symptomatic treatments. In patients with mild cognitive impairment (MCI), accurate prognosis is especially important, since MCI in some cases lead to cognitive decline and dementia (due to AD or other diseases), and in some cases is benign and stable.[3] The importance of correct prognostics has been further highlighted by recent advances in candidate disease-modifying treatments targeting amyloid-β (Aβ).[4,5]

Even at the MCI stage, AD can be identified *in vivo*, using cerebrospinal fluid (CSF) biomarkers that reflect key AD features, *e.g.*, the ratio of Aβ42 to Aβ40, and tau phosphorylated at threonine-181 (P-tau181),[6,7] or positron emission tomography (PET) of Aβ and tau.[8,9] But the use of these technologies is not widespread, due the invasiveness of lumbar punctures and the high cost and low availability of PET imaging. There is therefore a growing interest in blood-based biomarkers for AD, with greater accessibility and lower cost.

Blood-based biomarkers of Aβ (A), tau (T), and neurodegeneration (N) in AD[10] include the Aβ42/Aβ40 ratio,[11,12] P-tau181[13-15] and neurofilament light (NfL),[16,17] respectively. Aβ42/Aβ40 ratio and P-tau181 concentration in plasma correlate with Aβ and tau PET findings, respectively, and can distinguish AD dementia from controls and non-AD neurodegenerative disorders.[12-15,18] Blood-based NfL is associated with cortical atrophy and cognitive decline in AD.[19,20] Most previous studies on blood-based AD biomarkers report findings at the group level, and there is a gap in our understanding of how well these biomarkers predict clinical outcomes at the individual patient level. An individualized

approach to risk assessment has recently been applied using CSF and related imaging biomarkers in MCI.[21,22] A similar study is lacking for blood-based biomarkers, and it could be of great value for clinical practice and trials to investigate whether a combination of plasma ATN biomarkers performs as well as CSF biomarkers, and better than more basic prediction models. We have previously done a study with a multivariate approach to examine plasma biomarkers and the risk of progression from MCI to AD dementia,[13] but most other studies focused on evaluating the biomarkers individually.[12,14,15] None of these studies, however, have applied the ATN classification system,[10] or systematically aimed to find the best subset of ATN for individualized predictions. We therefore measured plasma Aβ42/Aβ40, P-tau181 and NfL in patients with MCI from two large clinical cohorts and tested which subset of plasma biomarkers best predicted individual risk for cognitive decline and progression to AD dementia. We compared the prognostic ability of plasma biomarkers to the same biomarkers measured in CSF, as well as to a more basic prediction model and cross-validated our individual-based risk assessment models both within and across cohorts. We made the models easily available for others to use, in an online tool.

Methods

The study procedures are outlined in Figure 1.

Participants

The Swedish BioFINDER (Biomarkers for Identifying Neurodegenerative Disorders Early and Reliably; clinical trial no. NCT01208675, www.biofinder.se) cohort used in the present analysis consisted of MCI patients with available plasma Aβ42/Aβ40, P-tau181, and NfL measurements. Patients with MCI were recruited at the memory clinics in the cities of Lund, Malmö and Ängelholm. They were between 60 and 80 years old and fulfilled the consensus criteria for MCI suggested by Petersen et al,[23] including cognitive complaints, preferably

corroborated by an informant; objective cognitive impairment, adjusted for age and education; preservation of general cognitive functioning and a Mini-Mental State Examination score (MMSE) of 24-30; no or minimum impairment of daily life activities, and not fulfilling criteria for dementia, as described previously in detail.[24] Exclusion criteria included cognitive impairment that could better be accounted for by another non-neurodegenerative condition, severe somatic disease, and current alcohol or substance abuse. After their baseline visit, all patients were seen at least every 2 years in order to assess clinical progression.

Validation was done on patients with MCI in the Alzheimer's Disease Neuroimaging Initiative (ADNI) (see adni.loni.usc.edu and www.adni-info.org for updated information). The ADNI was launched in 2003 as a public-private partnership, and is led by Principal Investigator Michael W. Weiner, MD. Ethical approval was given by the local ethical committees of all involved sites. All participants gave written informed consent.

Outcomes

The co-primary outcomes were the global cognitive measure MMSE and clinical conversion to AD dementia at 4 years after baseline. As secondary outcomes, we used MMSE and conversion to AD dementia at 2 years. As exploratory outcomes, we used the Clinical Dementia Rating Scale - Sum of Boxes (CDRSB) and conversion to dementia due to any cause evaluated at 2 and 4 years after baseline.

In BioFINDER, clinical status of dementia due to AD or other diseases was evaluated according to the diagnostic and statistical manual of mental disorders version 5 (DSM-5) criteria for major neurocognitive disorder (*i.e.*, dementia) and recorded at each visit by a senior neuropsychologist and an experienced memory disorder specialist (SP). In addition to the DSM-5 criteria, a diagnosis of AD was only used if the participant had an abnormal ratio

of CSF phosphorylated-tau (P-tau) to CSF Aβ42 ≥0.022 using the Elecsys immunoassays, which previously has been validated against Aβ PET.

Dementia in ADNI was… [add methods]

Predictors

We measured Aβ42/Aβ40, P-tau181, and NfL in both CSF and plasma. Biomarker values were binarized for certain parts of the analysis, whereby cutpoints were defined using Youden's index to maximize the separation between amyloid-negative (defined by CSF Aβ42 in ADNI and CSF Aβ42/Aβ40 in BioFINDER) cognitively unimpaired (Aβ- CU) participants and amyloid-positive patients with AD dementia (Aβ+ AD); note, none of the participants used to define cutpoints were used in the main analysis.

In BioFINDER, plasma Aβ42/Aβ40 levels were measured using Elecsys immunoassays on a Cobas e601 analyzer (Roche Diagnostics GmbH, Penzberg, Germany).[12] Plasma P-tau181 concentration was measured on a Meso-Scale Discovery platform (MSD, Rockville, MD), using an assay developed by Eli Lilly.[13] Sensitivity analyses were performed using a mass spectrometry-based plasma Aβ42/Aβ40 assay (Araclon Biotech Ltd., Zaragoza, Spain)[25] instead of the Elecsys Aβ42/Aβ40 assay.

In ADNI, plasma Aβ42/Aβ40 was analyzed by a mass spectrometry-based method,[26] and P-tau181 was analyzed on a Single molecule array (Simoa) HD-X Analyzer (Quanterix, Billerica, MA), using an assay developed in the Clinical Neurochemistry Laboratory, University of Gothenburg, Sweden.[14] In both BioFINDER and ADNI, NfL was analyzed using a Simoa-based assay.[19]

In both cohorts, CSF levels of Aβ42 (used in place of Aβ42/Aβ40 since Aβ40 measurements were not available in ADNI) and P-tau181 were measured using the Elecsys platform (Roche Diagnostics GmbH), while CSF NfL was measured using ELISA (UmanDiagnostics AB, Umeå, Sweden).

Statistical Analysis

In the first stage of analysis, three linear regression models were fit with the cognitive outcomes described above as response variable: a basic model (including age, sex, education and baseline MMSE), a plasma model (including age, sex, education and baseline MMSE, and plasma ATN biomarkers), and a CSF model (including age, sex, education and baseline MMSE, and CSF ATN biomarkers). For the plasma and CSF models, each possible combination of ATN biomarkers (either A only; T only; N only; A and T; A and N; T and N; or A and T and N) was tested. Models were compared using the coefficient of determination (R^2) and the Akaike Information Criterion (AIC), and statistical significance of models within CSF or plasma modalities was assessed using the likelihood ratio test while significance of models between CSF and plasma modalities was assessed by comparing R^2 values over 1000 bootstrapped samples. A statistically significant difference was defined as a non-overlapping 95% confidence interval (CI). Additionally, logistic regression models were fit with clinical conversion outcomes described above as response variables, with the same set of predictors and the same method of comparison but with area under the curve (AUC) instead of R^2 as the performance metric.

In the second stage of analysis, the best fitting model identified in the first stage according to AIC score (lower is better) was carried further and its individual-level predictive value was evaluated. This was done first separately within each cohort using 1000 repetitions of five-fold cross validation. In a second step, validation was done externally across cohorts by first fitting the model on all BioFINDER subjects and then testing the estimates derived in BioFINDER on all ADNI subjects, and vice-versa. The external validation procedure was performed with binarized biomarker levels according to the cutoff procedure described above in order to facilitate comparison across assays.

The fitted models were used to create an online application which provides individual risk assessment for all outcomes. All analyses were performed using the R programming language (v4.0.0), with significance set at $P < .05$, two-sided.

Results

Main study population characteristics

152 patients with MCI for which all plasma and CSF biomarkers were available were included from BioFINDER (Table 1). The mean MMSE score was 27.2 ± 1.7 at baseline, 24.8 ± 3.7 two years after baseline, and 21.8 ± 5.2 four years after baseline. Conversion to AD dementia occurred in 25.5% of patients within two years of baseline and 59.8% within four years of baseline. Continuous biomarker levels, and their associations, are shown in Figure S1 (appendix p 12). According to the biomarker cutoff procedure described above, 56.6% of patients were plasma Aβ-positive, 65.1% were plasma P-tau181-positive, and 41.4% were plasma NfL-positive. There was a significant negative correlation between plasma Aβ42/Aβ40 and plasma P-tau181 (R^{2}_{2}=-0.30, P<0.0001) and a significant positive correlation between plasma P-tau181 and plasma NfL (R^2=0.33, P<0.0001), but no significant correlation was observed between plasma Aβ42/Aβ40 and plasma NfL (R^2=-0.08, P=0.22). Similar information on the same biomarkers in CSF, and on the characteristics of the ADNI validation cohort (n=92 for model selection; n=320 for prognostic validation), are shown in Figures S2 and 3 (appendix, pp 13 and 14).

Model selection for longitudinal cognition

The MMSE scores evaluated at four years was a primary outcome. In the BioFINDER cohort (n=118), the model which included plasma Aβ42/Aβ40, P-tau181, and NfL as predictors (note that all models also included age, sex, and education, and baseline MMSE) provided a model fit (R^2=0.36, AIC=684) which was significantly better than the basic model, which

only included age, sex, education and baseline MMSE (R^2=0.24, AIC=702, P=0.0001 compared to full model). The best fitting model according to AIC was that which included only P-tau181 and NfL (R^2=0.36, AIC=683, P=0.32 compared to full model), where there was a significant effect of P-tau181 (β=-1.65 points / log std. dev, P<0.0001) but not for NfL (β=-0.70 points / log std. dev, P=0.13) (Figure 2A-B).

The model selection result was validated with four-year MMSE as outcome in the ADNI cohort (n=64). Here, the model which included all three biomarkers (R^2=0.25, AIC=310) fit the data better than using age, sex, education and baseline MMSE only (R^2=0.15, AIC=316, P=0.01 compared to full model) and the best fitting model according to AIC again included P-tau181 and NfL (R^2=0.25, AIC=309, P=0.39 compared to full model). In the best fitting model, the effect of P-tau181 was nearly significant (β=-0.64 points / log std. dev, P=0.06) while the effect of NfL was significant (β=-1.02 points / log std. dev, P=0.02) (Figure 2A-B).

Results for secondary (two-year MMSE) outcomes were similar to the primary outcome and are described in Table S1 (appendix p 5) and Figure S4 (appendix, p 16). Results for exploratory (four-year CDR-SB) outcomes showed similar findings, with both plasma NfL and P-tau181 contributing to the best performing model (Tables S2 and 3, appendix, pp 5-6; Figures S5 and 6 (appendix, pp 16-17).

Model selection for clinical conversion

Conversion to AD dementia at four years was a co-primary outcome. In the BioFINDER cohort (n=107), the model which included plasma Aβ42/Aβ40, P-tau181, and NfL as predictors provided a model fit (AUC=0.88, AIC=106) which was significantly better than the basic model (AUC=0.70, AIC=140, P<0.0001 compared to full model). The best fitting model according to AIC was the one including only P-tau181 and NfL (AUC=0.88,

AIC=104, P=0.95 compared to full model), where there was a significant effect of P-tau181 (HR=5.87, P=0.0001) but not for NfL (HR=1.73, P=0.10) (Figure 3A-B).

The model selection result was validated with four-year conversion to AD as outcome in the ADNI cohort (n=74). Here, the model which included all three biomarkers (AUC=0.88, AIC=50) fit the data better than using the basic model (AUC=0.74, AIC=57, P=0.005 compared to full model) and the best fitting model according to AIC again included P-tau181 and NfL (AUC=0.89, AIC=49, P=0.32 compared to full model). In the best fitting model, the effect of P-tau181 was significant (HR=4.58, P=0.009) while the effect of NfL was not significant (HR=2.15, P=0.20) (Figure 3A-B).

Results for the secondary (two-year conversion to AD) conversion outcome were similar to the primary conversion outcome and are described in Table S4 (appendix p 8) and Figure S7 (appendix p 19). Results for exploratory (dementia due to any cause) conversion outcomes are described in Tables S5 and S6 (appendix pp 9-10) and Figures S8 and 9 (appendix pp 20-21).

Sensitivity analysis using an alternative plasma Aβ42/Aβ40 assays

Because plasma Aβ42/Aβ40 was not selected as part of any best fitting models above, we tested whether this changed when using a mass spectrometry assay (Araclon Biotech Ltd) instead of the Elecsys assay in the BioFINDER cohort. The best fitting models still did not include Aβ42/Aβ40 for any of the co-primary outcomes (Tables S7 and 8, appendix pp 11-12; Figures S10 and 11, appendix pp 22-23).

Individual-level risk assessment within cohorts

Since the model which included both P-tau181 and NfL, but not Aβ42/Aβ40, consistently provided the best fit across co-primary outcomes, this model was taken forward to evaluate

out-of-sample prediction at the subject level using cross-validation within each cohort separately. Biomarkers were still used as continuous measures for this procedure, and all individuals with available plasma P-tau181 and NfL measurements were included in this analysis stage, regardless of plasma Aβ42/Aβ40 availability.

With four-year MMSE as outcome in BioFINDER (n=118), the best fitting plasma model (P-tau181 and NfL) significantly improved cross-validated, out-of-sample prediction compared to the basic model (mean absolute error [MAE]=3.07 points versus 3.36 points, P<0.001, 8.5% improvement) and showed no difference compared to the full CSF-based model (P=0.68 over 1000 bootstrapped trials). In the ADNI cohort (n=252), the plasma model significantly improved out-of-sample prediction of four-year MMSE compared to the basic model (MAE=2.42 points versus MAE=2.49 points, P<0.001, 2.9% improvement) (Figure 4).

With four-year conversion to AD as outcome in BioFINDER (n=107), the best fitting plasma model (P-tau181 and NfL) significantly improved out-of-sample prediction compared to the basic model (AUC=0.86 versus AUC=0.75, P<0.001, 14.7% improvement) and actually significantly out-performed the full CSF-based model (P=0.002 over 1000 bootstrapped trials, 5% improvement). In the ADNI cohort (n=320), the plasma model significantly improved out-of-sample prediction of four-year conversion to AD compared to the basic model (AUC=0.66 versus 0.76, P<0.001, 15.4% improvement) (Figure 4).

Individual-level risk assessment across cohorts

Finally, we evaluated out-of-sample prediction across cohorts by first fitting models on the BioFINDER cohort and testing on the ADNI cohort, and vice-versa. Again, only the model including plasma P-tau181 and NfL was carried forward to this stage. For this analysis, biomarkers were dichotomized according to the procedure described above in order to facilitate the application of fitted models across cohorts/assays.

With four-year MMSE as outcome (n=118 in BioFINDER of which 28 T-N-, 13 T-N+, 46 T+N-, 31 T+N+; n=252 in ADNI of which 118 T-N-, 35 T-N+, 46 T+N-, 44 T+N+), the dichotomized plasma model significantly improved prediction on the test cohort compared to the basic model, both when the model was fit on BioFINDER and tested on ADNI (MAE=3.74 versus 4.08, P=0.0006, 8.3% improvement) and when the model was fit on ADNI and tested on BioFINDER (MAE=4.15 versus 5.19, P<0.0001, 20.1% improvement).

With four-year conversion to AD as outcome (n=107 in BioFINDER of which 20 T-N-, 5 T-N+, 49 T+N-, 33 T+N+; n=320 in ADNI of which 34 T-N-, 16 T-N+, 13 T+N-, 11 T+N+), the dichotomized plasma model improved prediction on the unseen cohort both when the model was fit on ADNI and tested on BioFINDER (AUC=0.76 versus 0.88, P<0.0001, 14.7% improvement) and when the model was fit on BioFINDER and tested on ADNI (AUC=0.77 versus 0.82, P<0.0001, 6.9% improvement).

Discussion

We addressed the prognostic value of plasma AD biomarkers (Aβ42/Aβ40, P-tau181 and NfL) in MCI using a precision medicine-based approach for subject-level prediction. We found that P-tau181 in combination with NfL best modelled primary outcomes of decline in MMSE and clinical progression to AD dementia over four years. These results were robust to time horizon (two- or four-years follow-up), selection of outcome (MMSE, CDR-SB, conversion to AD dementia or all-cause dementia), different cohorts, and choice of Aβ assay. In general, the plasma-based models were non-inferior to predictions using CSF biomarkers, and significantly better than a basic model including age, sex, education and baseline MMSE. Our results also held over both internal and external cross-validation, demonstrating the generalizability of plasma biomarker models. We have implemented the models in an online

tool (https://brainapps.shinyapps.io/plasmaatnapp/), where prognostic information at the individual patient level can be obtained.

Our study is novel in the way we address the individualized predictive value of plasma AD biomarkers, but it can be compared to previous work examining CSF and imaging biomarker-driven prognosis at the MCI stage.[21,22] Using four separate prognostic models— including age, sex, Aβ42, T-tau and MMSE; and an ATN variant combining CSF Aβ42 and P-tau181 with hippocampal volume—van Maurik and colleagues looked at the likelihood of progression to dementia from MCI over time frames of 1, 3, and 5 years.[22] While all models performed well, the highest performance was seen using the CSF ATN model. Similarly, we found that a combined ATN model (Aβ42/Aβ40, P-tau181 and NfL), but now in plasma, outperformed a model combining age, sex, education and baseline MMSE. The fact that both P-tau181 and NfL were selected in the best models may reflect that P-tau181 detects AD-specific changes[13] while NfL is a more general marker of neurodegeneration.[19] Our results suggest that plasma Aβ biomarkers do not provide additional prognostic information for MCI patients when an efficient plasma tau measure is included. This is logical, since symptoms in AD are linked to tau,[27] and elevations in tau appear to be dependent on Aβ pathology.[28,29] Findings for plasma Aβ42/Aβ40 have also been more varied than for plasma P-tau181 and with only modest reductions (10-15% in AD dementia compared to CU).[12,30,31] However, it is still possible that plasma Aβ42/Aβ40 may have added value at the preclinical stage of the disease, where it has reached pathological levels,[12] while tau and neurodegeneration markers continue to increase during the symptomatic stages of the disease.[13,32] Biomarker-driven prediction models could provide a tool for more accurate diagnostic work-up, including individualized prognosis, in patients with MCI; this might improve treatment and care[33] and could fast-track participants with prodromal AD to clinical trials by only including those with a high risk of future progression.

In addition to the above described studies by van Maurik on individualized biomarker-based risk predictions of dementia in patients with MCI,[21,22] recent work from our group has also examined the association between plasma-based biomarkers and the risk of future AD dementia.[13] Though similar to the present work in terms of including plasma Aβ42/Aβ40, P-tau181 and NfL, the present study differs from our previous work in a number of important ways. First, the present work focused on identifying optimal models within the ATN framework. In terms of methods, the present study focused on risk prediction at the individual and not group level, and systematically tested all possible plasma ATN subsets. We also did extensive internal and external validation analyses (including in a new cohort, ADNI). In terms of results, one important difference compared to the previous study[13] is the findings that both NfL and P-tau181 (rather than just P-tau181) contribute to the best performing models.

When assessing ATN combinations, we tested for non-inferiority compared to the full ATN model. Though the relative importance of biomarkers may vary across contexts and intended applications, plasma biomarkers are likely to eventually be used across research, specialist clinical and general clinical settings due their accessibility and lower cost. Given the comparative recency of plasma-based AD biomarkers,[34] measurement standardization may be a limiting factor. Standardization efforts for plasma biomarkers are behind those for CSF, where the field has only recently resolved the methodological issues that have complicated widespread use. Similar to CSF, universal cutoffs will need to be complemented by fully validated clinical-grade assays for plasma biomarkers to be used in clinical practice.[35]

Strengths of our study include the use of CSF-based ATN models as an internal performance benchmark and the construction of models providing risk estimates at the subject level. We performed several sensitivity analyses, including for clinical outcomes and

for the method used to measure plasma Aβ42/Aβ40. Validation in two independent cohorts with greatly differing demographic makeup speaks to the robustness and clinical relevance of our findings. Patients in BioFINDER have been recruited in a consecutive fashion at three different memory clinics, with approximately 90% of these referred by primary care physicians. Patients in the ADNI study were recruited from many different clinics and may be more representative of a highly selected clinical trial population. One limitation of the study is the relatively modest sample size. Further studies on larger and more diverse populations, including in primary care, may result in more precise and generalizable models.

Conclusions

Plasma-based AD biomarkers can provide patient level prognostic information in patients with MCI, comparable to CSF biomarkers. Specifically, plasma P-tau181 in combination with NfL seems to best predict cognitive decline and clinical progression. The present results indicate that plasma biomarkers of core AD features may aid in individualized risk assessment for MCI patients, which represents a critical step towards accessible precision medicine for cognitive diseases. However, development of high-precision assays with established universal cutoffs anchored to certified reference materials for assay standardization, and replication of the findings in larger clinically relevant cohorts are needed.

Data sharing

Anonymized study data for the primary analyses presented herein are available upon request from any qualified investigator for purposes of replicating reported results.

Conflicts of interest

NC, AL, SP, NMC have nothing to disclose. OH has acquired research support (for the institution) from Roche, Pfizer, GE Healthcare, Biogen, Eli Lilly and AVID Radiopharmaceuticals. In the past 2 years, he has received consultancy/speaker fees (paid to the institution) from Biogen and Roche. HZ has served at scientific advisory boards for Denali, Roche Diagnostics, Wave, Samumed, Siemens Healthineers, Pinteon Therapeutics and CogRx, has given lectures in symposia sponsored by Fujirebio, Alzecure and Biogen.

OTHERS ADD

Author contributions

AL, NMC and OH conceived the study. AL and NC performed the statistical analysis. ES, SP and OH recruited participants and collected clinical data. HZ, JA, JD, KB, NP, PP and SJ were responsible for biochemical analyses. NMC developed an online tool implementing the statistical models. AL, NC, NMC and OH drafted the initial manuscript. All authors contributed to revision and editing of the manuscript.

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Panel: Online individualized risk prediction tool

We provide an online tool at https://brainapps.shinyapps.io/plasmaatnapp/ where individualized predictions can be done for MMSE, conversion to AD dementia, and CDR-SB at 2 year and 4 year after baseline, in patients with MCI at baseline. The tool allows the user to enter age, sex, baseline cognition (MMSE, CDR-SB), and dichotomous biomarker status for CSF or plasma Aβ42/Aβ40, P-tau181 and NfL. It is also possible to test predictions with sparse models including subsets of biomarkers. For example, for a 70-year-old female with MCI and baseline MMSE of 27, the predicted probability of conversion to AD is 33% (90% prediction interval 23-45%) at 2 years and 69% (56-80%) at 4 years, without biomarker information. If all plasma Aβ42/Aβ40, P-tau181 and NfL are known and negative, the probabilities are 6% (2-20%) at 2 years and 16% (5-38%) at 4 years. If all plasma Aβ42/Aβ40, P-tau181 and NfL are positive, the probabilities change to 43% (25-62%) at 2 years and 92% (77-97%) at 4 years.

Tables

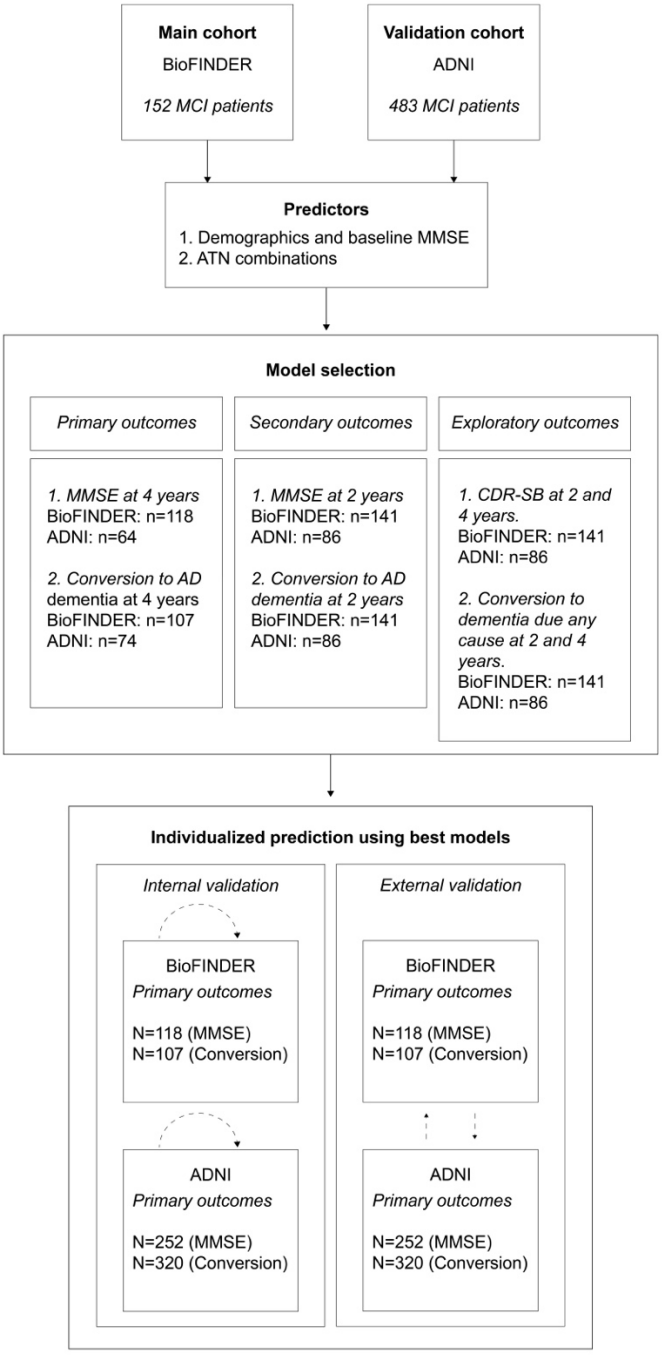
Table 1. Study participant characteristics

			ADNI cohorts		
		BioFINDER	Model selection	Prognostic validation sample	P
n		152	92	483	
Age (mean [SD])		71.28 (5.47)	71.30 (7.53)	71.09 (7.84)	0.942
Education (mean [SD])		11.16 (3.49)	16.41 (2.62)	15.99 (2.67)	<0.001
Sex (%)	M	98 (64.5)	46 (50.0)	235 (48.7)	0.003
	F	54 (35.5)	46 (50.0)	248 (51.3)	
MMSE scores					
Baseline (mean [SD])		27.20 (1.73)	28.33 (1.72)	28.13 (1.68)	<0.001
Two-years (mean [SD])		24.76 (3.66)	28.16 (2.00)	27.27 (2.76)	<0.001
Four-year (mean [SD])		21.78 (5.25)	27.62 (2.92)	26.57 (4.10)	<0.001
Conversion to AD dementia					
Two-year (%)	No	105 (74.5)	80 (93.0)	337 (79.7)	0.003
	Yes	36 (25.5)	6 (7.0)	86 (20.3)	
Four-year (%)	No	43 (40.2)	66 (89.2)	230 (66.9)	<0.001
	Yes	64 (59.8)	8 (10.8)	114 (33.1)	
Plasma biomarkers					
Aβ42/Aβ40 (mean [SD])		4.15 (0.12)	0.12 (0.01)	0.12 (0.01)	<0.001
Aβ42/Aβ40 status (%)	-	66 (43.4)	43 (46.7)	43 (46.7)	0.831
	+	86 (56.6)	49 (53.3)	49 (53.3)	
P-tau181 (mean [SD])		0.89 (0.80)	2.64 (0.60)	2.69 (0.62)	<0.001
P-tau181 status (%)	-	53 (34.9)	59 (64.1)	291 (60.2)	<0.001
	+	99 (65.1)	33 (35.9)	192 (39.8)	
NfL (mean [SD])		3.14 (0.45)	3.46 (0.43)	3.49 (0.48)	<0.001
NfL status (%)	-	89 (58.6)	60 (65.2)	322 (66.7)	0.188
	+	63 (41.4)	32 (34.8)	161 (33.3)	

This table summarizes characteristics for study participants in the BioFINDER and ADNI cohorts. The ADNI model selection cohort was used to validate the model selection procedure in the first analysis stage, while the ADNI prognostic validation cohort was used to test individualized prediction performance of the best model identified during the model selection step M=male; F=female; -/+ indicates negative (normal) or positive (abnormal) biomarker values. Biomarker concentrations are given as pg/mL.

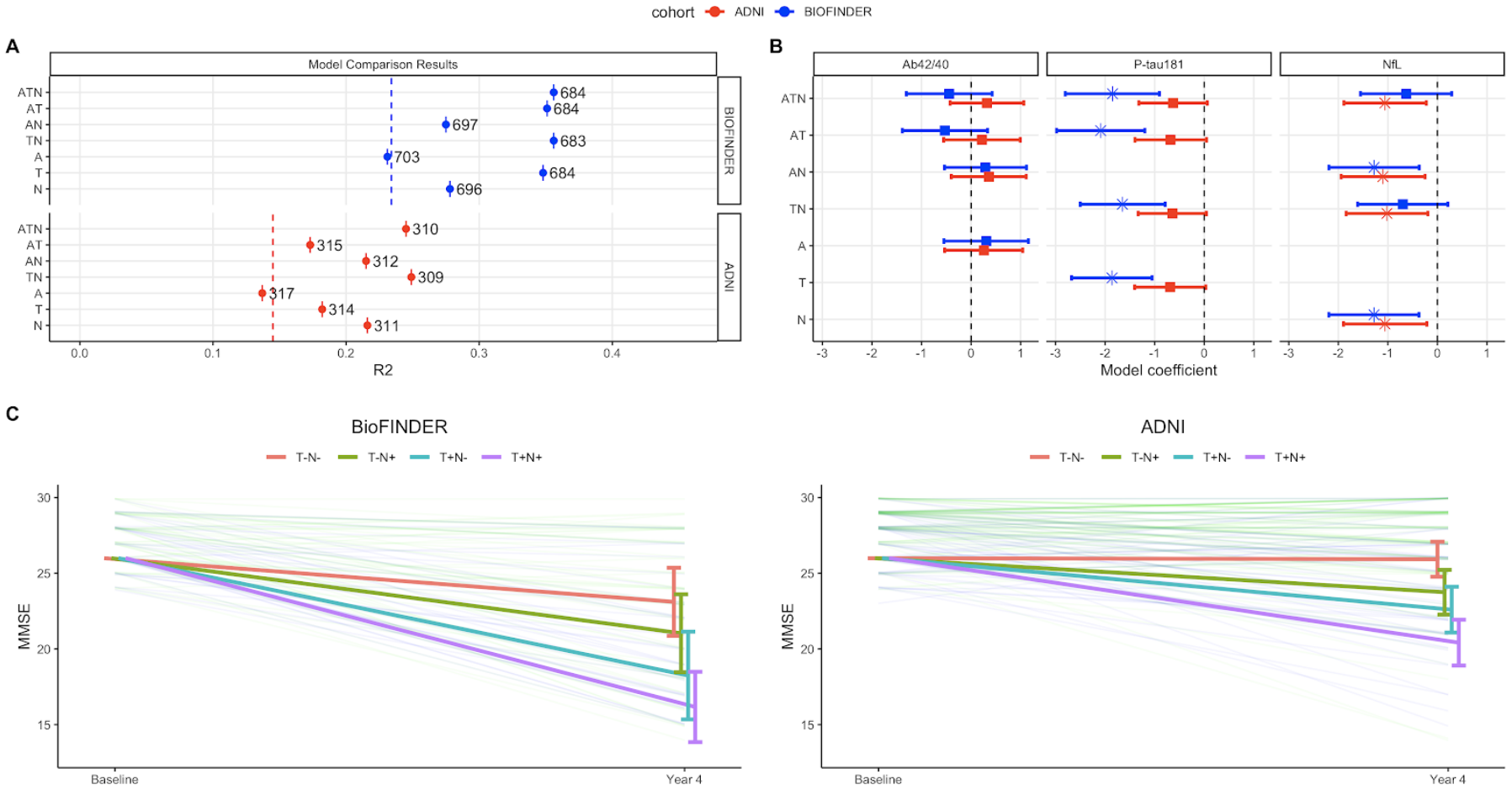
Figures

Figure 1. Flow chart of study procedures



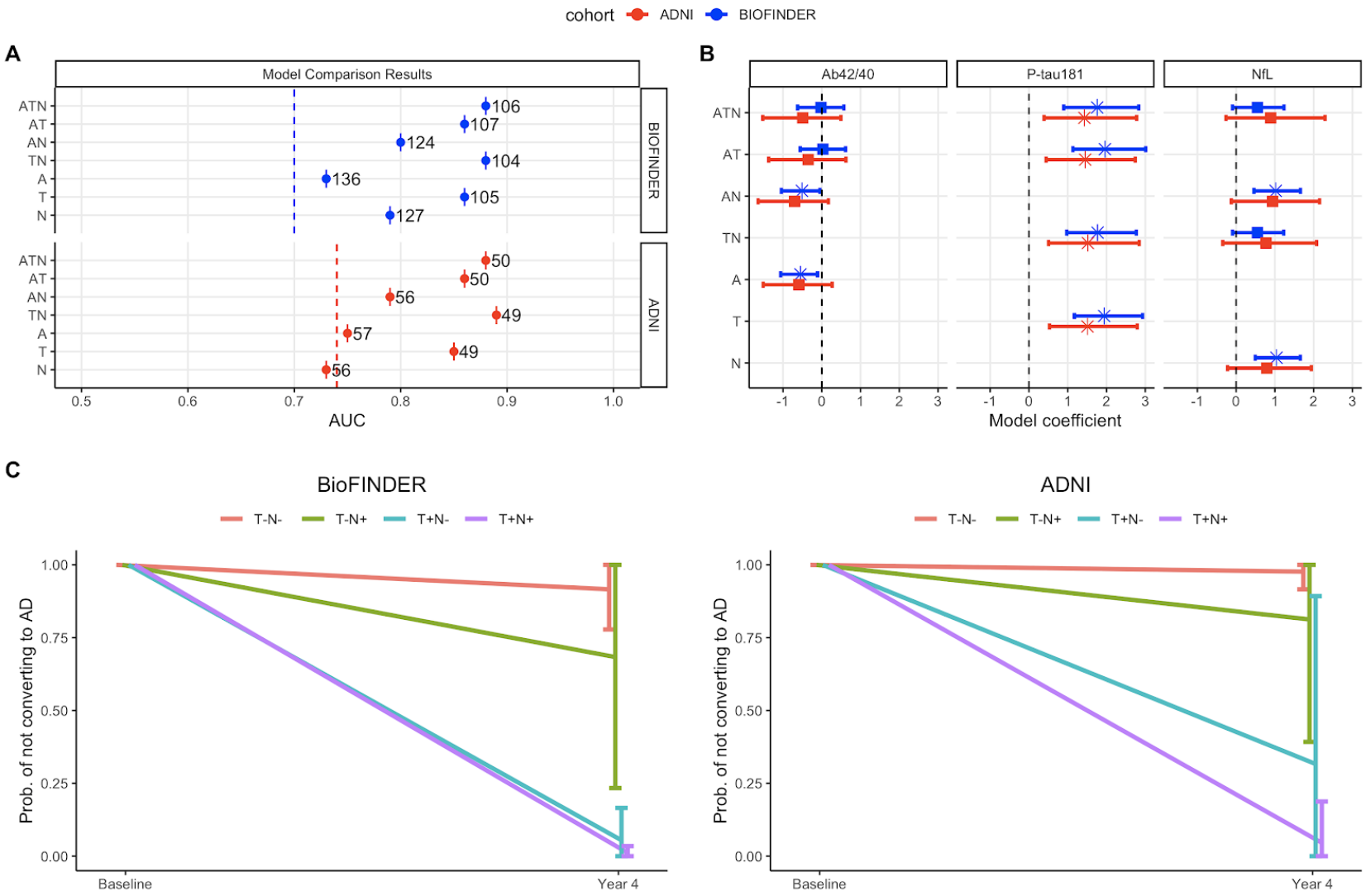
This figure summarizes the methods used in the study

Figure 2. Modelling cognitive decline using plasma Aβ42/Aβ40, P-tau181, and NfL



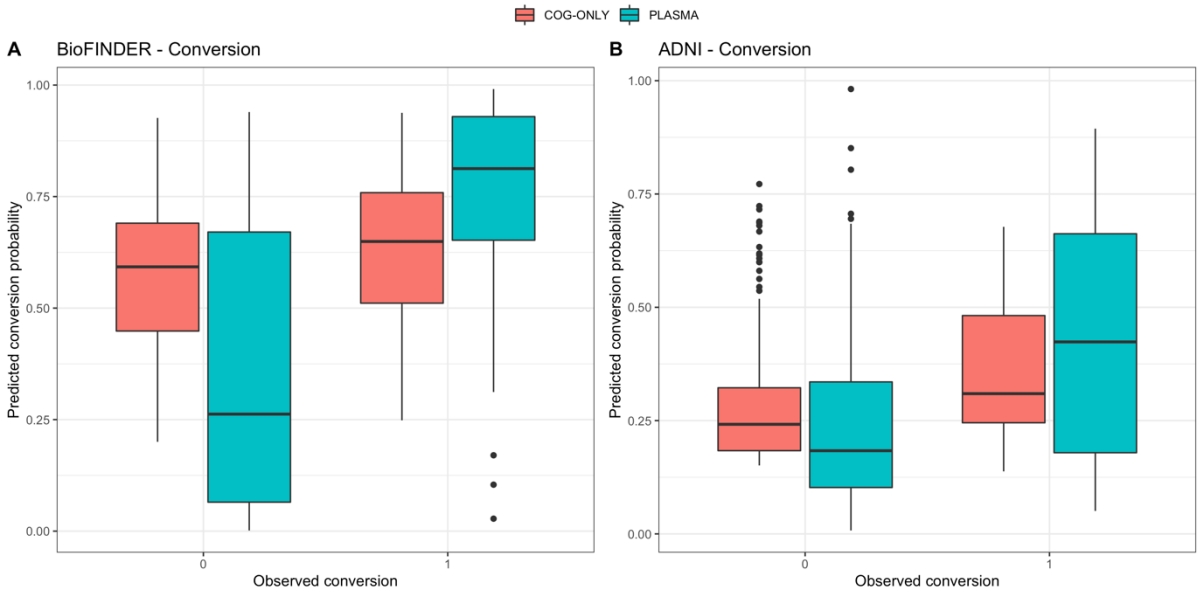
This figure shows the results from modelling cognitive decline in MCI patients using CSF and plasma biomarkers. (A) The R^2 (x-axis) and AIC (values in plot) for each plasma-based model with MMSE evaluated four years after baseline as outcome in ADNI and BioFINDER cohorts are plotted and the basic model (age, sex, education and baseline MMSE) and the CSF-based ATN model are shown for reference as dashed vertical lines. All models also included age, sex, education and baseline MMSE as predictors. (B) The coefficients from each plasma-based model are shown with MMSE evaluated four years after baseline as outcome in ADNI and BioFINDER cohorts. Statistically significant variables are plotted with a star instead of a square and lines represent 95% confidence intervals. (C) The observed MMSE trajectories (shaded lines) together with the estimated trajectories from the best fitting model (P-tau181 and NfL) according to biomarker status, adjusted for age, sex, education and baseline MMSE.

Figure 3. Modelling clinical conversion using plasma Aβ42/Aβ40, P-tau181, and NfL



This figure shows the results from modelling clinical conversion in MCI patients using CSF and plasma biomarkers. (A) The AUC and AIC values for each plasma-based model with conversion to AD within four years after baseline as outcome in BioFINDER and ADNI cohorts are plotted and the basic model and the CSF-based ATN model are shown for reference as dashed vertical lines. All models also included age, sex, education and baseline MMSE as predictors. (B) The coefficients from each plasma-based model are shown with conversion to AD within four years after baseline as outcome in BioFINDER and ADNI cohorts. Statistically significant variables are plotted with a star instead of a square. (C) The estimated probability of not converting to AD as predicted from the best fitting model (P-tau181 and NfL) according to biomarker status, adjusted for age, sex, education and baseline MMSE.

Figure 4. Individualized prediction of conversion from MCI to AD dementia



This figure shows the results from internal cross-validation for clinical conversion for the best performing models as identified in the first stage of analysis. The values plotted here show the predicted probability of conversion from MCI to AD dementia for each individual in the BioFINDER and ADNI cohorts, showing a 14.7% improvement of the plasma-based model over of the basic model in BioFINDER and a 15.4% improvement in ADNI. In BioFINDER, there were 43 (40) false (true) negatives and 3 (21) false (true) positives for the basic model (AUC=0.62) and 13 (30) false (true) negatives and 13 (52) false (true) positives for the P-tau181 and NfL model (AUC=0.83). In ADNI, there were 52 (175) false (true) negatives and 43 (50) false (true) positives for the model combining age, sex, education and baseline MMSE (AUC=0.66) and 35 (166) false (true) negatives and 52 (67) false (true) positives for the P-tau181 and NfL model (AUC=0.76).

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