Articles

Clinical and biomarker changes of Alzheimer's disease in adults with Down syndrome: a cross-sectional study

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Summary

Background Alzheimer's disease and its complications are the leading cause of death in adults with Down syndrome. Studies have assessed Alzheimer's disease in individuals with Down syndrome, but the natural history of biomarker changes in Down syndrome has not been established. We characterised the order and timing of changes in biomarkers of Alzheimer's disease in a population of adults with Down syndrome.

Methods We did a dual-centre cross-sectional study of adults with Down syndrome recruited through a population-based health plan in Barcelona (Spain) and through services for people with intellectual disabilities in Cambridge (UK). Cognitive impairment in participants with Down syndrome was classified with the Cambridge Cognitive Examination for Older Adults with Down Syndrome (CAMCOG-DS). Only participants with mild or moderate disability were included who had at least one of the following Alzheimer's disease measures: apolipoprotein E allele carrier status; plasma concentrations of amyloid β peptides 1–42 and 1–40 and their ratio ($A\beta_{1-42/1-40}$), total tau protein, and neurofilament light chain (NFL); tau phosphorylated at threonine 181 (p-tau), and NFL in cerebrospinal fluid (CSF); and one or more of PET with ^{18}F-fluorodeoxyglucose, PET with amyloid tracers, and MRI. Cognitively healthy euploid controls aged up to 75 years who had no biomarker abnormalities were recruited from the Sant Pau Initiative on Neurodegeneration. We used a first-order locally estimated scatterplot smoothing curve to determine the order and age at onset of the biomarker changes, and the lowest ages at the divergence with 95% CIs are also reported where appropriate.

Findings Between Feb 1, 2013, and June 28, 2019 (Barcelona), and between June 1, 2009, and Dec 31, 2014 (Cambridge), we included 388 participants with Down syndrome (257 [66%] asymptomatic, 48 [12%] with prodromal Alzheimer's disease, and 83 [21%] with Alzheimer's disease dementia) and 242 euploid controls. CSF $A\beta_{1-42/1-40}$ and plasma NFL values changed in individuals with Down syndrome as early as the third decade of life, and amyloid PET uptake changed in the fourth decade. ^{18}F-fluorodeoxyglucose PET and CSF p-tau changes occurred later in the fourth decade of life, followed by hippocampal atrophy and changes in cognition in the fifth decade of life. Prodromal Alzheimer's disease was diagnosed at a median age of 50·2 years (IQR 47·5–54·1), and Alzheimer's disease dementia at 53·7 years (49·5–57·2). Symptomatic Alzheimer's disease prevalence increased with age in individuals with Down syndrome, reaching 90–100% in the seventh decade of life.

Interpretation Alzheimer's disease in individuals with Down syndrome has a long preclinical phase in which biomarkers follow a predictable order of changes over more than two decades. The similarities with sporadic and autosomal dominant Alzheimer's disease and the prevalence of Down syndrome make this population a suitable target for Alzheimer's disease preventive treatments.

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Introduction

Down syndrome (also referred to as trisomy 21) is the most frequent form of genetic developmental and intellectual disability, affecting 5·8 million people worldwide.[1] Life expectancy for people with Down syndrome has greatly increased because of improved medical care, exceeding 60 years of age.[2] Consequently, age-related comorbidities in this group have emerged, particularly Alzheimer's disease. The lifetime risk of Alzheimer's disease in people with Down syndrome is now more than 90%,[3] and the disease is the leading cause of death in this population.[4] The strong association between Down syndrome and Alzheimer's disease has a genetic basis through a gene-dose effect of amyloid β (Aβ) precursor protein, which is located on chromosome 21 and is overexpressed in people with Down syndrome.[1]

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Research in context

Evidence before this study
We searched and reviewed the literature using PubMed, meeting abstracts, and presentations. We searched PubMed on Nov 30, 2018, for research studies on established Alzheimer's disease biomarkers in the plasma and cerebrospinal fluid (CSF; amyloid β (Aβ), tau protein, and neurofilament light protein [NFL]), MRI, or PET in people with Down syndrome using the search terms "biomarker" OR "plasma" OR "CSF" OR "MRI" OR "PET" AND "Down Syndrome" AND ("Alzheimer's" or "dementia"). Several previous studies in Down syndrome have assessed Aβ brain deposition with PET tracers, studied plasma and CSF biomarkers, or described the atrophy and cerebral metabolic Alzheimer's disease patterns. However, we found no previous multimodal studies assessing the natural history of Alzheimer's disease in individuals with Down syndrome.

Added value of this study
To our knowledge, this is the first large multimodal biomarker study to characterise the natural history of Alzheimer's disease in adults with Down syndrome. Biomarker changes begin more than two decades before Alzheimer's disease dementia onset in a strikingly similar order and timing to that described in patients with autosomal dominant Alzheimer's disease.

Implications of all the available evidence
Our study supports people with Down syndrome as a suitable population for clinical trials for Alzheimer's disease. The description of the natural history of Alzheimer's disease in this population would have an immediate effect on the design of such trials. It would also have implications in clinical practice because it supports the concept of Down syndrome as a form of genetically determined Alzheimer's disease with a predictable sequence of biomarker changes.

Consequently, Down syndrome is now conceptualised as a form of genetically determined Alzheimer's disease, similar to its autosomal dominant form.[5]

Alzheimer's disease pathology has been described in all adults with full trisomy 21 by the age of 40 years, and its hallmarks are qualitatively similar to those of sporadic Alzheimer's disease.[6] Increasing evidence from biomarker studies also suggest that the pathophysiology of the disease in Down syndrome is similar to that of the sporadic and autosomal dominant forms of Alzheimer's disease.[7,8] Several studies in individuals with Down syndrome have assessed Aβ brain deposition with PET tracers, studied plasma and cerebrospinal fluid (CSF) biomarkers, or described the atrophy and cerebral metabolic patterns of Alzheimer's disease.[9–14] Although longitudinal studies of Alzheimer's disease in individuals with Down syndrome exist,[15] the natural history of biomarker changes in Down syndrome has not been established.[1,8,16–18] In this study, we assessed the order and timing of Alzheimer's disease biomarker changes in a large cohort of adults with Down syndrome. We used biochemical, neuroimaging, and cognitive measures covering all three categories of the AT(N) biomarker system (assessing deposition of Aβ, hyperphosphorylated tau, and neurodegeneration).[19] Understanding this pattern of changes is crucial for the design of trials in Down syndrome aimed at preventing or moderating the progression of Alzheimer's disease.[8]

Methods

Study design and participants
We did a dual-centre cross-sectional study of adults with Down syndrome and euploid controls in Barcelona (Hospital of Sant Pau, Spain) and Cambridge (University of Cambridge, UK). Adults aged 18 years or older with Down syndrome in Barcelona were recruited from a population-based health plan designed to screen for Alzheimer's disease dementia, which includes yearly neurological and neuropsychological assessments. Individuals in this plan interested in research studies are included in the Down Alzheimer Barcelona Neuroimaging Initiative cohort.[14] In Cambridge, participants with Down syndrome were recruited through services for people with intellectual disabilities in England and Scotland, with the support of the UK Down Syndrome Association.[12] We included all adults with Down syndrome who had at least one biochemical or imaging Alzheimer's disease biomarker. For euploid controls, we recruited non-trisomic individuals aged 18 years or older up to 75 years from the Sant Pau Initiative on Neurodegeneration.[20] Euploid controls underwent a structured neurological assessment and a comprehensive battery of neuropsychological tests to establish normal cognitive health. Inclusion criteria were normal neuropsychological results for their age and education level, a Clinical Dementia Rating Scale score of 0, and normal levels of Alzheimer's disease biomarkers in CSF.[20]

The study was approved by the Sant Pau and the Cambridge Research Ethics Committees, following the standards for medical research in humans recommended by the Declaration of Helsinki. All participants or their legally authorised representative gave written informed consent before enrolment.

Procedures
For the purpose of dementia diagnosis, we provided the participants' caregivers with a semi-structured adapted health questionnaire, the Cambridge Examination for

For the **Clinical Dementia Rating Scale** see https://www.madrc.org/clinical-dementia-rating-cdr-scale

Mental Disorders of Older People with Down Syndrome and others with intellectual disabilities (CAMDEX-DS) developed in Cambridge, with a version adapted for the Spanish population. The CAMDEX-DS includes a comprehensive cognitive battery of neuropsychological tests, the Cambridge Cognitive Examination for Older Adults with Down Syndrome (CAMCOG-DS), that covers seven different cognitive domains.[21] We classified participants with Down syndrome into asymptomatic, having prodromal Alzheimer's disease, or having Alzheimer's disease dementia, in a consensus meeting between the neurologist or psychiatrist and the neuropsychologists who assessed the participants while masked to biomarker data, as previously described.[14] Because of the floor effects of the CAMCOG-DS in adults with Down syndrome with severe and profound levels of intellectual disability,[21] we only included participants with mild or moderate intellectual disability in this analysis. We stratified the level of intellectual disability according to the Diagnostic and Statistical Manual of Mental Disorders (fifth edition)[22] as mild, moderate, or severe or profound (which were grouped together) on the basis of the individuals' best-ever level of functioning, as determined from carers' reports. The information was obtained through family interviews and review of medical or educational records for past assessment results.

We screened patients for trisomy 21 using the Illumina Infinium Global Screening Array (Illumina, San Diego, CA, USA) as previously described.[14] We also determined apolipoprotein E (*APOE*)-ε4 allele carrier status through detection of polymorphisms rs429358 and rs7412 in exon 4 via Sanger sequencing.[7,14]

A subset of participants underwent a 3 Tesla MRI, a ^{18}F-fluorodeoxyglucose PET, amyloid PET, or a combination of these acquisitions. The amyloid tracer used was ^{18}F-florbetapir in Barcelona and ^{11}C-Pittsburg compound B in Cambridge. Structural T1 MRI was processed with Freesurfer (version 6)[23] to extract the adjusted hippocampal volumes (appendix pp 3–4).[24] ^{18}F-fluorodeoxyglucose and ^{18}F-florbetapir PET images were co-registered to the individual MRI, and standardised uptake value ratios were extracted from the corresponding Landau regions.[25,26] Methods and findings from amyloid PET or CT scans in the Cambridge cohort have been published previously,[12] but data were reanalysed and converted to the Centiloid scale and combined with the Barcelona ^{18}F-florbetapir data following standard procedures (appendix pp 5–6).[27]

CSF and blood samples were acquired concurrently on the same day. Plasma concentrations of Aβ 1–42 (Aβ$_{1-42}$) and Aβ$_{1-40}$, total tau protein, and neurofilament light chain (NFL) were measured using single molecule array (Simoa; Quanterix, Billerica, MA, USA) at Centre Hospitalier Universitaire Montpellier (Montpellier, France)[14] or at Hospital of Sant Pau (Barcelona, Spain). CSF concentrations of Aβ$_{1-42}$, Aβ$_{1-40}$, tau phosphorylated at threonine 181 (p-tau), and total tau were quantified with a commercially available immunoassay in a fully automated

platform (Lumipulse, Fujirebio-Europe, Ghent, Belgium).[20] CSF NFL concentrations were measured with a commercial ELISA (UmanDiagnostics, Umeå, Sweden), following the manufacturer's recommendations. All CSF samples were analysed at Hospital Sant Pau (appendix p 7). Methods and findings from plasma and CSF biomarkers have been published previously,[14] but the age-related changes were not assessed. All participants from our paper assessing the diagnostic performance[5] have been included in this Article. Notably, the methods for CSF biomarker measurement differs between the two studies (we used a commercially available ELISA in the previous study, but the Lumipulse fully automated platform for this study). Additionally, in this study, we have included 66 new plasma samples and 47 new CSF samples from adults with Down syndrome and 56 new plasma samples and 160 new CSF samples from control participants.

Statistical analysis

We assessed differences in baseline characteristics between the diagnostic groups with the Kruskal-Wallis test and a pairwise Wilcoxon test corrected for multiple comparisons. To determine the order and temporality of the biomarker changes and cognitive decline in participants with Down syndrome, we fitted a first-order locally estimated scatterplot smoothing curve for controls and adults with Down syndrome independently.[16,28] The model uses a standard tricubic weight function with a span parameter to 0·75. The exact age at which the intervals diverge is dependent on intrinsic limitations of studies assessing the natural history of biomarkers, such as the nature of the variable, the sensitivity of the assay, the slope of the association, and, in our study, the uneven sample sizes for the different biomarkers. Therefore, we defined biomarker change as the age at which the groups appear to start diverging visually. Nonetheless, we also provide the lower age at which the 95% CIs between groups did not overlap. When neurodevelopmental differences were present (different offsets such as hippocampal volumes in the youngest individuals) or when no data were available for the controls (eg, CAMCOG-DS scores), we visually described only the trajectory in adults with Down syndrome.

To compare the timing of changes in Down syndrome and autosomal dominant Alzheimer's disease, we presented the biomarker changes both according to the chronological age and with respect to the median age of diagnosis of prodromal Alzheimer's disease (referred in this study as expected symptom onset).

All statistical analyses were done with R statistical software. Further details for the statistical methods can be found in the appendix (pp 7–8).

Role of the funding source

The funders of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all

the data in the study and had final responsibility for the decision to submit for publication.

Results

Between Feb 1, 2013, and June 28, 2019 (Barcelona), and between June 1, 2009, and Dec 31, 2014 (Cambridge), we recruited participants with Down syndrome and euploid controls (appendix p 3). We recruited and included 388 adults with Down syndrome (347 in Barcelona and 41 in Cambridge) and 242 euploid controls (recruited in Barcelona) in our study. 174 (45%) of 388 participants with Down syndrome and 162 (67%) of 242 controls were women (table). 308 participants with Down syndrome were screened for trisomy 21; genetic confirmation of trisomy 21 was not available for 80 participants. Of participants with Down syndrome, 72 (19%) had mild intellectual disability, 175 (45%) had moderate, and 98 (25%) had severe or profound. The number of adults with Down syndrome and controls was different for each biomarker method (appendix p 9).

We observed no clinical evidence of dementia in 257 (66%) of the 388 participants with Down syndrome. Of the remaining, 48 (12%) had prodromal Alzheimer's disease and 83 (21%) had Alzheimer's disease dementia. The median age of diagnosis was 50·2 years (IQR 47·5–54·1) for prodromal Alzheimer's disease and

53·7 years (49·5–57·2) for Alzheimer's disease dementia. The prevalence of symptomatic Alzheimer's disease (prodromal Alzheimer's disease and Alzheimer's disease dementia combined) increased exponentially from age 40 years onwards, reaching 90–100% prevalence by the end of the seventh decade of life (figure 1). The evolution of the different biomarkers along the Alzheimer's disease continuum (asymptomatic, prodromal Alzheimer's disease, and Alzheimer's disease dementia) is shown in the appendix (p 11).

We plotted the scores on the CAMCOG scale as a function of age in individuals with mild and moderate intellectual disability separately (figure 1). Although neuropsychological performance cannot be directly compared between individuals with Down syndrome and controls, a visual inspection showed a decline in CAMCOG scores starting at about age 40 years in individuals with Down syndrome, particularly in participants with moderate intellectual disability, which is 10 years before the median age of diagnosis of prodromal Alzheimer's disease (figure 1).

We compared the pattern of cortical atrophy in individuals with Down syndrome who had symptomatic Alzheimer's disease with that in individuals who were asymptomatic (figure 2). Compared with participants who were asymptomatic, those who were symptomatic showed

	Asymptomatic (n=257)	Prodromal Alzheimer's disease (n=48)	Alzheimer's disease dementia (n=83)	Controls (n=242)	p value
Age, years	38·7 (31·1 to 48·2)	50·2 (47·5 to 54·1)	53·7 (49·5 to 57·2)	56·6 (50·4 to 63·8)	<0·0001
Sex	..	..	..	..	<0·0001
Men	144 (56%)	26 (54%)	44 (53%)	80 (33%)	NA
Women	113 (44%)	22 (46%)	39 (47%)	162 (67%)	NA
APOE-ε4 carriers	46 (19%)	11 (23%)	15 (20%)	67 (29%)	0·10
Level of intellectual disability	..	..	..	..	<0·0001
Mild	62 (27%)	6 (14%)	4 (5%)	NA	NA
Moderate	114 (50%)	19 (45%)	42 (56%)	NA	NA
Severe or profound	52 (22%)	17 (40%)	29 (38%)	NA	NA
CAMCOG-DS score (n=225)					
Mild (n=67)	88 (82 to 91)	80 (76 to 82)	70 (66 to 75)	NA	<0·0001
Moderate (n=158)	75 (65 to 82)	65 (60 to 73)	41 (18 to 55)	NA	<0·0001
CSF $A\beta_{1-42/1-40}$ (n=265)	0·078 (0·062 to 0·093)	0·042 (0·046 to 0·050)	0·046 (0·042 to 0·055)	0·106 (0·101 to 0·111)	<0·0001
CSF NFL (n=230)	368·7 (233·7 to 523·1)	667·4 (484·0 to 1013·6)	1139·2 (757·3 to 1436·3)	400·0 (338·6 to 544·6)	<0·0001
CSF p-tau (n=272)	30·0 (18·5 to 59·0)	110·4 (67·3 to 186·2)	155·3 (100·4 to 192·8)	32·5 (26·1 to 43·7)	<0·0001
Plasma $A\beta_{1-42}$ (n=435)	14·2 (12·2 to 16·4)	14·1 (11·9 to 15·8)	14·5 (12·5 to 17·0)	9·0 (7·7 to 10·1)	<0·0001
Plasma NFL (n=431)	6·0 (4·6 to 10·3)	13·3 (10·7 to 18·5)	22·3 (17·2 to 30·0)	4·4 (3·3 to 6·2)	<0·0001
Plasma total tau (n=427)	2·5 (2·0 to 3·1)	3·3 (2·4 to 4·0)	4·0 (2·9 to 5·0)	2·2 (1·7 to 3·4)	<0·0001
Centiloid amyloid PET (n=108)*	6·4 (2·0 to 15·7)	64·7 (44·9 to 84·8)	72·6 (53·6 to 97·8)	–1·9 (–10·9 to 4·4)	<0·0001
FDG SUVR (n=197)	1·3 (1·3 to 1·4)	1·2 (1·1 to 1·3)	0·9 (0·8 to 1·1)	1·4 (1·3 to 1·5)	<0·0001
Adjusted hippocampal volume 3 Tesla MRI (n=330)†	–0·14 (–0·50 to 0·27)	–0·87 (–1·50 to –0·59)	–1·86 (–2·37 to –1·03)	0·86 (0·35 to 1·40)	<0·0001

Data are n (%) or median (IQR). The n values on each row are the total number of assessments, including for both individuals with Down syndrome and controls. Percentages of APOE-ε4 carriers and level of intellectual disability were calculated according to the total of patients with available data in each group. CAMCOG-DS=Cambridge Cognitive Examination for Older Adults with Down Syndrome. $A\beta_{1-40}$=amyloid β peptide 1–40. $A\beta_{1-42}$=amyloid β peptide 1–42. CSF=cerebrospinal fluid. NFL=neurofilament light chain. p-tau=tau phosphorylated at threonine 181. FDG=[18]F-fluorodeoxyglucose. NA=not applicable. SUVR=standardised uptake value ratio. *n=74 with ^{18}F-florbetapir and n=34 with ^{11}C-Pittsburg compound B. †With MRI.

Table: Clinical, cognitive, imaging, and biochemical markers in individuals with Down syndrome and controls
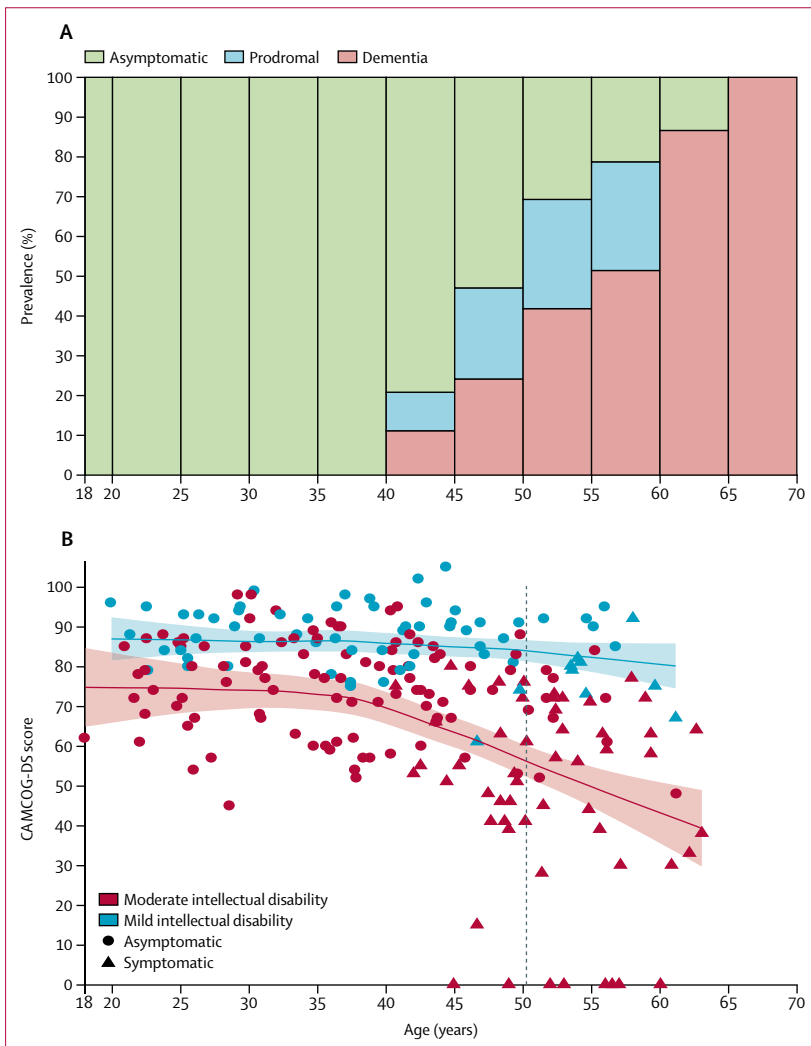
Figure 1: Clinical and neuropsychological changes with age in adults with Down syndrome
(A) Point prevalence of Alzheimer's disease dementia, prodromal Alzheimer's disease, and no clinical symptoms of Alzheimer's disease in individuals with Down syndrome. (B) Age-related changes in CAMCOG-DS scores in individuals with Down syndrome with mild or moderate intellectual disability (analysed separately), with bands representing 95% CIs. The vertical dashed line at 50·2 years represents the median age at the expected symptom onset (ie, prodromal Alzheimer's disease diagnosis). CAMCOG-DS=Cambridge Cognitive Examination for Older Adults with Down Syndrome.
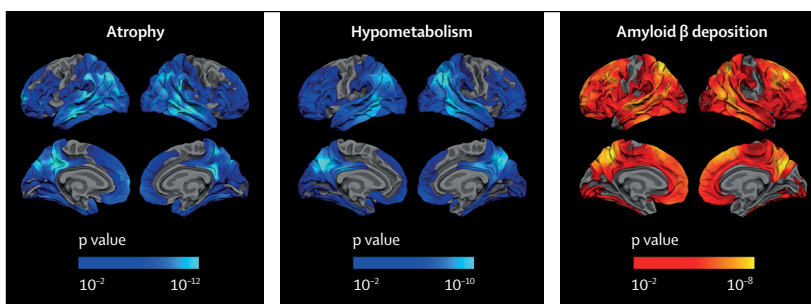
Figure 2: Imaging signature of Alzheimer's disease in adults with Down syndrome for brain structure, glucose metabolism, and amyloid β deposition
All panels compare individuals with Down syndrome with symptomatic Alzheimer's disease with those with no clinical evidence of dementia. The left panel represents loss of cortical thickness, the middle panel loss of brain glucose metabolism, and the right panel the increase in cerebral fibrillar amyloid β deposition.

decreased cortical thickness in the temporoparietal, precuneus-posterior cingulate, and frontal areas.

In the assessment of hippocampal atrophy with age, adults with Down syndrome had smaller hippocampi across the whole lifespan than did controls (figure 3G). A visual inspection of the data showed a steeper decline in hippocampal volumes at the beginning of the fifth decade of life in participants with Down syndrome, about 10 years before expected symptom onset.

A comparison of the pattern of cortical hypometabolism in individuals who were symptomatic with those without symptoms showed decreased glucose metabolism in the temporoparietal, precuneus-posterior cingulate, and frontal areas in participants who were symptomatic (figure 2). We assessed the trajectory with age of the cerebral glucose metabolism as measured in the Landau signature, which tracks early changes in sporadic Alzheimer's disease (figure 3H).[26] Visually, brain metabolism decreased in participants with Down syndrome starting from the earliest observed age. We observed a substantial further decrease in cerebral metabolism at age 37·5 years (12·7 years before expected symptom onset).

We compared the pattern of Aβ deposition in individuals who were symptomatic with that of individuals without symptoms (figure 2). Participants who were symptomatic showed increased global cerebral Aβ deposition, with a relative sparing in sensory and motor areas. Amyloid PET uptake started to increase in the latter half of the fourth decade of life, about 12–15 years before expected symptom onset (figure 3I). Notably, the curves for ^{11}C-Pittsburg compound B and ^{18}F-florbetapir followed similar trajectories and had overlapping CIs in the whole age span (appendix p 6).

Conversely, CSF $Aβ_{1-42/1-40}$ values decreased in individuals with Down syndrome starting from the earliest observed ages (third decade of life) and were significantly different from those of controls from age 28 years onwards (22 years before prodromal Alzheimer's disease diagnosis), levelling off in the sixth decade of life (figure 3A). CSF concentrations of p-tau and NFL showed similar trajectories: visually, the concentrations of both biomarkers started increasing in the fourth decade of life in individuals with Down syndrome (figures 3B, 3C). The CIs no longer overlapped for p-tau at age 39 years and for NFL at age 40 years (11–10 years before prodromal Alzheimer's disease diagnosis). Total tau concentrations in participants with Down syndrome evolved similarly to p-tau (appendix p 10), showing differences at age 36 years.

Plasma $Aβ_{1-42}$ concentrations were 58% higher in adults with Down syndrome than in controls across the whole age span (figure 3D) and did not differ between diagnostic groups (plasma $Aβ_{1-42/1-40}$ concentrations are shown in the appendix [p 10]). Plasma NFL concentrations also increased visually from the earliest observed ages and were significantly different at age 30 years (20 years before prodromal Alzheimer's disease
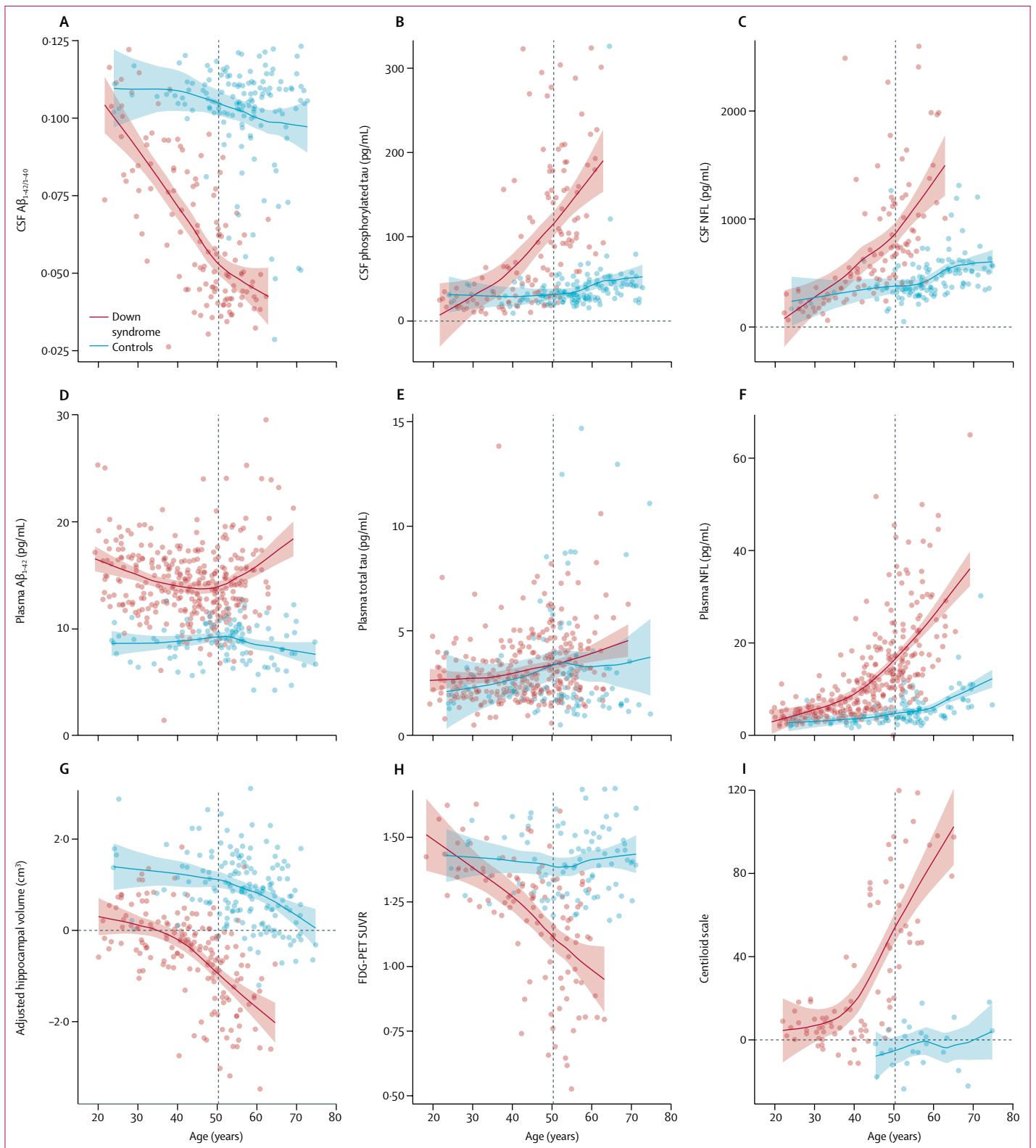
***Figure 3*: Biomarker changes with age in adults with Down syndrome and control participants**
Shading represents 95% CIs. The vertical dashed lines at 50·2 years represent the age at the expected symptom onset (eg, median age of prodromal Alzheimer's disease diagnosis). Aβ$_{1-40}$=amyloid β peptide 1–40. Aβ$_{1-42}$=amyloid β peptide 1–42. CSF=cerebrospinal fluid. FDG=^{18}F-fluorodeoxyglucose. NFL=neurofilament light chain. SUVR=standardised uptake value ratio.
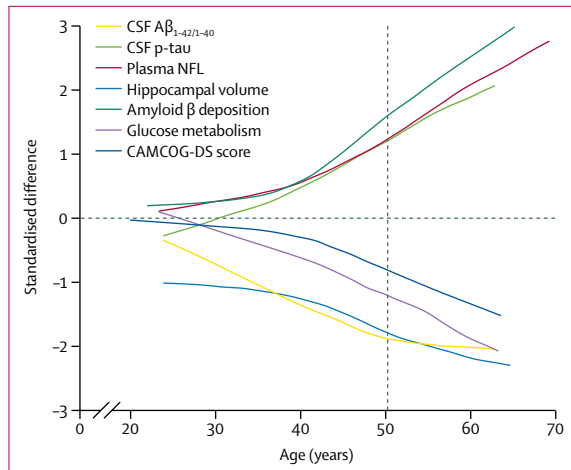
***Figure 4:* Integrated model of the natural history of Alzheimer's disease in individuals with Down syndrome**
Comparison of structural, metabolic, and biochemical biomarker changes as a function of age by use of the standardised differences between participants with Down syndrome and controls fitted with a locally estimated scatterplot smoothing curve. Positive standardised differences represent higher biomarker values in participants with Down syndrome than in euploid controls, and negative values represent lower biomarker values. The vertical dashed line at 50·2 years represents the age at the expected symptom onset (median age of prodromal Alzheimer's disease diagnosis). $A\beta_{1-40}$=amyloid β peptide 1–40. $A\beta_{1-42}$=amyloid β peptide 1–42. CAMCOG-DS=Cambridge Cognitive Examination for Older Adults with Down Syndrome. CSF=cerebrospinal fluid. NFL=neurofilament light chain. p-tau=tau phosphorylated at threonine 181.

diagnosis; figure 3F). Plasma tau concentrations in participants with Down syndrome and controls overlapped widely (figure 3E). The group comparisons for all the biomarkers along the Alzheimer's disease continuum (asymptomatic *vs* prodromal Alzheimer's disease *vs* Alzheimer's disease dementia) are shown in the appendix (p 11).

The changes in cognitive, biochemical, and imaging biomarkers in individuals with Down syndrome expanded for more than 20 years (figure 4). At age 30 years, the early decrease in CSF $A\beta_{1-42/1-40}$ values was accompanied by increases in plasma NFL concentrations, increases in CSF p-tau concentrations, and reductions in brain metabolism. At age 40 years, changes in Centiloid Aβ scale scores and brain atrophy occurred together with cognitive impairment, followed by prodromal Alzheimer's disease and Alzheimer's disease dementia at the beginning of the fifth decade of life in these individuals.

Discussion

To our knowledge, this was the first large multimodal biomarker study to characterise the natural history of Alzheimer's disease in adults with Down syndrome. We found sequential changes in biomarkers over decades, as well as progressive cognitive impairment. In accordance with the conceptualisation of Down syndrome as a form of genetically determined Alzheimer's disease,[5] these changes began more than two decades before the onset of

Alzheimer's disease dementia, in a strikingly similar order and timing to that described in autosomal dominant Alzheimer's disease.[16,18]

Symptomatic Alzheimer's disease prevalence in individuals with Down syndrome increased exponentially with age in our study, reaching 90–100% in the seventh decade of life. The median age of diagnosis of prodromal Alzheimer's disease and clinical Alzheimer's disease dementia in our study are in agreement with a large study that used clinical records in the UK (mean age at dementia onset of 55·8 years, SD 6·3).[29] The variability in dementia onset in both ours and the UK-only study was less than that reported in the Colombian Kindred study (mean age 38·5 years, SD 8·6), and similar to those reported for the other mutations in autosomal dominant Alzheimer's disease.[30] The prevalence estimates for Alzheimer's disease dementia in cross-sectional and longitudinal studies show substantial variability.[1,3] Our study is among those with the highest estimates, probably because of our research protocol with comprehensive neuropsychological and neurological assessments, which allowed us to detect individuals with prodromal Alzheimer's disease.

Early clinicopathological studies and recent biomarker studies have shown that Alzheimer's disease pathology and CSF and plasma biomarker changes in individuals with Down syndrome are qualitatively the same as in sporadic Alzheimer's disease.[6,14] On one hand, the patterns of cerebral Aβ deposition, atrophy, and hypometabolism in our study are similar to those found in previous reports, indicating that Alzheimer's disease in individuals with Down syndrome targets the same cortical regions affected in the sporadic and autosomal dominant forms.[9] On the other hand, cognitive performance and other biomarkers, such as plasma $A\beta_{1-42}$ concentrations or hippocampal volumes, had clear different starting points in the youngest individuals (and throughout all ages). These abnormalities, which have been consistently reported in the literature,[9,31] underscore the importance of considering the neurodevelopmental differences in individuals with Down syndrome and the level of baseline intellectual disability when interpreting cognitive and biomarker results.

In our study, the CSF $A\beta_{1-42/1-40}$ ratios and plasma NFL concentrations were the first biomarkers to change by age 28–30 years, more than 20 years before prodromal Alzheimer's disease diagnosis. Fibrillar amyloid deposition was not detectable with PET until almost 10 years after the $A\beta_{1-42/1-40}$ and NFL changes. This onset is similar to those reported in DIAN[16] or in the Colombian E280A PSEN1 kindred[18] studies and is in agreement with evidence showing the reduced sensitivity of amyloid PET in the earliest stages of amyloid deposition in individuals with Down syndrome.[32] Additionally, the change in CSF $A\beta_{1-42}$ concentrations might occur earlier than reported in this study because, at younger ages, both children with Down syndrome[33] and children with

a presenilin-1 mutation[16,34] have shown higher CSF $A\beta_{1-42}$ concentrations than those of age-matched controls. Changes in brain metabolism in individuals with Down syndrome was the biomarker that followed a more linear decrease, compared with that of other biomarkers, and we observed significant differences at about age 37·5 years, nearly 13 years before prodromal Alzheimer's disease diagnosis. CSF p-tau and NFL concentrations began to increase at about age 40 years, 10 years before prodromal Alzheimer's disease diagnosis, similar to autosomal dominant Alzheimer's disease.[16] Hippocampal atrophy was evident from an early age, probably reflecting neurodevelopmental differences. Nonetheless, hippocampal volumes decreased faster at about age 40 years, 10 years before prodromal Alzheimer's disease diagnosis, in parallel with a decline in CAMCOG-DS scores. Dementia diagnosis occurred approximately 3 years after prodromal Alzheimer's disease diagnosis. This sequence of changes, summarised in the combined model (figure 4), is again strikingly similar to that described in autosomal dominant Alzheimer's disease.[16]

The main strengths of this study are the large population size, with a wide age range, and the comprehensive and multimodal nature of the assessment. The main limitations are its cross-sectional design and the differences in sample sizes for the different biomarker tests. The unequal sample size of each biomarker affects the CI calculations and the exact estimates of the ages for the biomarker changes. For this reason, and because of the intrinsic limitations of studies assessing the natural history of biomarkers (eg, different nature of the variables with different slopes and different sensitivities of the assays), we used a descriptive analysis, which we complemented with the mathematical approach previously used in studies of autosomal dominant Alzheimer's disease.[16] This limitation is clearly exemplified by the differences between the estimates for plasma and CSF NFL concentrations. The neurodevelopmental and biological differences associated with Down syndrome also complicate the determination of the estimates, as exemplified in the cognitive performance assessment and in other biomarkers, such as hippocampal volumes or plasma $A\beta_{1-42}$ concentrations. Finally, as in previous studies in individuals with Down syndrome,[29] we used age at diagnosis as proxy of age of onset, but we acknowledge that this approach might have underestimated age of onset.

Our results have several important implications. First, they support the concept of Down syndrome as a form of genetically determined Alzheimer's disease,[5] which has been shown to have a profound effect in patient management and family counselling for autosomal dominant Alzheimer disease. Second, neurodegenerative changes, as measured by plasma NFL increases or brain hypometabolism, might occur much earlier than previously thought in individuals with Down syndrome,

even before fibrillar Aβ deposition is detectable by PET. These results reinforce the adequacy of the unbiased descriptive classification scheme for Alzheimer's disease biomarkers of the AT(N) system.[19] Third, our results showed that the biochemical changes in their direction and magnitude are similar to those described in sporadic and autosomal dominant Alzheimer's disease. The atrophy, hypometabolic, and Aβ deposition maps were also similar to those described in the sporadic and autosomal dominant forms of the disease. Finally, our finding of a long preclinical phase supports the consideration of people with Down syndrome as a suitable population for clinical trials of Alzheimer's disease.

Regrettably, individuals with Down syndrome have yet to be included in preventive clinical trials. Such trials would, admittedly, pose additional challenges compared with those done in the general population regarding informed consent and concerns about the feasibility of completing all assessments. However, our study shows that a substantial proportion of adults with Down syndrome are capable and willing to do all the multimodal studies required in a trial. Clinical trials in this population have obvious advantages: the ultra-high risk for developing symptomatic Alzheimer's disease and, as we showed here, a predictable sequence of events make this population ideal for preventive trials in Alzheimer's disease, together with individuals with its autosomal dominant form. Notably, Down syndrome is much more common than autosomal dominant Alzheimer's disease. Given the similarities between both sporadic and autosomal dominant Alzheimer's disease and Alzheimer's disease in individuals with Down syndrome, such therapies could prove beneficial for all of these groups.[35] Future studies should assess the effects of potential genetic modifiers (including the *APOE* haplotype and polygenic risk factors), the potential differences between individuals with full trisomy 21 and those with partial translocations or mosaicism and, importantly, the relationships between biomarkers. The different biomarkers do indeed show correlations, and multimodal studies are certainly needed to assess the effect of pathophysiological biomarkers on topographical biomarkers.[5] Additional studies specifically investigating cognition are also needed to assess whether the differences in the CAMCOG-DS rate of decline in individuals with mild versus moderate intellectual disability are due to insufficient test sensitivity for highly functioning individuals, differences in underlying biology supportive of the cognitive reserve hypothesis, or the population composition in our study.

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