Genetic risk for Alzheimer disease in children: Evidence from early-life IQ and brain white-matter microstructure

María Fernanda Vinueza-Veloz1,2 | Carlos Martín-Román3
María Paulina Robalino-Valdivieso1 | Tonya White4,5 | Steven A. Kushner6,7 | Chris I. De Zeeuw2,8

1School of Medicine, Escuela Superior Politécnica de Chimborazo, Riobamba, Ecuador
2Department of Neuroscience, Erasmus MC, Rotterdam, The Netherlands
3Leiden Institute for Advanced Computer Science, Leiden University, Leiden, The Netherlands
4Department of Child and Adolescent Psychiatry/Psychology, Erasmus MC, Rotterdam, The Netherlands
5Department of Radiology and Nuclear Medicine, Erasmus MC, Rotterdam, The Netherlands
6Department of Psychiatry, Erasmus MC, Rotterdam, The Netherlands
7Department of Psychiatry, Columbia University, New York City, United States of America
8Royal Netherlands Academy of Arts and Sciences, The Netherlands Institute for Neuroscience, Amsterdam, The Netherlands

Correspondence
Dr María Fernanda Vinueza-Veloz and Dr Chris I.De Zeeuw, Department of Neuroscience, Erasmus MC, 40, Faculty Building, 3015 GE Rotterdam, The Netherlands.
Email: mafervive@gmail.com (M. F. V) and Email: c.dezeeuw@erasmusmc.nl (C. I. Z)

Funding information
Dutch Organization for Medical Sciences, Life Sciences, Social and Behavioral Sciences; ERC-adv; ERC-POC; Secretaria de Educación Superior, Ciencia, Tecnología e Innovación; Ministry of Health; Netherlands Organization for Scientific Research; Health Research; Erasmus University Rotterdam; Erasmus MC

Abstract
It remains unclear whether the genetic risk for late-onset Alzheimer disease (AD) is linked to premorbid individual differences in general cognitive ability and brain structure. The objective of the present study was to determine whether the genetic risk of late-onset AD is related to premorbid individual differences in intelligence quotient (IQ) and characteristics of the cerebral white-matter in children. The study sample included children of the Generation R Study from Rotterdam, The Netherlands. IQ was measured using a well-validated Dutch nonverbal IQ test (n = 1908) at ages 5 to 9 years. White-matter microstructure was assessed by measuring fractional anisotropy (FA) of white-matter tracts using diffusion tensor imaging (DTI) (n = 919) at ages 9 to 12 years. Genetic risk was quantified using three biologically defined genetic risk scores (GRSs) hypothesized to be related to the pathophysiology of late-onset AD: immune response, cholesterol/lipid metabolism and endocytosis. Higher genetic risk for late-onset AD that included genes associated with immune responsivity had a negative influence on cognition and cerebral white-matter microstructure. For each unit increase in the immune response GRS, IQ decreased by 0.259 SD (95% CI [−0.500, −0.017]). For each unit increase in the immune response GRS, global FA decreased by 0.373 SD (95% CI [−0.721, −0.026]). Neither cholesterol/lipid metabolism nor endocytosis GRSs were associated with IQ or cerebral white-matter microstructure. Our findings suggest that elevated genetic risk for late-onset AD may in part be manifest during childhood neurodevelopment through alterations in immune responsivity.

KEYWORDS
Alzheimer’s disease, brain white-matter, children, cholesterol/lipid metabolism, diffusion tensor imaging, endocytosis, genetic risk score, IQ, immune response, Snijders Oomen Non-verbal Intelligence Test
INTRODUCTION

AD is a devastating adult-onset disease that causes progressive and irreversible brain neurodegeneration. It is the most common cause of dementia, accounting for more than 60% of the cases. People suffering from AD exhibit insidious and progressive memory loss usually associated with behavioral changes. As the disease progresses, other spheres of cognition such as language, visuospatial skills or executive functions become gradually impaired, leading to complete loss of the "sense of self." These symptoms probably result from brain atrophy, which at early stages affects the hippocampus and entorhinal cortex.

Nevertheless, brain white-matter abnormalities are among the earliest pathophysiological alterations detected in people suffering from AD. It has been observed, for example, that subjects in preclinical or prodromal stages already exhibit widespread abnormalities of brain white-matter, likely associated with altered microglial activity. Furthermore, disturbances of axonal transport, oligodendrocyte function and myelination have been observed in young animal models of AD. Altogether these findings suggest that cortical atrophy is preceded by brain white-matter alterations, which might manifest decades prior to the onset of clinical symptoms.

Furthermore, brain white-matter alterations might contribute to the impaired cognitive abilities that occur in individuals with AD. Various epidemiological studies have shown that in both children and young adults, low intelligence quotient (IQ) and low linguistic ability are strong predictors of poor cognitive function and AD diagnosis later in life. Likewise, impairments in memory retention and abstract reasoning have been observed in individuals who are later diagnosed with AD. Although it is unknown how cognitive ability early in life relates to AD, it has been suggested that "cognitive reserve" attenuates the impact of neurodegenerative processes. In this way, individuals with poorer cognitive reserve have a higher risk of developing AD in comparison to individuals with higher cognitive reserve.

There are two types of AD that are defined by the age of onset of the symptoms: early-onset (familial) and late-onset (sporadic). Although the cause of late-onset AD is unknown, genetic susceptibility appears to play a crucial role. Several genes are known to harbor risk variants, and among them the ε4 allele of the APOE gene is the strongest known genetic risk factor for late-onset AD. Approximately, one-third of the total phenotypic variance of late-onset AD is explained by common single nucleotide polymorphisms (SNPs). A total of 19 SNPs have been identified in the largest meta-analysis of genome-wide association data conducted to date. Additionally, a rare variant with an effect-size similar to that of APOE-ε4, rs75932628 in the TREM2 gene, has also been observed to increase the risk for late-onset AD.

The objective of this study was to determine whether the genetic risk of late-onset AD is related to premorbid differences in general cognitive ability and cerebral white-matter integrity during childhood. We developed biologically informed genetic risk scores (GRSs) and examined their association with IQ and cerebral white-matter fractional anisotropy (FA) in children from the Generation R Study. GRSs were constructed based on three pathways that have been shown to be related to the etiology of late-onset AD: immune response, endocytosis, and cholesterol/lipid metabolism.

METHODS

The current work was embedded in the Generation R Study, an ongoing population-based prospective cohort study carried out in Rotterdam, the Netherlands. The Generation R Study was designed to identify early environmental and genetic causes as well as underlying pathways leading to normal and abnormal growth, development and health.

Ethical issues

The Generation R Study, its general design, aims and specific measurements have been approved by the Medical Ethical Committee of Erasmus MC, Rotterdam. Written informed consent was obtained from the parents for all data collected as a part of the study. At the start of each phase, mothers and their partners received written and oral information about the study. Even with consent of the parents, if the child was not willing to participate, no measurements were performed.

Participants

The sample included children of the Generation R Study for whom IQ, diffusion tensor imaging (DTI) and genetic data were available (in total 5756 children). Children without Northwestern European ancestry (n = 2926), without IQ data or an IQ score lower than 70 (n = 797) were excluded from the sample. We restricted the study-population to Northwestern European children, because GRSs were based on studies performed in individuals of European ancestry. Participants with an IQ below 70 were excluded to assure that the association between outcome and exposure was independent from severe intellectual disability. Moreover, to assure independence of observations, we also excluded randomly selected siblings (n = 125). As a result, the study sample corresponded to 1908 children.

IQ assessment

IQ of children was assessed using the Snijders Oomen Nonverbal Intelligence Test 2.5-7-Revised (SON-R 2.5-7), which is suitable for children above the age of 2.5 years and does not require the use of spoken or written language. The full SON-R 2.5-7 test comprises six subtests that evaluate six different cognitive domains. However, due to time constraints, children underwent two subtests: categories and mosaics, which measure abstract reasoning and spatial visualization,
respectively. The correlation coefficient between the score obtained from these two subtests and the total set of all six subtests was relatively high in a previous study \( r = .86 \). Raw scores derived from the two SON-R 2.5-7 subtests were converted into IQ scores that were specific for each age group of the children.\(^25\) Low-performance levels on these two subtests were included in our analyses. DTI data included in the present article were acquired during the second neuroimaging wave, which included high-resolution structural MRI, 35-direction diffusion weighted imaging, and a 6 minutes 2 seconds resting-state functional MRI scan.\(^27\) Data were acquired using a 3 Tesla GE Discovery MR750w MRI System scanner (General Electric, Milwaukee, Wisconsin) using an 8-channel head coil for signal reception.\(^27\) Details on how DTI data were generated have been explained in detail in previous publications.\(^27-29\) Briefly, children underwent a mock scanning session before the actual MRI scan. DTI data were acquired using a single shot, echo-planar imaging sequence and processed using the FMRIB Library and the Camino Diffusion MRI Toolkit.\(^29\)

### 2.4 | Brain white-matter microstructure

Individual differences in the microstructure of brain white-matter in children were assessed with MRI using DTI. DTI takes advantage of anisotropic diffusion of water molecules in tissues to quantitatively detect individual differences in the microstructural properties of brain white-matter.\(^26\) Among the measures generated by DTI are scalar parameters that reflect the total amount of anisotropic diffusion, such as FA. FA expresses the relationship between axial and radial diffusivity and thus changes in any of those parameters will have an impact on FA. Increased axonal density, reduced axonal caliber and/or increased degree of myelination will all lead to reduced radial diffusivity and therefore an elevated FA. Accordingly, FA is highly sensitive to individual differences in white-matter microstructure, but limited in its specificity.\(^26\)

DTI data included in the present article were acquired during the second neuroimaging wave, which included high-resolution structural MRI, 35-direction diffusion weighted imaging, and a 6 minutes 2 seconds resting-state functional MRI scan.\(^27\) Data were acquired using a 3 Tesla GE Discovery MR750w MRI System scanner (General Electric, Milwaukee, Wisconsin) using an 8-channel head coil for signal reception.\(^27\) Details on how DTI data were generated have been explained in detail in previous publications.\(^27-29\) Briefly, children underwent a mock scanning session before the actual MRI scan. DTI data were acquired using a single shot, echo-planar imaging sequence and processed using the FMRIB Library and the Camino Diffusion MRI Toolkit.\(^29\)

### 2.5 | Genetic data

DNA from children was extracted from the blood of the umbilical cord at birth. In case blood of the umbilical cord was not available, DNA was extracted from a blood sample that was obtained by venipuncture during the first visit of the child to the Generation R Research Center.\(^23\) Genetic data were generated by a genome-wide association scan using Illumina HumanHap 610 or 660 Quad chips (Illumina, Inc., San Diego, California), following manufacturer protocols. Genotype calling was performed on normalized data of allele signal-intensity using the Genecall module from Illumina Genome Studio (version 1.1.0.28426). To keep only good-quality samples, the quality metric provided by Illumina Genome Studio was used; in this way, samples with scores below 97.5% were excluded.\(^30\)

The way in which DNA quality control was performed has been described in detail before.\(^29\) In short, individuals with low call rates (>97.5%) or sex mismatches were excluded. SNPs with low call rates (<95%) were excluded. Ethnic composition was estimated by identity-by-state analysis using multidimensional scaling. Participants with more than four standard deviations (SDs) from a European reference panel (HapMap CEU) on the first four principal components were considered as non-northwestern Europeans. In order to maximize genome coverage, MACH (version 1.0.15) was used to impute genotypes at each of the autosomal SNPs of the Haplotype Reference Consortium (HRC) reference panel for Europeans and European-Americans. Criteria to determine which SNPs to include in the imputation step included: SNP call rate < 98%, statistically significant departures from Hardy-Weinberg equilibrium \((P < 10^{-7})\) and low minor allele frequency <1%.\(^21,23\)

### 2.6 | Data analyses

#### 2.6.1 | Diffusion tensor imaging

Confirmatory factor analysis using lavaan package was used to compute global DTI FA.\(^29\) Briefly, multiple tracts were summarized as a single latent factor and the predicted factors scores for each subject were generated.\(^29\) These tracts included: right and left cingulum bundle (CCG), right and left corticospinal tract, forceps major and forceps minor of the corpus callosum, right and left inferior longitudinal fasciculus (ILF), right and left superior longitudinal fasciculus and right and left uncinate fasciculus.\(^29\)

#### 2.6.2 | Genetic risk scores

To construct the GRSs we considered SNPs that were associated with late-onset AD at a genome-wide significance level \((P < 5 \times 10^{-8})\) and were replicated in independent data sets. Consequently, 22 SNPs were considered for inclusion: one SNP for TREM2, one SNP for CD2AP, one SNP for SORL1, one SNP for EPHA1, one SNP for PICALM, one SNP for ZCWPW1 and 19 SNPs that were identified by the IGAP consortium. From the 22 SNPs originally considered, 21 remained after we excluded rs9271192 in the HLA-DRB5-DRB1 gene, because its imputation quality was below the generally accepted threshold \((R^2 < .30)\).

GRSs were constructed based on four pathways that have been shown to be related to the etiology of late-onset AD: immune response, endocytosis, cholesterol/lipid metabolism, and protein ubiquitination.\(^20\) To assign the SNPs to the pathways we used information on gene
ontology from previous publications and Gene Network.\textsuperscript{31-33} Five of the 21 SNPs could not be assigned to any pathway. Only one SNP could be assigned to protein ubiquitination, and therefore this pathway was not considered in the analysis. Thus, we ended up with 15 SNPs distributed in three pathways: immune response, endocytosis and cholesterol/lipid metabolism (Table 1). For each of the three pathways we calculated a weighted GRSs by adding the product of SNP dosages and their respective reported effects (Odds-ratio [OR]; Table 1), according to the formula below using R (version 3.6.2).\textsuperscript{34}

$$\text{GRS} = \sum_{i} \text{SNP dosage}_{i} \times \log(\text{OR})_{i}$$

### 2.7 Statistical analysis

To analyze the association between IQ, microstructural properties of the brain white-matter (global FA) and the GRSs we used multiple linear regression. We modeled IQ as an outcome and the three GRSs (immune response, endocytosis and cholesterol/lipid metabolism) as predictors in a single regression model. The same approach was used to analyze the association between global FA and the three GRSs. Both models were adjusted for age, gender and four principal components of ancestry. To help interpreting regression coefficients, we computed z-values for IQ and global FA. A complementary analysis was performed including children with an IQ lower than 70 using not only the original analysis, but also k-means cluster analysis. Variables used in the clustering included IQ, global FA and immune response GRS. Two clusters were computed.

### RESULTS

To study the association of genetic risk for late-onset AD on IQ and global FA, we implemented three biologically informed GRSs: immune response, endocytosis and cholesterol/lipid metabolism (see section 2).

#### 3.1 General characteristics of the sample

The sample included 1908 children, 51% (n = 973) of whom were female. IQ was assessed in children at a mean age of 6.09 years (SD = 0.40). The mean IQ score of children of the sample was 105.60 (SD = 13.58), ranging from 71 to 150. From the sample, 919 children had DTI data available. Children were subjected to DTI at a mean age of 10.18 years (SD = 0.62). The mean global FA of children was 0.23 (SD = 13.58), ranging from −6.98 to 5.47. The immune response GRS ranged from −0.646 to 1.027 with a mean of −0.073. The endocytosis GRS ranged from −1.340 to 3.003 with a mean of 0.276. The cholesterol/lipid metabolism GRS ranged from −0.261 to 0.867 with a mean of 0.366. As previously showed IQ exhibited a significant positive association with global FA. For each unit increase in global FA, IQ increased by 0.111 SD (95% CI [0.045, 0.177]; P = .001).\textsuperscript{29}

### Table 1 SNPs that were used to build the GRSs.

<table>
<thead>
<tr>
<th>SNP</th>
<th>Gene</th>
<th>Odds-ratio</th>
<th>Immune response</th>
<th>Endocytosis</th>
<th>Cholesterol/lipid metabolism</th>
</tr>
</thead>
<tbody>
<tr>
<td>rs4147929</td>
<td>ABCA7</td>
<td>1.15\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs7412</td>
<td>APOE2</td>
<td>0.67\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs429358</td>
<td>APOE4</td>
<td>3.86\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs6733839</td>
<td>BIN1</td>
<td>1.22\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs10948363</td>
<td>CD2AP</td>
<td>1.10\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs9331896</td>
<td>CLU</td>
<td>0.86\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs6656401</td>
<td>CR1</td>
<td>1.18\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs11771145</td>
<td>EPHA1</td>
<td>0.90\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs35349669</td>
<td>INPP5D</td>
<td>1.08\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs190982</td>
<td>MEF2C</td>
<td>0.93\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs983392</td>
<td>MS4A64</td>
<td>0.90\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs10792832</td>
<td>PICALM</td>
<td>0.87\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs28834970</td>
<td>PTK2B</td>
<td>1.10\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs11218343</td>
<td>SORL1</td>
<td>0.77\textsuperscript{1}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>rs75932628</td>
<td>TREM2</td>
<td>2.43\textsuperscript{2}</td>
<td></td>
<td>*</td>
<td></td>
</tr>
</tbody>
</table>

Note: 15 SNPs could be assigned to at least one of the pathways identified as showing enrichment for association with late-onset AD. The pathway protein ubiquitination was excluded because none of the SNPS could assigned to this pathway. Notice that only immune/inflammatory response and endocytosis are completely independent pathways.

Abbreviations: Immune resp., immune response; Cholesterol/lipid met., cholesterol/lipid metabolism; Synaptic trans., synaptic transmission; 1, OR extracted from reference 18; 2, OR extracted from reference 19; *, belongs to.
3.2 | Effect of GRS on IQ

We observed that in comparison to the other scores, the immune response GRS exhibited a significant negative association with IQ. For each unit increase in the immune response GRS, IQ decreased by 0.259 SD (95% CI [−0.500, −0.017]). In contrast, no significant associations were observed between either the endocytosis or cholesterol/lipid metabolism GRSs with IQ (Table 2). Moreover, we also found no evidence for an interaction between immune response GRS and gender, or between immune response GRS and age in relation to IQ (all p-values >.2) (Table S1; and Figures S1 and S2).

3.3 | Effect of GRS on global FA

The immune response GRS also exhibited a significant negative association with global FA. For each unit increase in the immune response GRS, global FA decreased by 0.373 SD (95% CI [−0.721, −0.026]). Similar to the results for IQ, the GRSs for neither endocytosis nor cholesterol/lipid metabolism were significantly associated with global FA (Table 2). We did not find evidence for an interaction between immune response GRS and gender, nor between immune response GRS and age in relation to global FA (all p-values >.2) (Table S1 and Figures S1 and S2).

3.4 | Impact of IQ filter

We wanted to know whether children with moderate to severe intellectual disability (IQ < 70) tended to also have lower global FA and higher immune response GRS. When we included them in the original analysis described above, we observed that the P-values slightly increase, while coefficients were only mildly different (Table S2). Next, to further explore the associations, we subjected the same data set to k-means cluster analysis. Two clusters emerged, in which children from cluster 1 had higher IQ, higher global FA, and a lower immune response GRS compared with those of cluster 2 (all p < .001) (Table 3). Furthermore, the effect of the immune response GRS did not depend on the number of SNPs it contains. The regression model implemented to test this hypothesis showed that the effect of an overall GRS is not statically significant associated with neither IQ nor global FA (Table S3).

4 | DISCUSSION

The objective of the present study was to determine whether the genetic risk of late-onset AD is associated with premorbid individual differences in general cognitive ability and cerebral white-matter microstructure. Genetic risk was evaluated through biologically informed GRSs that were constructed based on widely held hypothesis about the pathophysiology of late-onset AD. We found that the genetic risk for late-onset AD associated with immune responsibility negatively influenced IQ and cerebral white-matter microstructure. Accordingly, children with a high immune response GRS were more likely to have lower IQ and global FA, the latter of which is thought to reflect decreased axonal density, increased axonal caliber, and/or decreased axonal myelination.

<table>
<thead>
<tr>
<th>TABLE 2</th>
<th>Summary of k-means cluster analysis.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>GRS</td>
</tr>
<tr>
<td>IQ</td>
<td>Immune response</td>
</tr>
<tr>
<td></td>
<td>Endocytosis</td>
</tr>
<tr>
<td></td>
<td>Cholesterol/lipid metabolism</td>
</tr>
<tr>
<td>FA</td>
<td>Immune response</td>
</tr>
<tr>
<td></td>
<td>Endocytosis</td>
</tr>
<tr>
<td></td>
<td>Cholesterol/lipid metabolism</td>
</tr>
</tbody>
</table>

Abbreviations: CI, confidence interval; FA, fractional anisotropy; GRS, genetic risk score; IQ, intelligence quotient; SD, standard deviation; SE, standard error.

Note: Both models were adjusted by age, gender and four principal components of ancestry. Coefficients were calculated using z-values therefore β represents change in terms of SD from the mean.

<table>
<thead>
<tr>
<th>TABLE 3</th>
<th>Summary of k-means cluster analysis.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cluster 1</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
</tr>
<tr>
<td>IQ</td>
<td>111.850</td>
</tr>
<tr>
<td>Global FA</td>
<td>1.140</td>
</tr>
<tr>
<td>Immune response GRS</td>
<td>−.154</td>
</tr>
</tbody>
</table>

Abbreviations: FA, fractional anisotropy; GRS, genetic risk score; IQ, intelligence quotient; SD, standard deviation.

Note: Analysis included children with moderate to severe intellectual disability (IQ lower than 70). Values for IQ, global FA and immune response GRS are showed for the two clusters that were computed.
A few studies have examined the association between the genetic risk of late-onset AD using GRSs and premorbid phenotypic traits, including one using overlapping data from the Generation R Study. The results of these studies are partly contradictory. For example, we and others found no evidence of an association between the genome-wide GRS for AD risk and various outcomes related to education, cognition, and behavior in children. However, this was only true for GRSs constructed from SNPs that were associated with late-onset AD at a genome-wide significance level (P ≤ 5 × 10^{-8}). Furthermore, other studies using a similar approach, found that the genetic risk for AD was significantly associated with poorer cognition and decreased hippocampal volume in children 6 to 14 years of age and healthy young adults. Notably, in both studies GRSs were constructed using the P-values of the examined SNPs with respect to AD diagnosis as cut-off points. Furthermore, these studies, including our earlier work, did not construct GRSs on the basis of a priori defined biological pathways.

In contrast to previous studies, we found no evidence for early-life associations between either IQ or cerebral white-matter microstructure related to genes involved in cholesterol/lipid metabolism, such as APOE. For instance, our findings contrast those of Chang et al, who reported that AD genetic risk, specifically with respect to that of APOEε4, is negatively associated with brain and cognitive development in children. They found that on average in comparison to non-carriers, children carrying APOEε4 have a smaller hippocampus, reduced hippocampal FA, and thinner entorhinal cortex, which was associated with poorer performance on attention and working memory tasks. These findings also contrast those of Acqua et al and a recent meta-analysis, both of which found no evidence of association between APOE genotype and cognitive traits or brain structure in younger persons.

4.1 Immune response, IQ and cerebral white-matter microstructure

Our findings suggest that genetic risk for late-onset AD might in part be associated with immune responsivity, which was associated with both general cognitive ability and cerebral white-matter microstructure. Notably, we did not correct for multiple comparisons as we employed a single model comprising all comparisons for each outcome. Our data are corroborated by the finding that the AD risk score capturing the immune response has also been shown to associate modestly with white matter lesions in adults, confirming the link in a separate, yet different population. These findings raise the question as to how genes involved in the immune response may contribute to differences in IQ? Given the association between the immune response GRS and changes in cerebral white-matter microstructure, our data suggest that genes involved in immune responses might exert their influence on IQ indirectly. Possibly, genes involved in the immune response could drive changes in myelination and/or axonal growth that ultimately have an impact on IQ. Alternatively, the genes involved in the immune response might also impact cognition by participating in plasticity of synapses during development and adulthood.

How could genes involved in immune response alter the microstructure of the cerebral white-matter, that is, myelination and/or axonal morphology? Human and animal studies have shown that EPHA1, MEF2C, INPP5, CR1, CLU, MS4A6A and TREM2 are expressed in microglia, which are the main cell type of the innate immunity in the central nervous system (CNS). Microglial cells, which represent nearly 20% of glial cells in the cerebrum, might contribute to myelination and axonal growth in an immune "surveillant" and an immune "active" manner. Surveillant microglia support the survival of oligodendrocytes as well as the differentiation of oligodendrocyte progenitor cells (OPCs). In contrast, active microglia stimulate remyelination not only by removing debris created by the breakdown of myelin but also by priming the efficient recruitment of OPCs. Likewise, active microglial cells facilitate neuronal survival and axonal growth by supplying neurons with neurotrophic factors, including brain-derived neurotrophic factor and nerve growth factor among others.

4.2 Implications of our findings

Our findings suggest that intelligence, cerebral white-matter and Alzheimer disease (AD) share a common set of genes linked to the immune response. This conclusion has important implications. First, they support the hypothesis that pathophysiological changes underlying the characteristic neurodegeneration and neurological symptoms in people with AD begin early in life. Accumulation of β-amyloid and formation of neurofibrillary tangles may indeed constitute only part of the neurodegenerative process. Furthermore, our results also suggest that dysfunctional white-matter may be an insufficiently unappreciated component of the pathogenesis of AD. In this respect, it is important to note that white-matter abnormalities have been detected in preclinical stages of AD preceding overt neurodegenerative changes in the cerebral cortex. Indeed, white-matter demyelination as well as signs of neuronal dysfunction can occur prior to the accumulation of β-amyloid and the formation of neurofibrillary tangles.

Second, our findings highlight the importance of the immune system in cerebral white matter development. Accordingly, diverse risk factors, whether they might be genetic or environmental, that interfere with the "normal" functioning of the immune system potentially contribute to AD risk. This hypothesis is consistent with several GWA studies of AD risk, which have implicated a group of linked to immune responsivity, particularly those related to innate immunity. It is possible that these genetic factors confer risk for AD by directly compromising the function of microglial cells, and perhaps indirectly for astrocytes, oligodendrocytes, and ultimately neurons, which may well establish the basis for our observed premorbid association with cerebral white-matter microstructure.

Third, our findings open the possibility of developing sensitive and specific criteria that might facilitate AD diagnosis and therapeutic intervention. These criteria would be envisioned as based on a combination of polygenic risk profiles, cognitive evaluation, and cerebral DTI. Together, this integrated approach might help to identify
individuals at high risk for developing AD during the early stages of the disease, thereby allowing for preventative interventions, such as cognitive training and lifestyle changes.53

4.3 | Strengths and limitations

Although the effects of the genetic risk for AD have been studied extensively in older populations, their impact on children has been less frequently investigated. To our knowledge, this is the first study to utilize biologically informed GRSs in childhood to examine associations with cognition or white-matter microstructure. In contrast to earlier work within the Generation R Study and other cohorts, our study used GRSs that were constructed by clustering variants according to pathways previously associated with AD. Although this approach constitutes a strength, it is also a limitation given that the findings are based on a limited number of SNPs without being certain of the genes being influenced. Future studies are warranted to further explore the potential causality and biological mechanisms underlying these genetic associations.

It is important to note that our findings suggesting that the generic risk for developing AD is associated with both intelligence and global FA should be interpreted with caution. Given that FA only denotes individual differences in the microstructural properties of cerebral white-matter, lower FA does not necessarily imply a particular pathological change. Moreover, the size of the effects we found was relatively small (about 1% of the phenotypic variance of IQ or FA). Therefore, this finding likely reflects the complex multidimensional nature of the brain and cognitive development. Studies attempting to replicate these findings will be important to validate these results.

ACKNOWLEDGMENTS

The Generation R Study is conducted by the Erasmus MC in close collaboration with the School of Law and Faculty of Social Sciences of the Erasmus University Rotterdam, the Municipal Health Service Rotterdam area, Rotterdam, the Rotterdam Homecare Foundation, Rotterdam and the Stichting Trombosedienst & Artsenlaboratorium Rijnmond (STAR-MDC), Rotterdam. The general design of Generation R Study is made possible by financial support from the Erasmus MC, Rotterdam, the Erasmus University Rotterdam, the Netherlands Organization for Health Research and Development (ZonMw), the Netherlands Organization for Scientific Research (NWO), the Ministry of Health, Welfare and Sport and the Ministry of Youth and Families. Support was also received from The Dutch Organization for Medical Sciences (C. I. D. Z.), Life Sciences (C. I. D. Z.), Social and Behavioral Sciences (C. I. D. Z.), Albinism Vrienden-Fonds Foundation (C. I. D. Z.), Medical Neuro-Delta, Crossover-NWO-LSH INTENSE (C. I. D. Z.), ERC-adv and ERC-POC programs of the EU (C. I. D. Z.) and the Secretaría de Educación Superior, Ciencia, Tecnología e Innovación (SENESCYT) of Ecuador (M. F. V. V).

CONFLICT OF INTEREST

The authors declare no competing financial interests in relation to the present work.

DATA AVAILABILITY STATEMENT

Data that support the findings of this study are available on request from the Generation R Study (https://generationr.nl/researchers/collaboration/). Data are not publicly available due to privacy or ethical restrictions based on European Union general data protection regulations.

ORCID

Maria Fernanda Vinueza-Veloz https://orcid.org/0000-0002-2493-0769

REFERENCES

42. Dantzer R. Neuroimmune interactions: from the brain to the immune system and vice versa. Physiol Rev. 2018;98(1):477-504.

SUPPORTING INFORMATION
Additional supporting information may be found online in the Supporting Information section at the end of this article.