ORIGINAL CONTRIBUTION

A Longitudinal Study of Brain Volume Changes in Normal Aging Using Serial Registered Magnetic Resonance Imaging

Rachael I. Scahill, MA; Chris Frost, MA, DipStat; Rhian Jenkins, BSc; Jennifer L. Whitwell, BA; Martin N. Rossor, MD, FRCP; Nick C. Fox, MD, MRCP

Objective: To investigate the effect of age on global and regional brain volumes and rates of atrophy, and to compare directly results based on cross-sectional and longitudinal data.

Methods: Thirty-nine healthy control subjects (age range, 31-84 years) underwent serial magnetic resonance imaging assessments. Measurements included the whole-brain, temporal lobe, hippocampal, and ventricular volumes at baseline and for repeat scans.

Results: We found significant decreases in cross-sectional whole-brain ($P<.001$), temporal lobe ($P<.001$), and hippocampal ($P=.003$) volumes and a significant increase in ventricular volume ($P<.001$) with increasing age. Cross-sectional and longitudinal estimates of atrophy rates were similar. We also found directional evidence of acceleration in atrophy rates with increasing age in all analyses, with the most marked changes occurring after 70 years of age. This increase in rates after 70 years of age was particularly marked in the ventricles ($P<.001$) and the hippocampi ($P=.01$).

Conclusions: We found a significant age-associated decrease in global and regional brain volumes. Some evidence indicates that this decline in brain volumes may be due to a nonlinear acceleration in rates of atrophy with increasing age. A better understanding of this process may help to discriminate normal age-related changes from neurodegenerative diseases.

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From the Dementia Research Group, Department of Clinical Neurology, Institute of Neurology, University College London (Mss Scahill, Jenkins, and Whitwell; Mr Frost; and Drs Rossor and Fox); the Medical Statistics Unit, London School of Hygiene and Tropical Medicine (Mr Frost); and the Division of Neuroscience and Psychological Medicine, Faculty of Medicine, Imperial College of Science, Engineering and Medicine (Dr Rossor), London, England.

THE INCREASES in life expectancy during the past 100 years constitute one of the great achievements of medical research and public health. However, the aging brain is associated with a greatly increased prevalence of dementia. Addressing this public health problem is a major challenge currently facing medical research.

Magnetic resonance imaging (MRI)–based measurements of the brain have been proposed as aids in the diagnosis of Alzheimer disease (AD) and other types of dementia. These measures have shown global brain atrophy,[1-3] reduced temporal lobe and in particular hippocampal volumes,[4-8] and increased cerebrospinal fluid (CSF) spaces[9] when subjects with AD are compared with control subjects. Rates of regional or global atrophy have also been proposed as surrogate markers of disease progression for use in clinical trials.[10] For diagnostic purposes, it is important to understand the structural aging process to differentiate pathologic rates of atrophy from normal age-related changes. For trial purposes, the best one could expect of a drug designed to slow disease progression in AD is to reduce the rate of atrophy to that seen in normal aging. As a result, normative longitudinal data are needed for sample size calculations for progression trials.

Numerous cross-sectional imaging studies have found a correlation between increasing age and decreasing brain volumes,[11-18] and these findings are substantiated by postmortem data.[19,20] Some studies have shown age-related decreases in hippocampal,[18,21,22] temporal,[13,22] and frontal lobe[16,23-25] volumes and increases in CSF spaces.[9,11,12,14,16-18] However, some contradictory evidence from other studies shows no significant association between age and ventricular CSF spaces[13,14] and whole-brain,[21] temporal lobe,[21,26] or hippocampal[27,28] volume.

Cross-sectional studies have a number of disadvantages. First, the large amount of between-individual variation that exists in the normal cerebral morphology reduces the sensitivity of methods to detect true cerebral volume differences between groups of subjects of different ages. For example, a large biological variation exists in head size be-

Table 1. Subject Demographics

Age Group, y (No. Male/Female)	Age, Mean (SD), y	Interval Between Scans, Mean (SD), d
30-39 (4/4)	36.1 (2.5)	575 (435)
40-49 (5/5)	45.6 (2.9)	667 (318)
50-59 (5/5)	53.9 (3.5)	698 (406)
60-69 (3/3)	62.7 (2.3)	754 (443)
70-84 (1/4)	76.8 (5.5)	356 (153)

tween sexes, and men have brain volumes that are on average 10% larger than those of women.[19] This intersex variation can only partly be adjusted for by normalizing to total intracranial volume (TIV).[29] Second, in case of cohort effects, caution must be exercised when drawing conclusions about true age-related changes. In much the same way that secular changes in heights have occurred during the past century, cerebral volumes may also be changing as maternal health and nutrition improve. In addition, the variety of age spans covered in these studies may account for some of the discrepancies in findings. For example, Jack et al[27] found no significant association between age and hippocampal volume in a group of subjects aged 20 to 40 years, but a more recent study of subjects aged 51 to 89 years by the same group found a significant age-related change.[22] These findings are entirely consistent if a nonlinear relationship exists between age and brain atrophy. Such nonlinear relationships make comparisons between different cross-sectional studies problematic.

Concensus is lacking about the age-related changes that occur in the brain during the whole range of adult life. Because of the large differences in methods and sample groups between centers, it is not clear whether the probable decline in cross-sectional brain volume is a result of consistent low-level atrophy, or whether the rate of atrophy accelerates after a certain age. To establish this, it is necessary to examine rates across the whole adult age range.

Longitudinal studies avoid some of the problems of secular trends and between-individual variation, as each subject forms his or her own control. Recent studies have used serial MRI to measure global and regional atrophy.[13,14] Such studies may allow more sensitive measurements of atrophy rates in subjects with AD and in normal aging.[1]

This study examined the effect of normal aging on global and regional brain volumes across an age range of 31 to 84 years. We made a direct comparison between predicted rates based on cross-sectional volumes and longitudinal rates of atrophy in the whole brain, ventricular CSF, temporal lobes, and hippocampi.

METHODS

SUBJECTS

We studied 39 subjects ranging in age from 31 to 84 years. Most of these individuals were spouses of subjects with AD who volunteered to take part in a longitudinal research study. These subjects had no history of neurological problems and no memory complaints or impairment, but did not undergo screening to exclude risk factors for vascular disease, such as smoking, diabetes, or hypertension, and so were relatively representative of the typical aging population. They were divided into 5 age groups, the demographics of which are given in **Table 1**.

All subjects underwent 2 MRI assessments. The mean Mini-Mental State Examination[30] score at baseline assessment for the whole group was 29.6 (SD, 0.7) of a possible 30. All subjects had given their consent for participation in longitudinal studies, which had approval from the local research ethics committee (National Hospital for Neurology and Neurosurgery and Institute of Neurology Joint Research Ethics Committee, London, England).

BRAIN IMAGING

We performed T1-weighted volumetric MRI scans on a 1.5-T Signa unit (GE, Milwaukee, Wis) using a 256×128 image matrix (acquisition variables: repetition time, 35 milliseconds; echo time, 5 milliseconds; number of excitations, 1; flip angle, 35°). These scans consisted of 124 contiguous 1.5-mm coronal slices through the head, which were transferred to a Sun workstation (Sun Microsystems, Inc, Santa Clara, Calif) for analysis.

IMAGE ANALYSIS

We used the software package MIDAS[31] for all volumetric measurements, which were performed by operators (R.I.S., R.J., and J.L.W.) masked to each subject's details.

GLOBAL VOLUMETRIC ANALYSIS

Whole-brain segmentation was performed on T1-weighted images using a semiautomated technique, which separates brain tissue from CSF by means of thresholding and a series of erosions and dilations.[31] Serial images were then positionally matched by means of a registration program using 9 *df*.[32]

A measure of whole-brain volume change, the brain boundary shift integral, was calculated as described previously.[33] Serial measurements of the TIV were also performed on the same T1-weighted scans according to the protocol described by Whitwell et al.[29] The lateral ventricles were measured using a consistent threshold of 60% of mean brain intensity as the boundary between the CSF and brain.

REGIONS OF INTEREST

The structure of interest was delineated using intensity thresholding and a mouse-driven cursor for tracing around the boundaries. Two orthogonal views (ie, any 2 of axial, coronal, and sagittal) could be seen by the operator simultaneously, which improves boundary definition for volumetric measurements. Left- and right-side structures were measured separately. All scans were presented twice each, once conventionally and once flipped across the midsagittal plane. The scans were presented in a random order, and the operator was masked to the laterality of the structures and to whether it was a baseline or a repeat scan. Reproducibility error was assessed using the mean absolute percentage of difference between repeat measures.

Temporal Lobe

The brain-CSF boundary was standardized by setting a consistent lower threshold of 60% of the mean brain intensity. The lobe was separated from the remainder of the brain by drawing a straight line across the temporal stem from the superior point of the medial side to the inferomedial extreme of the sylvian fissure. The mean absolute percentage of difference was 3%.

Hippocampus

The hippocampus was defined as including the dentate gyrus, the hippocampus proper, the subiculum, and the alveus, according to a neuroanatomical atlas.[34] The tail of the hippocampus was excluded. Measurement commenced at the longest length of the fornix, visualized in the coronal plane, and proceeded rostrally to the junction with the amygdala. The hippocampus was bounded superiorly, medially, and laterally by CSF and inferiorly by the white matter of the subiculum. The mean absolute percentage of difference was 3%.

STATISTICAL ANALYSIS

We analyzed data using STATA software, version 6 (Stata Corp, College Station, Tex). Whole-brain and regional volumes were standardized to the group mean TIV to account for differences in head size. We fitted a linear regression model relating each volume to the TIV, with all variables on a logarithmic scale.

We performed all analysis of volume reductions using logarithmic scales. Expansion of ventricular volumes were analyzed on an arithmetic scale.

Left- and right-side volumes of the temporal lobes and hippocampus were combined. The geometric (arithmetic for ventricles) means of the TIV-adjusted baseline and repeat measurements were used as cross-sectional measurements. We used regression models to investigate the effects of age and sex on the cross-sectional volumes before and after TIV correction.

Rates of ventricular enlargement were calculated as the difference between the TIV-adjusted baseline and repeat volumes, expressed as cubic millimeters per year. Rates of atrophy for all other structures were expressed as proportional rates.

A regression model was used to investigate the effect of sex on the rates of atrophy. Continuous acceleration in atrophy was assessed by regressing atrophy rates on age, weighting by the square of the follow-up time. A further regression model was used to investigate whether the rates of change between the groups younger and older than 70 years differed. For the regression analysis, we used age as a continuous variable, with the exception of the comparison of groups younger and older than 70 years, for which we used discrete groupings.

RESULTS

CROSS-SECTIONAL DATA

Regression analysis showed a significant effect of sex on all volumetric measurements. Men had larger volumes than women for the whole brain (15%; $P<.001$), ventricles (44%; $P<.001$), temporal lobes (14%; $P<.001$), and hippocampi (8%; $P<.001$). When the TIV was used to correct for head size, this sex effect was removed, and there were no significant differences in total brain ($P=.95$), ventricular ($P=.62$), temporal lobe ($P=.67$), or hippocampal volume ($P=.33$). We found no significant relationship between age and TIV ($P=.70$).

Part A of the **Figure** shows the cross-sectional volumetric data for each age group. Regression analysis showed a significant reduction in total brain ($P<.001$), temporal lobe ($P<.001$), and hippocampal volumes ($P=.003$) associated with age, accompanied by a significant increase in ventricular volume ($P<.001$).

The mean rates of atrophy predicted by the cross-sectional data are shown in **Table 2**. Adding quadratic terms to the regression models provided directional evidence of continuous acceleration in rate of change, but the effects were not statistically significant across the whole age range for the whole brain ($P=.58$), the ventricles ($P=.28$), the temporal lobes ($P=.051$), or the hippocampi ($P=.44$).

LONGITUDINAL DATA

Part B of the Figure shows the rates of volume change for each age group. Regression-based annualized rates of volume change for the whole group are shown in Table 2. For whole-brain volume, the mean rate of atrophy across the whole age range was 0.32% (95% confidence interval [CI], 0.10%-0.54%) per year, which is very similar to the rate estimated from the cross-sectional data. No evidence suggested a significant continuous acceleration in the rate of global atrophy with increasing age ($P=.54$), or an increase after 70 years of age ($P=.51$).

The mean rate of ventricular volume change was 650 mm^3/y, which is consistent with the cross-sectional data. The test of continuous acceleration was statistically significant ($P=.02$), as was a comparison of rates in the groups younger and older than 70 years ($P<.001$).

For the temporal lobes and hippocampi, the mean rates of atrophy were 0.68% (95% CI, 0.42%-0.93%) and 0.82% (95% CI, 0.53%-1.11%) per year, respectively. The hippocampal rate in particular was somewhat larger than that seen with the cross-sectional data. We found a nonsignificant trend for gradually accelerating rates of atrophy with increasing age in the hippocampi ($P=.11$) and the temporal lobes ($P=.21$). A comparison of rates before and after 70 years of age was statistically significant for hippocampi ($P=.01$), but not for the temporal lobes ($P=.80$).

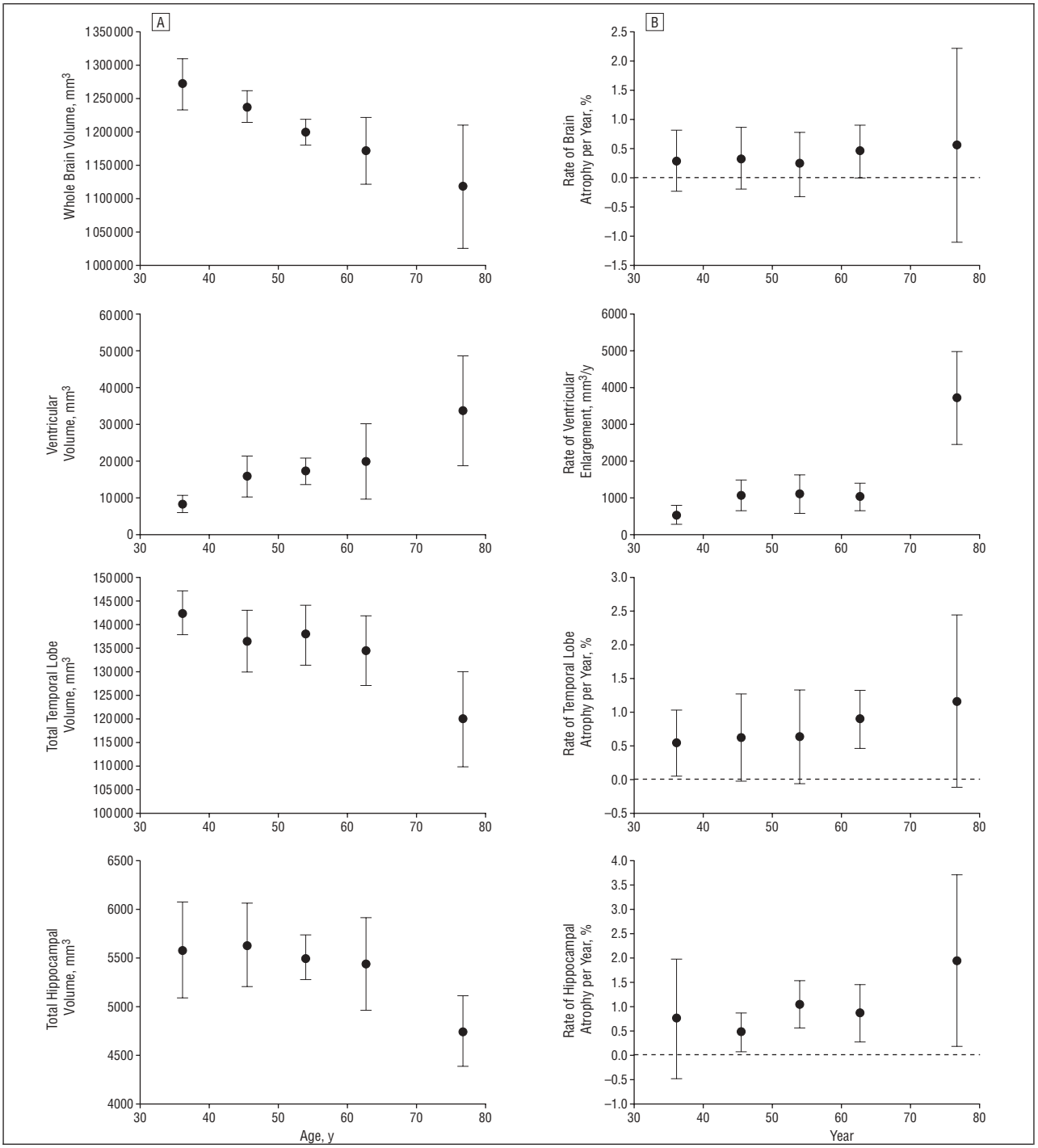
We found no significant effect of sex on the rates of change in any of the brain structures ($P>.23$).

COMMENT

Using a combination of semiautomated and manual assessment of MRI scans, we have demonstrated a highly significant age-associated difference in cross-sectional volumes, which concurs with the findings of several previous studies.[11-14,16-18,22] Cross-sectional age-related volume differences were seen at both a regional and global level. We found no evidence of a relationship between TIV and age, which concurs with previous work[29] and suggests that cross-sectional differences in cerebral volumes are not due to a cohort effect, but are more likely to reflect real volume loss during the aging process.

We found a highly significant and well-recognized effect of sex on volume, with men having larger brain volumes. This effect was removed when a correction for head size, TIV, was included in the regression analysis, which again concurs with previous data.[12,29] This finding suggests that studies considering the effect of sex on cross-sectional volumes should include a correction for head size to reduce this potential confounding effect.

Atrophy rates may be constant throughout adult life, and the cross-sectional differences seen may be the accumulation of this low-level atrophy. Alternatively, atrophy may accelerate with increasing age. Our longitu-



Total intracranial volume–adjusted cross-sectional volumes (A), and longitudinal rates of change (B) in the volume of the whole brain, ventricles, temporal lobes, and hippocampi across ages ranging from 30 to 84 years. Mean values with 95% confidence intervals are plotted against the mean age for each of the 5 age groups.

dinal results did not show a significant, consistent continuous age-related acceleration in the rates of global or regional brain atrophy, which concurs with the findings of previous longitudinal studies.[13,14] However, a significant acceleration in ventricular expansion with age was observed, as in recent work.[14] Although the study by Resnick et al[14] was based on much older subjects, the rates of ventricular enlargement we report in the subjects older than 60 years are entirely consistent with those of that much larger study. Nevertheless, ventricular expansion may not be wholly attributable to brain atrophy. Other factors, such as CSF dynamics, change the relative distribution of CSF in ventricular and sulcal spaces and thereby may influence these observations.

Although we did not find a consistent acceleration in rates across the whole adult age range, our results suggest a nonlinear pattern, with rates remaining comparatively stable before 70 years of age, but accelerating thereafter. The small sample sizes make it difficult to determine the point at which this acceleration occurs. In addition,

Table 2. Annual Rates of Atrophy and Enlargement Based on Cross-sectional and Longitudinal Data

Region	Mean (95% Confidence Interval)	
	Cross-sectional Data	Longitudinal Data
Whole brain*	0.33 (0.25-0.41)	0.32 (0.10-0.54)
Temporal lobes*	0.35 (0.20-0.51)	0.68 (0.42-0.93)
Hippocampi*	0.35 (0.13-0.57)	0.82 (0.53-1.11)
Lateral ventricles, mm^3/y	521 (323-719)	650 (333-968)

*Reported as percentage of atrophy per year.

the small number of subjects may contribute to the larger variance in the group older than 70 years. This variability may reflect heterogeneity within this older group. Some of these subjects, although apparently cognitively normal, may have been in the early presymptomatic stages of a degenerative disease, and consequently show elevated rates of atrophy. Serial MRI using larger sample sizes and including older subjects is needed to show significant changes in rates of atrophy as opposed to simple volume differences with age.

The rates of whole-brain atrophy and ventricular expansion predicted from the cross-sectional and longitudinal data were consistent. However, we found discrepancies between the cross-sectional and longitudinal rates of regional atrophy, with higher rates of atrophy of the hippocampus and temporal lobes seen using the longitudinal data. Again, this finding may be due to the small number of subjects in each age group or to the presence of individuals in the earliest stages of disease, which arguably might first manifest itself with increased rates of hippocampal atrophy on longitudinal analysis. In addition, the measurement error associated with these structures results in increased variability and may reduce the sensitivity of the technique to detect small changes in a single individual over time.

Because of the care and time required in manual outlining, a possible limitation of this study is the focus on a small number of anatomical regions. Other areas are thought to be involved in early AD and may also be subject to age-related decline. For example, volume reductions in the entorhinal cortex[8,24] and the posterior cingulate[35] have been shown in subjects with AD. Further work examining these regions may provide important additional information on the distribution of age-associated changes within the brain. In particular, the application of an automated technique such as statistical parametric mapping[36] to longitudinal data has the potential to detect regional atrophy without the need for time-consuming volumetric analysis.[37]

CONCLUSIONS

Using volumetric analysis, we have demonstrated age-related decreases in brain volumes across the adult age range, with some evidence of an acceleration in rates of atrophy after 70 years of age. Our findings have implications for the use of similar techniques for the diagnosis of dementia and the design of disease modification trials.

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Corresponding author: Rachael I. Scahill, MA, Dementia Research Group, Box 16, Institute of Neurology, Queen Square, London WC1N 3BG, England (e-mail: rscahill@dementia.ion.ucl.ac.uk).

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