Structural connectivity relates to risk of dementia in the general population: evidence for the disconnectivity hypothesis

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ABSTRACT

Background The 'disconnectivity hypothesis' postulates that partial loss of connecting white matter fibers between brain regions contributes to the development of dementia, including Alzheimer's disease (AD). Using diffusion-magnetic resonance imaging (MRI) to quantify global and tract-specific white matter microstructural integrity, we tested this hypothesis in a longitudinal population-based setting.

Method Global and tract-specific measures of fractional anisotropy (FA) and mean diffusivity (MD) were obtained in 4,415 non-demented persons (mean age 63.9 years (SD 11.0), 55.0% women) from the prospective population-based Rotterdam Study with brain MRI (2005-2011). We modelled the association of these diffusion measures with risk of dementia (follow-up until 2016), and with changes on repeated cognitive assessment over 5.4 years of follow-up, adjusting for age, sex, education, macrostructural MRI markers, depressive symptoms, cardiovascular risk factors, and *APOE* genotype.

Findings During a mean follow-up of 6.7 years, 101 participants had incident dementia, of whom 83 had AD. Lower global values of FA and higher values of MD associated with an increased risk of dementia (adjusted hazard ratio (95% confidence interval) per standard deviation increase in MD: 1.79 (1.44-2.23)). Similarly, lower global values of FA and higher values of MD related to more cognitive decline in non-demented individuals (difference in global cognition per standard deviation increase in MD (95% CI) was: -0.04 (-0.07;-0.01)). Associations were most profound in the projection, association and limbic system tracts.

Interpretations Structural disconnectivity is associated with and an increased risk of dementia and more pronounced cognitive decline in the general population.

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INTRODUCTION

Dementia is among the leading causes of death and disability worldwide, and its socioeconomic burden on society will continue to increase as the number of persons suffering from dementia is predicted to nearly triple to 131.5 million in 2050.[1] Effective preventive and curative interventions are urgently needed, but their development and timely application is hampered by incomplete understanding of pathophysiology, lack of markers that can identify changes in the very early, subclinical stages of disease and lack of prognostic markers. Subclinical brain changes are thought to occur years, if not decades, prior to onset of clinical symptoms,[2] which is beyond the scope of currently applied subclinical macrostructural imaging markers of neurodegeneration, such as hippocampal volume and presence of white matter hyperintensities.

One of the recent insights in dementia is that brain damage can lead to disruption of brain networks, so called disconnectivity.[3-5] Disconnectivity, which can be investigated using diffusion-magnetic resonance imaging (MRI), seems to occur prior to changes in conventional structural MRI markers such as white matter hyperintensities load in dementia,[6] and is thought to reflect early cerebral white matter damage.[7,8] Previous studies have shown more pronounced disconnectivity both in patients with dementia compared to healthy controls,[9,10] and in relation to more rapid progression in patients with Alzheimer's disease (AD).[11] However, evidence from longitudinal studies for a role of preclinical disconnectivity in the development of dementia is scarce. Two clinical studies among patients with small vessel disease found that network disruption increases the risk of dementia,[12,13] but these studies were hampered by limited precision (observing 18 and 32 cases of dementia, respectively), and it remains undetermined whether findings extend to the wider population without prior TIA or stroke.

Therefore, we aimed to test the disconnectivity hypothesis by investigating whether global and tract-specific disconnectivity are associated with dementia and cognitive decline in a population-based setting.

METHODS

Study population

This study was embedded within the Rotterdam Study, a population-based cohort study including participants of 45 years and older living in Ommoord, a suburb of Rotterdam.[14] The study started in 1990 with 7,983 participants and was extended with 3,011 participants in 2000 and with 3,932 participants in 2006. Participants were examined at baseline with a home interview and an extensive set of examinations in the research center. Follow-up examinations were repeated every 3-4 years. Also, all participants were continuously monitored through electronic linkage of the study

database with their own medical records. All details of the study have been previously described.[14] From 2005 onwards, MRI-scanning was implemented in the core protocol. Between 2005 and 2011, 5,715 participants without contraindications for MRI (metal implants, pacemaker, claustrophobia) were eligible for scanning, of whom 4,888 (86%) underwent a multi-sequence MRI acquisition of the brain, and 4,813 (98%) participants completed the diffusion-weighted sequences. We excluded 245 individuals due to technical scanning issues, e.g. failed segmentations, as well as 38 participants with prevalent dementia and 100 participants with insufficient dementia screening at baseline, resulting in a study sample of 4,430 individuals. Of these individuals, 4,317 persons had detailed cognitive assessment at baseline and 3,402 (79%) had repeated assessment during follow-up examination after on average 5.4 (SD 0.6) years. The Rotterdam Study has been approved by the medical ethics committee according to the Population Study Act Rotterdam Study, executed by the Ministry of Health, Welfare and Sports of the Netherlands. All participants gave written informed consent.

MRI acquisition and processing

Multi-sequence MR imaging was performed on a 1.5 tesla MRI scanner (GE Signa Excite). The imaging protocol has been described extensively elsewhere.[15] The conventional scan protocol consisted of a T1-weighted image, a T2-weighted fluid-attenuated inversion recovery (FLAIR) sequence, and a proton density weighted image.

Scans were spatially co-registered using rigid registration. Scans were segmented with an automated tissue segmentation approach into grey matter, white matter, cerebrospinal fluid (CSF) and background tissue,[16,17] followed by a white matter hyperintensity (WMH) segmentation based on the tissue segmentation and the FLAIR image.[18] Supratentorial intracranial volume (ICV), to correct for head size, was estimated by summing total grey and white matter and CSF volumes.[17] We visually assessed the presence of infarcts on conventional MRI sequences, and in case of involvement of cortical grey matter we classified these as cortical infarcts.

Diffusion-MRI processing and tractography

For diffusion-MRI, we performed a single shot, diffusion-weighted spin echo echo-planar imaging sequence. Maximum b-value was 1000 s/mm^2 in 25 non-collinear directions; three volumes were acquired without diffusion weighting (b-value = 0 s/mm^2). All diffusion data were pre-processed using a standardized pipeline.[19] In short, eddy current and head-motion correction were performed on the diffusion data. The resampled data was used to fit diffusion tensors to compute mean fractional anisotropy (FA) and mean diffusivity (MD) in the normal-appearing white matter, through combination with the tissue segmentation. The diffusion data was also used to segment white matter tracts using a diffusion tractography approach described previously.[20]

We segmented 15 different white matter tracts (12 bilateral, 3 singular) and obtained mean FA and MD in these tracts, with subsequent combination of left and right measures.[20] In general, lower FA and higher MD values are considered indicative of lower microstructural integrity and as such reflecting disconnectivity. Missing data for tract-specific measurements due to tractography or segmentation failures were limited to 33-78 participants (0.8-1.8%) per tract. Tracts were categorized, based on anatomy or presumed function, into brainstem tracts (middle cerebellar peduncle, medial lemniscus), projection tracts (corticospinal tract, anterior thalamic radiation, superior thalamic radiation, posterior thalamic radiation), association tracts (superior longitudinal fasciculus, inferior, longitudinal fasciculus, inferior fronto-occipital fasciculus, uncinated fasciculus), limbic system tracts (cingulate gyrus part of cingulum, parahippocampal part of cingulum) and callosal tracts (forceps major, forceps minor).[20]
We obtained tract volumes and tract WMH volumes by combining the tissue and tract segmentations. Tract-specific WMH volumes were natural-log transformed, to account for their skewed distribution.
Between February 2007 and May 2008, an erroneous swap of the phase and frequency encoding directions for the diffusion acquisition led to a mild ghosting artifact, which was addressed by adjustment in the analysis.[20] There was only partial coverage of one of the brainstem tracts (medial lemniscus) due to incomplete coverage of the cerebellum in the field of view, and we used alternative seed masks for tractography and adjustment in the model to overcome this problem.[20]

Dementia screening and surveillance
All participants were screened for dementia at baseline and during subsequent center visits using the Mini-Mental-State Examination (MMSE) and the Geriatric Mental Schedule (GMS) organic level.[21] Participants with an MMSE score <26 or a GMS score >0 underwent further cognitive examination and informant interview, including the Cambridge Examination for Mental Disorders of the Elderly. In addition, the entire cohort was under continuous surveillance for dementia through electronic linkage of the study database with medical records from general practitioners and the regional institute for outpatient mental health care. Clinical neuroimaging was used when required for dementia subtype diagnosis. A consensus panel led by a consultant neurologist established the final diagnosis in accordance with standard criteria for dementia (DSM-III-R) and Alzheimer's disease (NINCDS-ADRDA). Follow-up until 1st January 2016 was virtually complete (96% of potential person years). Participants were censored at date of dementia diagnosis, death, loss to follow-up, or 1st January 2016, whichever came first.

Assessment of cognitive function

During center visits, all participants underwent routine cognitive assessment comprising a word fluency test (number of animal species within 1 minute), 15-word learning test (immediate and delayed recall of 15 items), letter-digit substitution task (LDST, number of correct digits in 1 minute), Stroop test (error adjusted time in seconds taken for completing the reading, colour naming, and interference tasks), and the Purdue Pegboard task for manual dexterity.[21] To obtain a composite measure of test performance, we calculated the G-factor by principal component analysis,[21] which explained 49-54% of variance in cognitive test scores at each examination round in our population. For each participant, Z-scores were calculated for each test separately, by dividing the difference between individual test score and population mean by the population standard deviation. Scores for the Stroop tasks were inverted such that higher scores indicated better performance.

Other measurements

Information on smoking habits, educational attainment, and use of antihypertensive and lipid-lowering medication was ascertained at baseline by structured questionnaires. Blood pressure was measured twice in sitting position using a random-zero sphygmomanometer and the mean of two readings was used in the analyses. Total serum cholesterol and high-density lipoprotein (HDL) cholesterol were determined in fasting blood samples. Presence of type 2 diabetes at baseline was determined on the basis of fasting serum glucose level (>= 7.0 mmol/l) or, if unavailable, non-fasting serum glucose level (>=11.1 mmol/l) or the use of anti-diabetic medication.[22] Body-mass index (BMI) was calculated, dividing weight in kilograms by the squared height in meters. History of stroke was assessed by interview, and verified in medical records, and participants were continuously monitored for incident stroke through computerized linkage of medical records from general practitioners and nursing home physicians with the study database. We used the validated Dutch version of the Center for Epidemiology Depression Scale (CES-D) for assessment of depressive symptoms.[23]
APOE genotype was determined using polymerase chain reaction on coded DNA samples (original cohort), and using a bi-allelic TaqMan assay (rs7412 and rs429358; expansion cohort). In 179 participants with missing *APOE* status from this blood sampling, genotype was determined by genetic imputation (Illumina 610K and 660K chip; imputation with Haplotype Reference Consortium reference panel [v1.0] with Minimac 3).

Statistical analysis

Analyses included all eligible participants, with the exception of 15 participants whose diffusion measures deviated >7 standard deviations from the mean, leaving 4,415

participants for analysis. We used Cox proportional hazard models to determine the association of global and tract-specific diffusion-MRI measures (FA and MD) with incident dementia. The proportional hazard assumption was met. We assessed risk of dementia per standard deviation increase in FA and MD. We repeated the analyses 1) for Alzheimer disease only, 2) after excluding participants with prevalent stroke while censoring at time of incident stroke, 3) excluding persons with MRI-defined, subclinical cortical infarcts at baseline, and 4) stepwise excluding the first five years of follow-up from the analysis.

We then determined the association of global and tract-specific diffusion-MRI measures with change in cognitive performance using linear regression models. Cognitive test scores at follow-up were adjusted for baseline cognitive test results. These analyses were repeated after exclusion of all participants who developed dementia during follow-up. All models were adjusted for age, sex, education, intracranial volume, white matter volume, and the log-transformed volume of white matter hyperintensities and the correction for swapping gradients and varying field of view (model I), and additionally for education, depressive symptoms (CES-D score), and cardiovascular risk factors (systolic blood pressure, diastolic blood pressure, antihypertensive medication, serum cholesterol, HDL-cholesterol, lipid-lowering medication, diabetes, smoking, and BMI), and *APOE*-ε4 allele carriership (model II).

For the tract-specific analyses, we corrected the p-value (alpha level of 0.05) for multiple comparisons with the number of independent tests on the basis of the variance of the eigenvalues of the correlation matrix of all 30 variables used in the main analysis (i.e. FA and MD for the 15 tracts). The following formula was used: M_{eff}= 1 +(M-1)(1-var (λ_{obs})/M) in which M is the number of variables, λ_{obs} is the variance of the eigenvalues of the correlation matrix,and M_{eff} is the number of independent variables.[24,25] This resulted in an M_{eff} of 17.45, which then, using the Šidák formula (α sidak = 1 - ((1 - α)^(1/M_{eff}))), translated into a significance level of $p<0.0029$ for the tract-specific analyses with dementia as outcome.[24]

For the analyses assessing global diffusion-MRI measures with the separate cognitive tests as outcome, the above-mentioned method generated a significance level of $p<0.008$. For analyses assessing tract-specific diffusion-MRI measures with global cognition and separate cognitive tests as outcome, the adjusted p-value was $p<0.0022$. All analyses were carried out using SPSS Statistics 21.0 (IBM Corp, Armonk, NY, USA) or R version 3.0.3 (packages “GenABEL”, “survival, “stargazer” and “data. table”).

RESULTS

Table 1 presents the baseline characteristics of the study population. Mean age of the 4,415 participants was 63.9 years (SD ±11.1 years), and 55.0% were women. During a median follow-up of 6.8 years (IQR 5.8-8 .0 years), 101 persons developed dementia, of whom 83 had AD.

Table 1. Population characteristics

Characteristic	N= 4,415
Age, years	63.9 (±11.0)
Female	2,426 (55.0 %)
Smoking	
Never	1367 (31.0%)
Former	2120 (48.0%)
Current	928 (21.0%)
Education	
Lower education	1266 (28.7%)
Middle education	2107 (47.7%)
Higher education	1042 (23.6%)
Systolic blood pressure, mmHg	140.0 (±21.5)
Diastolic blood pressure, mmHg	83.2 (±10.9)
Antihypertensive medication	1573 (35.6)
Total cholesterol, mmol/L	5.5 (±1.1)
HDL cholesterol, mmol/L	1.5 (±0.4)
Lipid-lowering medication	1113 (25.2%)
Diabetes mellitus	531 (12.0%)
BMI, kg/m^2	27.4 (±4.1)
Center for Epidemiology Depression Scale score	8 (2-12)
APOE-ε4 carriership	1216 (28.3%)
Mean FA	0.34 (±0.02)
Mean MD	0.74 (±0.03)
Intracranial volume, mL	1142.0 (±116.4)
White matter volume, mL	409.3 (±60.7)
White matter hyperintensities volume, mL	2.90 (1.6-6.3)

Continuous variables are presented as means (±standard deviations) and categorical variables as n (percentages), except for white matter hyperintensities volume and Center for Epidemiology Depression Scale score, which are presented as median (inter-quartile range).

Abbreviations: N:number of participants, HDL:high-density lipoprotein, BMI: body mass index, s;seconds, FA:fractional anisotropy, MD:mean diffusivity x 10^{-3} mm^2/sec, mL: milliliter.

Lower microstructural integrity, reflected in lower values of global FA and higher values of global MD was associated with a higher risk of dementia (fully adjusted hazard ratio [HR], 95% confidence interval (CI), per SD increase in FA: 0.65 (0.52-0.80), and for MD: 1.79 (1.44-2.23) (**Figure 1, Supplementary Table S1**). Results were similar for AD only, and unaltered after excluding participants with prevalent stroke while censoring at time of incident stroke, or excluding participants with subclinical MRI-defined cortical infarcts (**Figure 1, Supplementary Table S1**). Stepwise exclusion of the first five years of follow-up from the analysis did not alter the risk estimates (**Figure 2, Supplementary Table S2**).

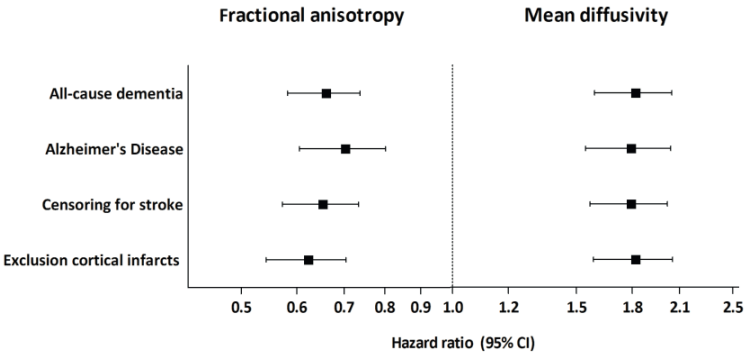

Figure 1. Global mean microstructural integrity and incident dementia

In tract-specific analyses, the strongest associations with dementia risk were observed for MD in the projection tracts, association tracts and limbic system tracts (per SD increase HR of 2.35 (1 .53;3.62) for the superior thalamic radiation, 1.79 (1.36;2.37) for the inferior fronto-occipital fasciculus, and 1.62 (1.41;1.86) for the parahippocampal part of the cingulum respectively (**Table 2, Figure 3**)). Similarly, lower FA in the association tracts and in the limbic system tracts were most profoundly associated

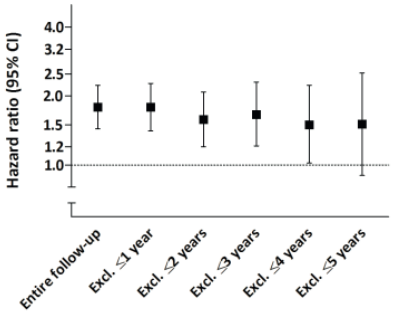

Figure 2. Global mean diffusivity and incident dementia, with stepwise exclusion of the first five years of follow-up

with a higher risk of dementia (per SD increase HR 0.59 (0.45;0.76) for the uncinated fasciculus and HR 0.67 (0.53;0.84) for the parahippocampal part of the cingulum, respectively, in the fully adjusted model (**Table 2**)).

Table 2. Tract-specific white matter microstructure and incident dementia

	Fractional Anisotropy		*Mean Diffusivity*	
White matter tracts	**Model I**	**Model II**	**Model I**	**Model II**
Brainstem tracts				
Middle cerebellar peduncle	1.05 (0.85;1.30)	1.08 (0.87;1.35)	1.05 (0.85;1.30)	1.04 (0.83;1.30)
Medial lemniscus	1.09 (0.86;1.39)	1.11 (0.86;1.44)	1.06 (0.88;1.28)	1.06 (0.87;1.29)
Projection tracts				
Corticospinal tract	1.17 (0.95;1.44)	1.19 (0.96;1.47)	**1.52 (1.13;2.06)**	**1.52 (1.11;2.08)**
Anterior thalamic radiation	0.85 (0.66;1.09)	0.87 (0.67;1.13)	**1.68 (1.23;2.30)***	**1.73 (1.26;2.38)***
Superior thalamic radiation	1.17 (0.95;1.45)	1.20 (0.97;1.50)	**2.29 (1.49;3.52)***	**2.35 (1.53;3.62)***
Posterior thalamic radiation	**0.69 (0.52;0.90)**	**0.74 (0.56;0.97)**	**1.41 (1.15;1.72)***	**1.42 (1.15;1.75)***
Association tracts				
Superior longitudinal fasciculus	0.77 (0.60;1.00)	0.79 (0.60;1.04)	**1.65 (1.30;2.11)***	**1.65 (1.28;2.14)***
Inferior longitudinal fasciculus	0.79 (0.62;1.01)	0.84 (0.65;1.09)	**1.73 (1.36;2.21)***	**1.69 (1.31;2.18)***
Inferior fronto-occipital fasciculus	**0.66 (0.50;0.86)***	**0.71 (0.53;0.93)**	**1.75 (1.34;2.27)***	**1.79 (1.36;2.37)***
Uncinate fasciculus	**0.60 (0.47;0.77)***	**0.59 (0.45;0.76)***	**1.67 (1.39;2.00)***	**1.73 (1.42;2.10)***
Limbic system tracts				
Cingulate gyrus part of cingulum	0.69 (0.54;0.87)	**0.71 (0.55;0.90)**	**1.55 (1.26;1.92)***	**1.58 (1.26;1.97)***
Parahippocampal part of cingulum	**0.67 (0.54;0.84)***	**0.67 (0.53;0.84)***	**1.61 (1.41;1.85)***	**1.62 (1.41;1.86)***
fornix	**0.76 (0.59;0.99)**	0.78 (0.60;1.02)	1.13 (0.80;1.58)	1.06 (0.75;1.50)
Callosal tracts				
Forceps major	**0.77 (0.59;1.00)**	0.79 (0.61;1.04)	1.15 (0.93;1.41)	1.12 (0.90;1.38)
Forceps minor	0.78 (0.60;1.01)	0.80 (0.61;1.06)	**1.38 (1.12;1.71)**	**1.39 (1.11;1.75)**

Data are presented as Hazard ratios (95% CI) per standard deviation increase of FA and MD. Results in bold were significant at $p<0.05$ and those that were starred * at $p<0.0029$. Model I: adjusted for age, sex, education, intracranial volume, white matter volume and the log transformed white matter lesion volume of the investigated tract. Model II: Model I, and additionally adjusted for CES-D score, and cardiovascular risk factors (systolic blood pressure, diastolic blood pressure, antihypertensive medication, serum cholesterol, HDL-cholesterol, lipid lowering medication, diabetes, smoking, BMI) and *apolipoprotein E-ε4 allele* carriership.
Abbreviations: FA: fractional anisotropy, MD; mean diffusivity, WML: white matter lesion, BMI: Body Mass Index, CES-D: Center for Epidemiology Depression Scale score.

The association between global white matter microstructure and cognitive decline is presented in **Table 3**. Higher values of global MD were associated with greater decline in global cognition, driven by worse performance on the Word Fluency Test and Stroop reading and interference subtasks. Results were unaltered by exclusion of all incident dementia cases (**Supplementary Table S3**). Similar associations, albeit somewhat attenuated, were observed for FA.

Table 3. Global white matter microstructure and cognitive decline

	FA	MD
G-factor	0.02 (-0.004;0.041)	**-0.04 (-0.07;-0.01)**
Immediate memory	-0.002 (-0.04;0.03)	-0.03 (-0.07;0.02)
Delayed memory	0.007 (-0.03;0.04)	-0.03 (-0.07;-0.01)
Stroop 1	**0.04 (0.01;0.07)***	**-0.06 (-0.09;-0.02)***
Stroop 2	0.02 (-0.001;0.05)	-0.02 (-0.05;0.02)
Stroop 3	**0.04 (0.01;0.07)***	**-0.09 (-0.12;-0.05)***
Letter-Digit Substitution Task	0.004 (-0.02;0.03)	-0.004 (-0.04;0.03)
Word Fluency Test	0.03 (0.001;0.06)	**-0.06 (-0.10;-0.02)***
Purdue Pegboard	**0.03 (0.005;0.06)**	-0.04 (-0.07;-0.00)

Data are presented as mean difference in Z-score (95% CI) per standard deviation increase of FA and MD. Results in bold were significant at p<0.05 and those that were starred * at p<0.008.

Model adjusted for age, sex, education, intracranial volume, white matter volume and the log transformed white matter lesion volume, CES-D score, and additionally adjusted for cardiovascular risk factors (systolic blood pressure, diastolic blood pressure, antihypertensive medication, serum cholesterol, HDL-cholesterol, lipid lowering medication, diabetes, smoking, BMI) and *apolipoprotein E-ε4 allele* carriership.

Abbreviations: FA: fractional anisotropy, MD; mean diffusivity, WML: white matter lesion, BMI: Body Mass Index, CES-D score: Center for Epidemiology Depression Scale.

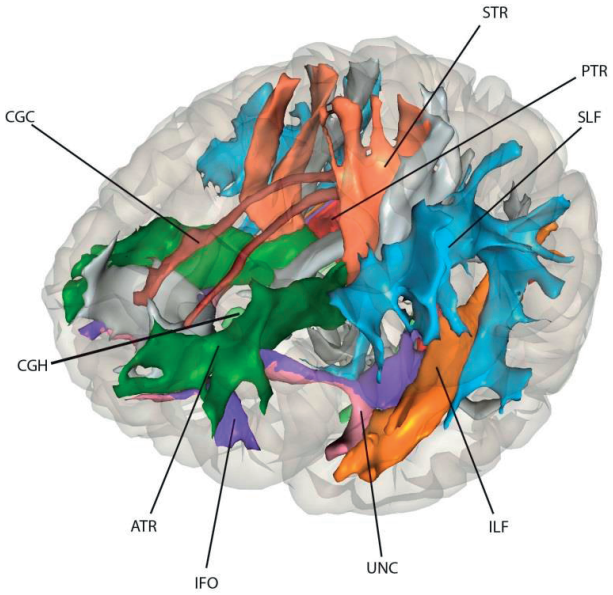

Figure 3. Association of tract-specific diffusion-MRI measures and incident dementia.

Tracts that were significantly associated with dementia risk are color-coded, as represented in the color legend. Other tracts are presented in grey.

Abbreviations:, ATR: anterior thalamic radiation, STR: superior thalamic radiation, PTR: posterior thalamic radiation, SLF: superior longitudinal fasciculus, ILF: inferior longitudinal fasciculus, IFO: inferior-fronto-occipital fasciculus, UNC: uncinate fasciculus, CGC: cingulate gyrus part of cingulum, CGH: parahippocampal part of cingulum.

DISCUSSION

In this longitudinal population-based study, we found that structural disconnectivity is associated with increased risk of dementia and with more pronounced cognitive decline. These associations were most profound for the projection, association and limbic system tracts, and extended into the pre-clinical phase of the disease.

Longitudinal studies provide higher evidence for causal relations. Our main results provide evidence for the 'disconnection hypothesis', which states that loss of brain connections precedes cognitive decline and dementia. In line with this hypothesis, our results suggest that disconnectivity plays a role already in the preclinical stages of dementia. The findings in this study are consistent with previous clinical studies concluding that diffusion-MRI measures in persons with cerebral small vessel disease add in the prediction of cognitive decline and dementia.[12,13] We can now further extend this conclusion to the general population.

Various potential pathways could lead to disconnectivity. A vascular pathway has been proposed in which reduction in white matter perfusion, e.g. due to impaired autoregulation may result in white matter damage.[26] Oligodendrocytes might shrink because of hypoxia and ischaemia in white matter, with subsequent loss of myelin.[27,28] However, in our fully adjusted model we corrected for several cardiovascular risk factors and the estimates did not change substantially. This may be explained by residual confounding (due to age-specific effects of vascular factors or subclinical vascular factors), or a more complex, multifaceted pathway, in which there is a complex interplay of traditional vascular risk factors, hypoxia, and neuro-inflammation.[29] Inflammation-induced disconnectivity may be caused by inflammation-related cytokines (TNF- α, IL-8, IL-10, IFN- γ) and growth factors (IGFBP2, PDGF-BB) which have been associated with a lower integrity of myelin sheaths.[30,31].Yet, reverse causality as an explanation for our findings is very unlikely since the risk estimates did not change after excluding the first five years of follow-up. Also, disconnectivity associated with cognitive decline also in individuals who did not develop dementia during the study duration, suggesting an association already in the preclinical phase of dementia and with normal aging.

We found that structural disconnectivity, indicated by a low FA and high MD throughout the brain, but in particular in the projection, association and limbic system tracts, related to a higher risk of dementia. This is in line with previous research in cross-sectional studies which found lower FA in white matter tracts including the association tracts[32,33] and projection tracts[34,35] associated with dementia. Lower FA values in limbic system tracts (in particular in the parahippocampal cingulum) and the association with dementia, more specifically AD, has been most consistently reported in previous studies.[34,36,37]

A small number of studies reported higher FA values in specific regions in AD.[38,39] This counterintuitive finding may be explained by selective degeneration of a fiber population in regions with crossing white matter tracts, leading to paradoxical higher FA.[40] MD is therefore thought to be a more sensitive and reliable measure in these crossing fiber regions (and therefore also globally),[41] and presumably more sensitive to white matter damage.[9,10] Moreover, in a small group of patients with AD, increases in MD preceded changes in FA which only occurred in a more progressive disease state.[9] Accordingly, in our study we found stronger associations with MD than with FA.

The exact pathological substrate underlying the changes in FA and MD leading to disconnectivity is still unknown. There is pathological evidence that changes in diffusion-MRI measures correlate with myelin damage and axonal count,[42] that myelin is increasingly suggested as an important factor in AD pathology and that myelin breakdown is at the core of the earliest changes involved in dementia.[43] However, the presence of other possible processes such as inflammation generates difficulties to assigning change in diffusion-MRI measures to a specific underlying pathological process causing the observed structural disconnectivity.[44]

Strengths of the study are the population-based setting, the large sample size, the automated publicly available diffusion MRI processing methods that facilitate replication[6], and the longitudinal assessment of cognitive performance with meticulous follow-up for dementia. Some limitations need to be considered. First, the averaging of FA and MD measures over the normal-appearing white matter for analyses discards some spatial information. Second, given the long preclinical phase of dementia, our median follow-up time of 6.8 years is still relatively short, and longer duration studies with repeated imaging are required to further map changes in diffusion-MRI in the process of neurodegeneration. Nevertheless, our results were unaffected by excluding the first five years of follow-up, and independent of macrostructural white matter pathology (white matter hyperintensity volume). Third, although we found associations similar for all-cause dementia and clinical AD, confirmation of subtype diagnosis by biomarkers or pathological examination was not available.

In conclusion, structural disconnectivity increases the risk of dementia and more pronounced cognitive decline. Our study suggests that diffusion-MRI may be useful in risk prediction.

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Supplementary Table S1. Global white matter microstructure and incident dementia

	Model	FA	MD
All dementia (n=101)	model I	**0.65 (0.53;0.80)**	**1.77 (1.43;2.17)**
	model II	**0.65 (0.52;0.80)**	**1.79 (1.44;2.23)**
AD (n=83)	model I	**0.70 (0.55;0.88)**	**1.71 (1.35;2.16)**
	model II	**0.69 (0.54;0.88)**	**1.76 (1.38;2.24)**
Censoring for stroke (n=98)	model I	**0.65 (0.53;0.80)**	**1.75 (1.41;2.16)**
	model II	**0.64 (0.52;0.80)**	**1.76 (1.42;2.20)**
Exclusion cortical infarcts (n=97)	model I	**0.63 (0.51;0.78)**	**1.75 (1.41;2.17)**
	model II	**0.61 (0.49;0.77)**	**1.79 (1.43;2.24)**

Data are presented as Hazard ratios (95% CI) per standard deviation increase of FA and MD. Results in bold were significant at p<0.05.

Model I: adjusted for age, sex, education, intracranial volume, white matter volume and the log transformed white matter hyperintensity volume.

Model II: Model I, and additionally adjusted for CES-D score, cardiovascular risk factors (systolic blood pressure, diastolic blood pressure, antihypertensive medication, serum cholesterol, HDL-cholesterol, lipid lowering medication, diabetes, smoking, BMI), and *apolipoprotein E-ε4 allele* carriership.

Abbrevations: FA: fractional anisotropy, MD; mean diffusivity, BMI: Body Mass Index, CES-D: Center for Epidemiology Depression Scale score, n=number of cases.

Supplementary Table S2. Global white matter microstructure and incident dementia, incrementally excluding the first five years of follow-up

	Excluding ≤ 1 year (n=4374/88)	Excluding ≤ 2 years (n=4305/74)	Excluding ≤ 3 years (n=4224/56)	Excluding ≤ 4 years (n=4131/41)	Excluding ≤ 5 years (n=3872/25)
FA	**0.63 (0.50;0.78)**	**0.70 (0.54;0.89)**	**0.64 (0.48;0.86)**	0.73 (0.52;1.03)	**0.60 (0.39;0.91)**
MD	**1.77 (1.41;2.21)**	**1.56 (1.20;2.04)**	**1.64 (1.21;2.22)**	**1.49 (1.02;2.16)**	1.58 (0.98;2.54)

Data are presented as Hazard ratios (95% CI) per standard deviation increase of FA and MD after excluding the first 5 years of follow-up. Results in bold p<0.05. Adjusted for age, sex, education, intracranial volume, white matter volume and the log transformed white matter hyperintensity volume.

Abbreviations: FA: fractional anisotropy, MD; mean diffusivity, n; sub-population/number of cases.

Supplementary Table S3. Global white matter microstructural integrity and cognitive decline (after exclusion of all incident dementia cases)

	FA	MD
G-factor	0.01 (-0.01;0.04)	**-0.03 (-0.06;-0.001)**
Immediate memory	-0.007 (-0.04;0.03)	-0.01 (-0.06;0.03)
Delayed memory	0.002 (-0.03;0.03)	-0.02 (-0.06;0.02)
Stroop 1	**0.04 (0.01;0.07)***	**-0.06 (-0.09;-0.02)***
Stroop 2	**0.03 (0.003;0.05)**	-0.02 (-0.05;-0.01)
Stroop 3	**0.04 (0.009;0.06)**	**-0.08 (-0.12;-0.04)***
Letter-Digit Substitution Task	0.005 (-0.02;0.03)	-0.004 (-0.04;0.02)
Word Fluency Test	**0.03 (0.002;0.06)**	**-0.06 (-0.10;-0.02)***
Purdue Pegboard	**0.03 (0.002;0.06)**	-0.03 (-0.06;0.009)

Data are presented as mean difference in Z-score (95% CI) per standard deviation increase of FA and MD. Results in bold were significant at $p<0.05$ and those that were starred * at $p<0.008$.

Model adjusted for age, sex, education, intracranial volume, white matter volume and the log transformed white matter hyperintensity volume, and additionally adjusted for CES-D score, cardiovascular risk factors (systolic blood pressure, diastolic blood pressure, antihypertensive medication, serum cholesterol, HDL-cholesterol, lipid lowering medication, diabetes, smoking, BMI) and *apolipoprotein E-ε4 allele* carriership.

Abbreviations: FA: fractional anisotropy, MD; mean diffusivity, BMI: Body Mass Index, CES-D score: Center for Epidemiology Depression Scale score.