Pergamon

Neurobiology of Aging, Vol. 15, No. 4, pp. 393–397, 1994

0197-4580/94 $6.00 + .00

0197-4580(94)E0027-W

# Human Corpus Callosum in Aging and Alzheimer's Disease: A Magnetic Resonance Imaging Study

A. BIEGON,*[1] J. L. EBERLING,* B. C. RICHARDSON,*† M. S. ROOS,* S. T. S. WONG,* B. R. REED*† AND W. J. JAGUST*†

*Center for Functional Imaging, Lawrence Berkeley Laboratory, University of California, Berkeley, CA 94720
†Department of Neurology, University of California, Davis, CA 95616

Received 22 September 1992; Revised 1 December 1993; Accepted 27 December 1993

BIEGON, A., J. L. EBERLING, B. C. RICHARDSON, M. S. ROOS, S. T. S. WONG, B. R. REED AND W. J. JAGUST. *Human corpus callosum in aging and Alzheimer's disease: A magnetic resonance imaging study.* NEUROBIOL AGING **15**(4) 393–397, 1994.—The involvement of the corpus callosum in aging and Alzheimer's disease (AD) is not clear. We measured cross sectional areas of the entire corpus callosum (CC), as well as the front 20% (genu), middle 60% (body), and posterior 20% (splenium) of the structure from a midsagittal MRI slice in AD patients ($N$ = 20), and young ($N$ = 16) and old ($N$ = 13) control subjects. We found that mean CC area in young controls was 570 ± 107 mm$^2$. Aging did not significantly affect the mean area of the CC (562 ± 98 mm$^2$). A small, significant reduction was seen in AD in comparison to the young control group (480 ± 133 mm$^2$). However, AD is accompanied by a large and statistically significant reduction in the genu area in comparison to both young and old control subjects. A trend toward an age-dependent reduction in the body area is also accentuated in AD patients who showed significantly smaller callosal bodies than young controls. We conclude that selective changes within the corpus callosum accompany aging and AD pathology.

Quantitative MRI  Corpus callosum  Aging  Alzheimer's disease  Human brain

THE CORPUS callosum (CC) is the major connection between the cerebral hemispheres. An intact corpus callosum is essential for the integration of information between the two hemispheres. Disruption or atrophy of the corpus callosum, as in "split brain" or multiple sclerosis patients, is accompanied by specific cognitive and neuropsychological deficits (3,14,20,22).

Alzheimer's disease (AD) is accompanied by both severe and progressive cognitive loss and brain atrophy (4,5). A variable degree of cognitive loss as well as some brain atrophy are also associated with normal aging (5). Despite the well documented changes in the cerebral hemispheres and ventricular system that occur in both aging and AD, relatively little is known about changes in the corpus callosum. The advent of nuclear magnetic resonance imaging (MRI) techniques has facilitated the measurement of callosal integrity and size in living patients. Midsagittal brain slices have been used for quantitative examination of changes in callosal cross sectional area as well as the areas of the genu, isthmus, and splenium, in a variety of conditions including psychiatric disorders, multiple sclerosis, dyslexia, and normal development (1,15,17,21). We have chosen this approach to examine the possible effects of AD on the corpus callosum and its components, by comparing a group of AD patients to a group of age-matched controls. An additional group of young controls was studied to examine the effects of aging.

## METHOD

### *Subject Selection*

Twenty AD patients were recruited from a university dementia clinic and were part of a longitudinal study of changes in brain structure and function in AD. All patients met current criteria for probable or possible AD (18), and in addition were free of all significant medical illnesses and were taking no psychoactive medications. A standard laboratory evaluation to exclude reversible causes of dementia was performed on each subject and all patients showed impairment in multiple areas of cognitive function using an extensive neuropsychological battery.

Three patients were severely demented (MMSE of 10 or less), 9 were moderately demented (MMSE 11 to 20), and 8 were mildly demented (MMSE of 21 or greater). Seven patients had early onset dementia (before 65) and 13 were late onset cases. All patients had a CT or MRI scan before participating in this MRI protocol, and in all cases the scans were normal or revealed cerebral atrophy or a small number of hyperintense regions on T2-weighted MRI scans.

A total of 31 control subjects were recruited from the community by advertisement, and were divided into a group of 16 young controls (YC) and a group of 15 "old" controls (OC) using a cutoff at age 50. To create an age matched group for comparison

[1] Requests for reprints should be addressed to Anat Biegon, Ph.D., at his current address: Pharmos Corp., Kiryat Weizmann Science Park, Rehovot Israel.

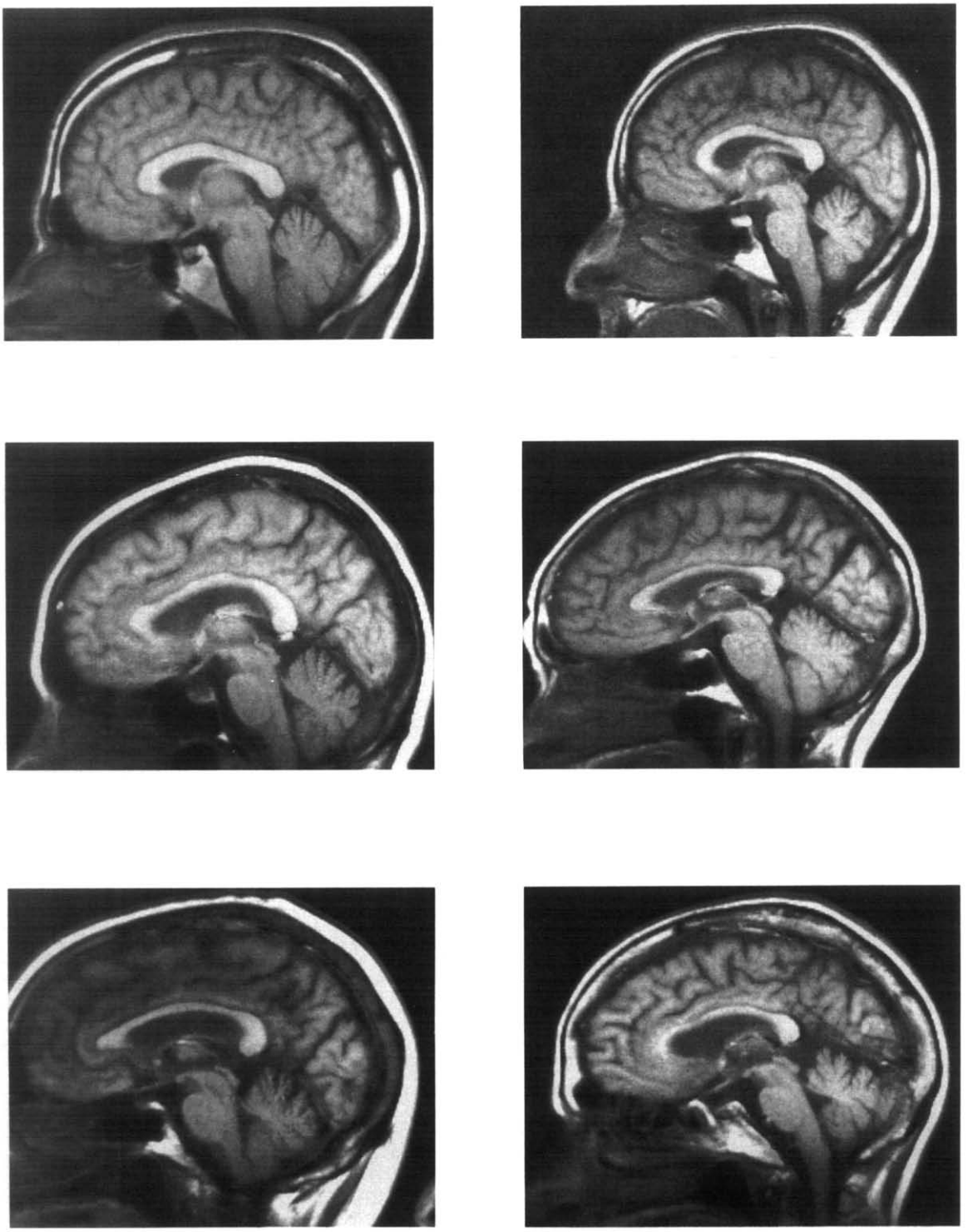

FIG. 1. Corpora callosa of young and elderly controls and AD patients. The range of sizes and shapes of corpus callosum found among young, elderly, and AD patients participating in the study is illustrated by representative images which were acquired as described in the text. Top row: Range of CC sizes in young healthy controls. The CC on the left (male, 29 years old) is among the largest in the study population. The CC on the right (female, 30) is the smallest in the young control group. Middle row: Range of CC sizes among healthy elderly subjects. The image on the left (female, 63 years old) represents a group of elderly controls with large, mostly intact callosa. The image on the right (female, 76 years old) shows one of the smallest callosa in this group. Some ventricular enlargement, cortical atrophy and thinning of the body of the CC can be seen in both images, when compared to the young controls. Bottom row: Range of CC sizes among AD patients. The patient on the left (female, 61 years old) has the best preserved CC in the AD group. Still, the genu is smaller than in the age matched control above. The patient on the right (female, 74 years old) represents a group of AD patients with extensive degeneration of the genu coupled with ventricular enlargement.

with the AD patients, two control subjects aged 50 and 52 who were included in correlational analyses of CC with age were excluded from the group comparison with AD patients (Table 1). All control subjects were interviewed and were free of any medical or psychiatric illnesses, were taking no medications, and had full social and occupational functional levels. The institutional review boards of all participating institutions approved the procedures for obtaining informed consent for both patients and controls.

*MRI Procedure*

MRI scans were obtained using a 0.5 Tesla magnet with a standard spin echo study using a TE = 33 ms and TR = 0.4 s. The slice thickness was 1.0 cm, with a field of view of 25.6 cm and a resolution of 1 mm in-plane. This protocol was not designed specifically for callosal measurements but rather as a means for orienting subsequent axial and coronal studies. Subjects were placed in the magnet with their head immobilized using a plastic form-fitting mask. A single midsagittal slice was chosen for analysis. This was followed by multi-slice acquisitions in the transaxial and coronal planes which were not analyzed in the current study. The whole procedure took 45 min.

*Data Analysis*

The midsagittal slice was chosen based on anatomical landmarks. From our data base of MRI scans of AD patients and controls, an experimenter blind to the age and diagnosis of the subjects (AB) picked only those slices (and cases) with a midline representation of the corpus callosum, as judged by the appearance of the cortical mantle surrounding the structure (no white matter), the presence of an undistorted spinal cord in the field of view, and the appearance of cerebellum, fourth ventricle and colliculi. Those sections which showed evidence of head rotation were not used.

The MRI slices were displayed and analyzed on a SPARC workstation using VIDA (University of Pennsylvania), an image processing and analysis software package. Initially, grey level histograms were obtained from a sample region deep in the CC and from sample regions in surrounding grey matter. These histograms were used to select thresholds for region growing. When thresholding and region growing failed, segmentation was completed manually, and the CC grey level set to 255. A grey level histogram of the segmented image was then used to compute the cross-sectional area of the CC (1 pixel = 1 $mm^2$). A digital ruler was then "anchored" to the most anterior point of the callosum, i.e., a point lying at the tip of the genu. The ruler was then swivelled toward the posterior end of the callosum, in parallel to the position of the structure in the field of view. The resulting line pointed either straight across or diagonally up or diagonally down, depending on the patient's head position. The position in which the intersection of the posterior end of the splenium produced the longest distance was then used as supplying the longest axis of the callosum. The longest axis of the structure was then measured, and the anterior 20% defined as genu, the middle 60% defined as body and the posterior 20% defined as splenium (e.g., Duara *et al.* (10). Each subregion was assigned a different grey level and its area was then computed from a final grey level histogram. The experimenter (AB) performing the analysis and area measurements was blind to the diagnosis and sex of the subjects. The reproducibility of the measurements was found to be better than 95%: a subset of four scans was measured twice by the same experimenter. A subset of 8 scans was independently measured by a student assistant. In both cases, the two sets of measurements agreed within <5%.

In addition to measurements of the areas of various segments of the *corpus callosum*, we also computed the ratio of the genu and body to splenium in each subject. These ratios control for variations in the size of the whole brain between subjects.

*Statistical Analysis*

The multivariate analyses of variance (MANOVA) was performed to evaluate callosal area measurements and ratios. Because this result was significant, a series of two-way (group × sex) analyses of variance (ANOVA) for each dependent variable were run to assess possible sex differences. A series of one-way ANOVAs, and Fisher PLSD and Scheffé posthoc tests ($\alpha = 0.05$) were used to evaluate group differences. Chi-square tests were used to evaluate categorical data.

TABLE 1

SUBJECT CHARACTERISTICS

| | Young Controls | Old Controls | AD Patients |
|---|---|---|---|
| *n* | 16 | 13 | 20 |
| Age (years, mean ± SD) | 33.4 ± 5.9* | 64.8 ± 9.0 | 70.1 ± 7.4 |
| Range | 26–41 | 54–78 | 55–79 |
| Sex (m/f) | 8m/8f | 6m/7f | 6m/14f |
| Handedness (l/r) | 1/15 | 0/13 | 1/19 |
| MMSE (mean ± SD) | — | — | 17 ± 7.2 |
| Range | | | 9–25 |

* $F(2, 46) = 119.7$, $p = 0.0001$; significantly different from old controls and AD patients, Scheffé and Fisher PLSD post hoc tests. MMSE = Mini Mental State Examination Score.

## RESULTS

The chi-square test revealed that the three groups did differ on sex, ($\chi^2 = 1.68$, $p = 0.43$). An overall MANOVA (including group, gender, and regions) comparing callosal measurements was significant ($p = 0.0001$). The two way (group × sex) ANOVAs revealed no significant ($p > 0.05$) sex effects or group × sex interactions. Whereas the mean values for genu and body tended to be lower in females than in males, the differences were not significant. Two-way repeated measures ANOVA by group and region revealed significant effects of group ($p < 0.05$) and region ($p < 0.0001$) and a highly significant group by region interaction ($p < 0.0001$).

Subsequent one-way ANOVAs of CC size and ratios revealed significant, region specific effects of both age and AD (Table 2). The cross-sectional area of the whole CC showed a small, nonsignificant reduction with age and a further reduction in AD, such that total CC area was significantly lower in AD when compared to young but not old controls. Whereas there was no significant decrease in the size of the genu with age, this region was strongly affected by AD. Mean genu area in AD patients was 30–35% lower when compared to old and young controls, respectively, a highly significant difference. The size of the body of the CC was diminished with aging as well as AD. However, a significant reduction in body cross-sectional area was seen only when AD patients we compared to young controls as the trend observed in the elderly control group did not reach statistical significance. The decrease in mean body area was smaller than the decrease in genu area and amounted to 14%–22% in old controls and AD patients, respectively.

Subdividing the control subjects to young and old with a cutoff age of 60 rather than 50 did not change the observed pattern of changes in callosal measurements. Again, the only subregion showing a trend towards decreased size with age was the body of the callosum. This was also confirmed by correlation analysis:

TABLE 2

EFFECTS OF AGE AND AD ON CORPUS CALLOSUM MEASUREMENTS

| Callosal Measurement (Mean ± SD) | Young Controls ($n$ = 16) | Old Controls ($n$ = 13) | AD Patients ($n$ = 20) |
|---|---|---|---|
| Total ($mm^2$) | 570 ± 107 | 562 ± 98 | 480 ± 133* |
| Genu ($mm^2$) | 176 ± 34 | 166 ± 40 | 115 ± 34† |
| Body ($mm^2$) | 224 ± 45 | 192 ± 45 | 174 ± 58‡ |
| Splenium ($mm^2$) | 171 ± 36 | 204 ± 39 | 190 ± 65 |
| Genu/Splenium | 1.04 ± 0.12 | 0.83 ± 0.19§ | 0.63 ± 0.19‖ |
| Body/Splenium | 1.33 ± 0.18 | 0.94 ± 0.18¶ | 0.95 ± 0.26¶ |

Young and old controls defined in Table 1.

* $F(2, 46) = 3.34$, $p = 0.04$; significantly different from young controls, Fisher PLSD post hoc test. †$F(2, 46) = 14.8$, $p = 0.0001$; significantly different from old and young controls, Scheffé and Fisher PLSD post hoc tests. ‡$F(2, 46) = 4.3$, $p < 0.02$; significantly different from young controls, Scheffé and Fisher PLSD post hoc tests. §$F(2, 46) = 24.9$, $p = 0.0001$; significantly different from young controls and AD, Scheffé, and Fisher PLSD post hoc tests. ‖significantly different from old and young controls, Scheffé and Fisher PLSD post hoc tests. ¶$F(2, 46) = 14.2$, $p = 0.0001$; significantly different from young controls, Scheffé, and Fisher PLSD post hoc tests.

While the Pearson correlation coefficients between age and whole callosum and genu were small and nonsignificant ($R = -0.03$, $p = 0.8$; $R = -0.1$, $p = 0.6$, respectively), the body area was significantly negatively correlated with age ($R = -0.36$, $p = 0.05$).

The size of the splenium did not appear to decrease at all with either aging or AD, and, in fact, the area of the splenium in the OC and AD groups was larger than in young controls although not significantly. Because there were no significant differences in splenium area across the three groups, we used the splenium as a denominator, representative of individual differences in overall brain size, to compute individual ratios of CC measurements.

Comparison of genu/splenium and body/splenium ratios across the three groups reaffirmed the finding of age-related decreases in the mean area of the body and severe degeneration of the genu in AD. MMSE results did not correlate significantly with any of the callosal measurements.

## DISCUSSION

Using magnetic resonance imaging, we have shown here that the human *corpus callosum* is affected by both age and AD in a specific, region-selective manner. No significant sex differences were observed in our study population or in any of the subgroups (young and elderly controls and AD patients). Although the range of normal values encountered in the subjects included in this study is quite wide, both the mean values for cross sectional area of the whole CC (approximately 600 $mm^2$) and the range of values (300–800 $mm^2$) are in good agreement with previously reported MRI studies of callosal size in healthy individuals (8).

The cross sectional area of the total CC in our sample did not appear to be reduced with age and was only slightly reduced with AD (~15% mean reduction when compared to OC). However, different callosal regions were affected differentially by aging and dementia. We found a significant (~30%) decrease in the area of the genu in the AD patients, compared to a small (6%), nonsignificant difference between young and old control groups. Because considerable brain atrophy is expected in AD, one could argue that the decrease in genu merely reflects a generalized, nonspecific decrease in brain and callosum size. If that were the case, the group differences would disappear when individual ratios of callosal measurements, rather than absolute measurements, were compared. This does not appear to be the case in our sample because the decrease is even more significant when genu/splenium ratios in AD patients are compared to those computed for age-matched elderly controls.

The effects of age alone appear to be most pronounced in the body of the CC. This region was the only one showing a significant negative correlation with age in the control group.

These results, taken in conjunction with other work reported in the literature, suggest that the effect of age on callosal size is complex. While we, and others (16,21) using MRI found that there was no effect of age alone on total callosal area, there are also reports of age effects on this variable (8). Doraiswamy et al. (8) suggest that differences in the literature may be explained by the variable exclusion of older patients with white matter lesions from some studies, because these lesions are likely to reduce the size of the large white matter tracts. In this respect, it is worth noting that our older subjects were relatively free of these lesions. In addition, however, our results suggest that while large, statistically significant declines are not seen in total callosal area, different portions of the CC are variably affected by aging, with the body showing the greatest effect.

Trends for smaller CC, genu, and body in women when compared to men were observed in all groups (AD, old, and young controls) but were not statistically significant. This finding is in agreement with a published large scale MRI study of sexual dimorphism of the CC, where no gender differences were found (19). In general, published studies evaluating the size of the CC in human brains have been contradictory, with one postmortem study reporting greater callosal area in women (7), which was not entirely confirmed in another post mortem study and MRI studies (8,19,24). Although technical factors and sample variability may explain some discrepancies, it is also possible that the relationship between CC size and total brain size may confound the interpretation of results.

Our results confirm a previous report showing reduced CC size in AD (25). However, genu, body, and splenium measurements were not included in previous studies of the effect of AD on CC. The region-specific decreases reported here are intriguing. The mechanism underlying a region-specific CC degeneration is not known. It is possible that progressive ventricular enlargement with resultant compression of callosal fibres contributes to the observed findings. Another likely explanation is that changes in CC thickness reflect degeneration of axons of cortical neurons lost during the disease process (23). If the latter explanation were true, and

given the topography of the CC (6), the implication would be that neuronal loss due to age and AD is more significant in frontal and parietal cortical regions (connected by the genu and body, respectively) than in the posterior cortical regions such as the occipital cortex connected via the splenium. In this respect, it is interesting to note that functional PET studies of AD patients repeatedly show decreases in brain metabolism in parietal, temporal, and frontal cortical areas with relative sparing of the occipital cortex (9,13), findings which parallel structural studies of the distribution of Alzheimer neuropathology in postmortem tissue (2,11,12).

ACKNOWLEDGEMENTS

This work was supported by NIH grants R01 AG07793 and P30 AG10129 from the National Institute on Aging, and by the Director, Office of Energy Research, Office of Health and Environmental Research of the United States Department of Energy under contract DE-AC03-76SF00098.

## REFERENCES

1. Barkovich, A. J.; Kjos, B. O. Normal postnatal development of the corpus callosum as demonstrated by MR imaging. AJNR 9:487–491; 1988.
2. Brun, A.; Englund, E. Regional pattern of degeneration in Alzheimer's disease: Neuronal loss and histopathological grading. Histopathology 5:549–564; 1981.
3. Chiarello, C. A house divided? Cognitive functioning with callosal agenesis. Brain Lang 11:128–158; 1980.
4. Creasey, H.; Schwartz, M.; Ferderickson, H.; Haxby, J. V.; Rapoport, S. I. Quantitative computed tomography in dementia of the Alzheimer type. Neurology 36:1563–1568; 1986.
5. DeCarli, C.; Kaye, J. A.; Horwitz, B.; Rapoport, S. I. Critical analysis of the use of computer-assisted transverse axial tomography to study human brain in aging and dementia of the Alzheimer type. Neurology 40:872–883; 1990.
6. De LaCoste, M. C.; Kirkpatrick, J. P.; Ross, E. D. Topography of the human corpus callosum. J. Neuropathol. Exp. Neurol. 44:578–591; 1985.
7. De Lacoste-Utamsing, C.; Holloway, R. L. Sexual dimorphism in the human corpus callosum. Science 216:1431–1432; 1982.
8. Doraiswamy, P. M.; Figiel, G. S.; Husain, M. M.; McDonald, W. M.; Shah, S. A.; Boyko, O. B.; Ellinwood, E. H.; Krishnan, K. R. R. Aging of the human corpus callosum: Magnetic resonance imaging in normal volunteers. J. Neuropsych. Clin. Neurosci. 3:392–397; 1991.
9. Duara, R.; Grady, C.; Haxby, J.; Sundaram, M.; Cutler, N. R.; Heston, L.; Moore, A.; Schlageter, N.; Larson, S.; Rapoport, S. I. Positron emission tomography in Alzheimer's disease. Neurology 36: 879–887; 1986.
10. Duara, R.; Kushch, A.; Gross-Glenn, K.; Barker, W. W.; Jallad, B.; Pascal, S.; Loewenstein, D. A.; Sheldon, J.; Rabin, M.; Levin, B.; Lubs, H. Neuroanatomic differences between dyslexic and normal readers on magnetic resonance imaging scans. Arch. Neurol. 48:410–416; 1991.
11. Esiri, M. M.; Pearson, R. C. A.; Steele, J. E.; Bowen, D. M.; Powell, T. P. S. A quantitative study of the neurofibrillary tangles and the choline acetyltransferase activity in the cerebral cortex and the amygdala in Alzheimer's disease. J. Neurol. Neurosurg. Psychiatr. 53:161–165; 1990.
12. Friedland, R. P.; Brun, A.; Budinger, T. F. Pathological and positron emission tomographic correlations in Alzheimer's disease. Lancet i: 228; 1985.
13. Friedland, R. P.; Budinger, T. F.; Ganz, E.; Yano, Y.; Mathis, C. A.; Koss, B.; Ober, B. A.; Huesman, R. H.; Derenzo, S. E. Regional cerebral metabolic alterations in dementia of the Alzheimer type: positron emission tomography with [18F]Fluorodeoxyglucose. J. Comput. Assist. Tomogr. 7:590–598; 1983.
14. Gazzaniga, M. S.; Kutas, M.; Van Petten, C.; Fendrich, R. Human callosal function: MRI-verified neuropsychological functions. Neurology 39:942–946; 1989.
15. Hauser, P.; Dauphinais, D.; Berrettini, W.; DeLisi, L. E.; Gelernter, J.; Post, R. M. Corpus callosum dimensions measured by magnetic resonance imaging in bipolar affective disorder and schizophrenia. Biol. Psychiatry 26:659–558; 1989.
16. Hayakawa, K.; Konishi, Y.; Matsuda, T.; Kuriyama, M.; Konishi, K.; Yamashita, K.; Okumura, R.; Daizaburo, H. Development and aging of brain midline structures: Assessment with MR imaging. Radiology 172:171–177; 1989.
17. Mathews, R. J.; Partain, C.; Prakash, R.; Kulkarni, M. V.; Logan, T. P.; Wilson, W. H. A study of the septum pellucidum and corpus callosum in schizophrenia with MR imaging. Acta Psychiatr. Scand. 72:414–421; 1985.
18. McKhann, G.; Drachman, D.; Folstein, M.; Katzman, R.; Price, D.; Stadlan, E. M. Clinical diagnosis of Alzheimer's disease: Report of the NINCDS-ADRDA work group under the auspices of Department of Health and Human Services Task Force on Alzheimer's Disease. Neurology 34:939–944; 1984.
19. Oppenheim, J. S.; Lee, B. C. P.; Nass, R.; Gazzaniga, M. S. No sex-related differences in human corpus callosum based on magnetic resonance imagery. Ann. Neurol. 21:604–606; 1987.
20. Rao, S. M.; Leo, G. J.; Haughton, V. M.; St. Aubin-Faubert, P.; Bernardin, L. Correlation of MRI with neuropsychological testing in multiple sclerosis. Neurology 39:161–166; 1989.
21. Simon, J. H.; Schiffer, R. B.; Rudick, R. A.; Herrdon, R. M. Quantification of MS-induced corpus callosum atrophy in vivo using MR imaging. AJNR 8:599–604; 1987.
22. Sperry, R. Some effects of disconnecting the cerebral hemispheres. Science 217:1223–1226; 1982.
23. Terry, R. D.; Peck, A.; DeTeresa, R.; Schechter, R.; Horoupian, D. S. Some morphometric aspects of the brain in senile dementia of the Alzheimer type. Ann. Neurol. 10:184–192; 1981.
24. Witelson, S. F. The brain connection: The corpus callosum is larger in left handers. Science 229:665–668; 1985.
25. Yoshi, F.; Duara, R. Size of the corpus callosum in normal subjects and patients with Alzheimer's disease: Magnetic resonance imaging study. Rinsho Shinkeigaku. 29:1–7; 1989.