Brain Morphometry, T2-Weighted Hyperintensities, and IQ in Children With Neurofibromatosis Type 1

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Background: Larger gray matter (GM) volume in healthy children is correlated with higher IQ. Children with neurofibromatosis type 1 (NF1) have larger brains, their magnetic resonance images frequently show T2-weighted hyperintensities, and their IQs are lower.

Objectives: To confirm the hypotheses that (1) children with NF1 have larger GM and white matter volumes, (2) the greatest volume differences are in the frontal and parietal regions and in children with NF1 with hyperintensities, and (3) GM volume is inversely related to IQ in children with NF1.

Design: Wechsler Intelligence Scale for Children–Third Edition IQ testing and measurement of cerebral volumes and hyperintensities in brain magnetic resonance images were performed on 36 children with NF1 and on 36 matched relatives who served as control subjects.

Results: Gray matter and white matter volumes were significantly larger in children with NF1. The greatest difference was observed in cerebral white matter volume, predominantly in the frontal lobes, whereas the greatest difference in GM volume was in the temporal, parietal, and occipital regions. In controls, IQ was significantly related to GM volume, but in children with NF1, IQ was not inversely associated with GM volume, although IQs of children with NF1 were significantly lower.

Conclusions: Children with NF1 do not have the normal relationship between GM volume and IQ. Larger GM volume in the posterior brain regions and larger white matter volumes in the frontal brain regions contribute to the larger brain volume in children with NF1.

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Patients with neurofibromatosis type 1 (NF1) often have a large head size, and structural abnormalities, particularly the signal hyperintensities, are commonly found in their brain magnetic resonance (MR) images. Children with NF1, on average, have lower IQs than those of healthy children. Brain volume, especially gray matter (GM) volume, in healthy children correlates positively with general intelligence. Increased brain volume in specific brain regions has been associated with cognitive dysfunction. Because NF1 involves some loss of growth control, we may gain a better understanding of the cognitive deficits in NF1 by defining whether the greater brain volume in NF1 is of GM or white matter (WM) origin, by specifying which brain regions comprise the volume changes, and by determining whether IQ is related to these changes.

Four studies examined brain MR imaging volumetric differences between patients with NF1 and control subjects. These studies confirmed that children with NF1 have significantly larger brain size, but each reached different conclusions regarding the contribution of GM and WM to the brain volume increase. Only one of these, a study limited to boys with NF1, attempted to define which regions of the brain have significant increases in WM or GM volume. Two studies found that increased brain volume was correlated with the presence of T2-weighted hyperintensities in children with NF1. Results from these previous studies have limited applicability to many children with NF1 because they enrolled special populations and did not have genetic controls for all of the subjects with NF1. For these reasons, we undertook a larger prospective study of cerebral lobar and regional GM and WM volumes and IQ in children with NF1 compared with family-matched genetic controls.

We hypothesized that whole-brain volumes, primarily supratentorial WM volume and the cross-sectional area of the corpus callosum, would be larger in children...
with NF1 and that IQ would be inversely related to brain volume in children with NF1. Because motor, visuospatial, and attentional processes are typically compromised in NF1, we also hypothesized that significant differences in WM volume between children with NF1 and family controls would be observed in the anterior brain and in predominantly parietal brain regions. We hypothesized further that children with NF1 with hyperintensities in the globus pallidus would have significantly larger WM volume.

**METHODS**

The human subjects research review boards at the University of North Carolina School of Medicine, Chapel Hill, and Duke University Medical Center, Durham, NC, approved the study protocols. All subjects (age range, 6-13 years) were from North Carolina or adjacent states and had been evaluated by a pediatric geneticist or a child neurologist to confirm the NF1 diagnosis. Children were excluded if they had the following: (1) a history of a clinically significant medical disease or condition that might severely compromise their ability to participate; (2) a history of serious psychiatric illness; (3) continuing seizures or significant neurologic deficit; (4) a diagnosis of brain tumor, except stable optic glomas; or (5) a documented IQ less than 70. The order of priority for matching controls to children with NF1 was as follows: (1) sibling, (2) age within 3 years of the child with NF1, (3) same sex as the child with NF1, and (4) second-degree relative, chosen in priority order of first cousin, aunt, or uncle of the same sex.

We identified 85 potential subjects with NF1. Forty-six subjects were excluded for the following reasons: (1) they did not have a closely related control subject (24 subjects), (2) they were not interested in participating in the study (10 subjects), (3) they could not undergo MR imaging (5 subjects), or (4) they met 1 of the medical exclusion criteria (7 subjects). The final cohort consisted of 36 children with NF1 (21 male; mean ± SD age, 9.3 ± 2.3 years) and 39 siblings or second-degree relatives (20 male; mean ± SD age, 9.5 ± 2.5 years). After study entry, we excluded MR imaging data from 2 NF1 and control pairs because 1 subject was too fearful to undergo MR imaging and because the MR image of 1 subject could not be analyzed because of technical difficulties.

**MR IMAGING AND ANALYSIS**

Unsedated subjects underwent MR imaging with a 1.5-T whole-body MR imaging system (Signa; GE Medical Systems, Milwaukee, Wis) using the standard head (volumetric) radiofrequency coil. High-resolution images were obtained with a dual-echo fast spin-echo sequence (repetition time, 4000 milliseconds; echo times, 30 milliseconds [first echo] and 100 milliseconds [second echo]; and echo train length, 16 echoes [8 for each echo]), with a 256 × 256-pixel matrix, 3-mm section thickness with no gaps between sections, 1 signal acquired per phase-encoding increment, and a 22-cm field of view.

The MR images were processed using the Duke University Medical Center Neuropsychiatric Imaging Research Laboratory–modified version of MRX software (GE Corporate Research and Development, Schenectady, NY) for image segmentation and the GRID program developed by the laboratory, as previously described. The segmentation protocol used by the laboratory was modified from that developed by Kikinis et al and Byrum et al. Reliability was established by repeated measurements on multiple MR images before raters were approved to process the study data.

Total hemisphere volumes included GM and WM in cortical and subcortical structures, excluding the cerebellum and infratentorial structures. The GM and WM components were separately assessed after segmentation. Cortical GM volume was assessed after excluding subcortical structures by means of MRX software.

The volumes of the left and right hemispheres were further subdivided using the GRID program into brain regions that approximated lobar regions of the brain. We realigned the 3-dimensional images to a standard orientation, including making the anterior commissure–posterior commissure plane horizontal, and defined planes according to a previously described regional stereotactic coordinate system (Figure). To measure the area of the corpus callosum, it was outlined in a midsagittal MR image reformatted from an axial T2-weighted fast spin-echo image on a commercial imaging workstation (Advantage Windows Workstation; GE Medical Systems).

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INTELLECTUAL PERFORMANCE

Each subject’s intellectual performance was assessed using the full version of the Wechsler Intelligence Scale for Children—Third Edition (WISC-III). The WISC-III yielded measures of full-scale IQ, verbal IQ, and performance IQ.17

STATISTICAL ANALYSIS

Paired t tests and Pearson χ² tests were used to compare the children with NF1 and the controls for demographic variables, brain volume measurements, and intellectual performance. SAS statistical software version 9.1 (SAS Institute Inc, Cary, NC) was used to analyze cortical GM and WM in relation to 16 parcellation regions in a repeated-measures analysis of variance, with group (NF1 vs control) and sex as between-subjects variables and with 3 within-subjects variables (2 levels of laterality, 4 levels of the anteroposterior plane, and 2 levels of the superoinferior plane). Given any significant interaction between group status and any of these within-subjects variables, individual parcellation regions were examined to better define the basis for the differences, with P<.05 indicating statistical significance. A general linear mixed-effects model18 was used to test group differences in the associations between brain volumes and intellectual performance. The matched pairs consisted of 20 full-sibling pairs, 9 half-sibling pairs, and 10 second-degree relative pairs, all first cousins.

RESULTS

The mean ages (t=−0.49, P=.63) and sex composition (χ²=0.05, P=.82) of the groups were comparable, and maternal education was not significantly different between the groups (χ²=1.63, P=.80). The matched pairs consisted of 20 full-sibling pairs, 9 half-sibling pairs, and 10 second-degree relative pairs, all first cousins.

BRAIN VOLUMES

Right and left hemisphere volumes, total GM volume, and total WM volume were significantly greater in children with NF1 (Table 1). The greatest percentage difference between the children with NF1 and their close relatives was in WM volume in both hemispheres. The greater WM volume was further reflected in a significantly larger mean area of the corpus callosum in the children with NF1. We did not find a differential effect for right vs left hemisphere volumes in the children with NF1.

Results of the repeated-measures analysis of variance demonstrated significant interactions between group status and within-subjects variables for GM and WM. For GM, a significant interaction with group was observed for the anteroposterior plane (F3,68=9.09), anteroposterior × laterality planes (F1,68=8.36), and anteroposterior × superoinferior planes (F3,68=10.02) (P<.001 for all). For WM, an interaction with group was observed for the anteroposterior × superoinferior planes (F1,68=5.47, P<.001). The significantly larger GM volumes were predominantly posterior to plane 5 (roughly corresponding to the parietal, occipital, and temporal regions of the brain) (Figure), but the significantly greater WM volumes were predominantly anterior to plane 5 (roughly corresponding to the postero-frontal, orbitofrontal, and anterotemporal regions of the brain). The parcellation volumes with greater WM volume were principally anterior to the parcellation volumes greater GM.

The mean ±SD brain volumes of the 25 children with NF1 with hyperintensities (GM, 986.83±21.80 cm³; and WM, 435.15±15.72 cm³) were not significantly different from those of the 11 children without hyperintensities (GM, 938.88±33.65 cm³; and WM, 389.66±23.28 cm³).

INTELLECTUAL PERFORMANCE AND BRAIN VOLUME

The mean WISC-III full-scale, verbal, and performance IQs were significantly lower in children with NF1 (P<.001 for all) (Table 2). Total GM volume but not WM volume was significantly positively related to WISC-III full-scale IQs for the controls, but not for the children with NF1 (Table 3). This positive relationship held for GM volumes in the right and left hemispheres.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control Group</th>
<th>NF1 Group</th>
<th>Difference, %</th>
<th>P Value</th>
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<td></td>
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<td>360.94 ± 42.97</td>
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<td>WM volume</td>
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<tr>
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<td>197.25 ± 39.00</td>
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<td>188.89 ± 36.52</td>
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<td>749.58 ± 94.60</td>
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</tbody>
</table>

Abbreviations: CC, corpus callosum; GM, gray matter; NF1, neurofibromatosis type 1; WM, white matter.

*Data are given as mean ± SD cubic centimeters unless otherwise indicated.

Table 1. WM Volume, GM Volume, and CC Area in Control Subjects and in Children With NF1

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The results of this study confirm previous findings that the larger brain volume of children with NF1 reflects greater GM and WM volumes compared with controls. In children with NF1, WM volume was significantly greater in parcellation regions that included most of the parietal, occipital, and temporal lobes, whereas greater WM volume was predominantly in parcellation regions corresponding to the posterofrontal and orbitofrontal lobes and the anterotemporal lobe. We also established for the first time, to our knowledge, that the expected relationship between cortical GM volume and cognitive function is disturbed in children with NF1. Children with NF1 had lower general cognitive function than their relatives, but they did not have the hypothesized inverse relationship between IQ and brain volume. Instead, only the controls showed a significant positive relationship between cortical GM volume and IQ.

The significantly larger brains of children with NF1 comprise greater GM and WM volumes, as demonstrated in the previous study, using different volume measurement methods, and as shown in a study of boys with NF1 without attention-deficit/hyperactivity disorder. One study of brain volume in children with NF1 attributed the greater brain size to a significant increase only in GM volume, and 2 other studies attributed the greater brain size to a significant increase in WM volume. These previous studies did not use closely related control subjects as we did, included younger subjects or only male subjects, or allowed inclusion of subjects with optic glioma. The measurements of brain volume in the latter study included the brainstem and cerebellum. We found the greatest percentage difference in cerebral WM volume, and post hoc analysis showed that significant changes were predominantly in frontal regions of the brain, as reported by Cutting et al in boys with NF1. They found a significant increase in frontal lobe WM volume, and sublobar analysis showed that the significant changes were in the premotor region of the frontal cortex. Contrary to our hypothesis and the findings of Cutting et al, WM volumes in the parietal lobe were not significantly different from those of controls in the present study. Instead, in our study, the GM volumes were significantly greater posteriorly. In the study by Cutting et al, GM volume in the frontal lobe was significantly greater than that in controls only in the boys with NF1 without attention-deficit/hyperactivity disorder, and this was attributed to an increase in the prefrontal region of the frontal lobe. We conclude that, when school-aged children with NF1 are compared with family members, their larger brain sizes reflect significantly greater GM and WM volumes, but the greatest difference is in WM volume in the posterior aspect of the frontal lobe. Furthermore, GM volume was significantly larger in the parietal and occipital cortices, areas of importance for understanding the visuospatial deficits that have usually been found in children with NF1.

Magnetic resonance imaging hyperintensities were not significantly related to any aspect of brain volume or IQ. Other investigators found a significant relationship between hyperintensities and brain volume, but as already noted, those studies utilized different types of subjects with NF1, and the findings were different. One study found an association between thalamic hyperintensities and GM volume, and the other study found an association between globus pallidus hyperintensities and WM volume. In view of our negative findings and the inconsistency of the findings in the previous studies, we conclude that the relationship between T2-weighted hyperintensities and brain volume is unclear and remains of unknown significance.

To our knowledge, our study is the largest study of family members paired with children with NF1 to show reduced IQ among the children with NF1. Previous studies demonstrated similar findings in children with NF1, but they were smaller and were not pair matched. Our study also indicates that the normal positive relationship between IQ and GM volume is absent in children with NF1. Our findings suggest that alteration in brain growth may be a factor in the cognitive deficits among children with NF1. A prospective longitudinal study design would be required to confirm such a hypothesis. Our study clearly raises the possibility that understanding what cellular elements cause the abnormal brain growth in NF1
may lead to a better understanding of the pathophysiology of learning disabilities in children with NF1.

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