

# Clinical Application of Measurement of Hippocampal Atrophy in Degenerative Diseases

Josephine Barnes<sup>1</sup> and Sebastien Ourselin<sup>1,2</sup> and Nick C. Fox<sup>1</sup>

<sup>1</sup> Dementia Research Centre, Institute of Neurology, University College London, UK,

<sup>2</sup> Centre for Medical Imaging Computing, Department of Medical Physics and Bioengineering, University College London, UK.

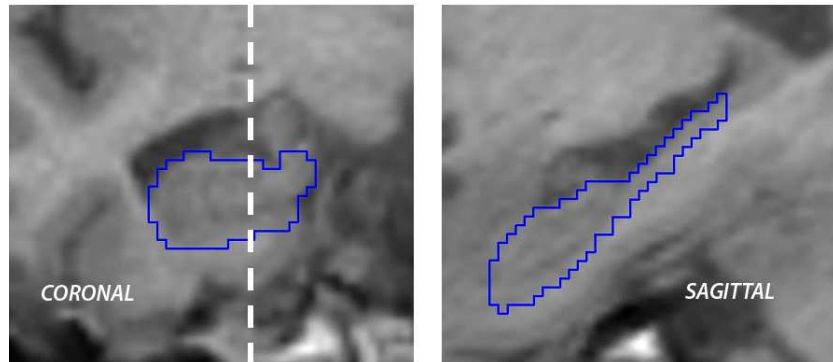
## 1 Manual measures

Hippocampal atrophy in degenerative dementias has been reported using structural imaging, both using computed tomography and magnetic resonance imaging (MRI), since the 1980s. Methods of quantifying hippocampal size began with estimating area of the hippocampus using a few 2D slices through the structure, extracted from the 3D volume. Over time, techniques were developed adopting a stereological approach, with hippocampal volume being measured using many "cuts" through the hippocampus. A large number of research groups use manual voluming of the hippocampus and this remains the "gold standard" of hippocampal volume measures.

Many groups use manual segmentation protocols which involve delineation of most but not all of the hippocampal formation. Anatomically, the hippocampal head and body which is bounded superiorly, medially and laterally by cerebrospinal fluid (CSF) and inferiorly by white matter: these are high contrast boundaries which are relatively easy to trace. More difficult is to separate the head of the hippocampus from the amygdala, and the alveus, which is best seen as a band of high signal intensity on the sagittal sections, is often used as a border. Part of the tail of the hippocampus is usually systematically excluded owing to difficulty in determining the posterior limits of the structure, and some groups have found the reliability of segmentation improved by its exclusion [1] (see Figure 1). The use of standard thresholds to exclude partial volume and CSF can also improve both reliability and speed of segmentation. Our group utilises thresholds of 70% mean brain intensity with whole brain regions having been previously determined by a semi-automated procedure.

Our early studies involved segmenting the structure in native space with no pre-processing to correct for scan inhomogeneity or head orientation. More recently, we have performed segmentations using N3-corrected [2] images registered to the MNI305 template [3] using a six degrees-of-freedom (dof) registration. We proposed this would have a theoretical advantage as all images would have reduced inhomogeneity and be in a similar orientation allowing comparable arbitrary cut-offs to be made between subjects. In addition, images are flipped across the mid-sagittal plane (mirror-image volumetry) to allow segmentation of left and right structures on the same side and reducing laterality bias. For

serial scans, scaling correction is also required. This can be achieved by either co-registration of all images using 9 or 12 dof or correction of serial measured volumes using estimates of total intracranial volume (TIV). Registration is advantageous as TIV estimation requires further operator-dependent segmentation. Co-registration of serial images also allows similar arbitrary protocol cut-offs to be made within subject, improving precision of measures of change. This can either be achieved by segmenting a series of regions relative to one another or copying and pasting baseline regions onto repeat scans followed by editing and intensity thresholding (cut-and-paste). The former of these two approaches is less biased as no borders are defined on paired scans prior to editing and therefore is used in our most recent studies. Segmentation software development has allowed images requiring delineation to be selected in advance so they can be loaded in a random order with blinding of the rater to diagnosis, scan laterality and if necessary chronological ordering of serial scans. In summary, our current processing stream for manual segmentation involves N3 correction of images followed by registration of images to the MNI305 template using six dof and then co-registration of serial images using either nine or twelve dof. This is followed loading of images or serial images in a random order and the blinding of the rater to all scan and patient information during segmentation.



**Fig. 1.** A manually-segmented hippocampus in coronal and sagittal views. The hashed line represents the location of the sagittal slice on the right. In the sagittal view the exclusion of the tail is apparent.

## 2 Familial AD : a model of anatomical change in AD

In order to assess the earliest features of change in AD, imaging needs to be performed on pre-symptomatic or pre-clinical AD: either a familial form where diagnosis is certain or subjects with mild cognitive impairment who have a high

risk of converting to AD. We have investigated a small group of subjects with an autosomal dominant form of AD. Such subjects are important to study as the level of certainty concerning their pathology is very high and owing to the early onset of the disease (often in their 30s-50s), and there are fewer existing co-morbidities that may affect the disease progression.

The first of such studies investigated seven at-risk members who underwent serial scanning and neuropsychological assessments over a three year period. Over this time, three of these subjects became clinically affected. Manual assessment of hippocampal volumes was performed on images in native space with no pre-processing for image inhomogeneity. Hippocampal volumes were normalised for TIV. Results showed asymmetrical atrophy in those subjects prior to appearance of symptoms together with verbal and visual memory loss. Losses of up to 8% per year in hippocampal volume occurred in these subjects in the two years over which these symptoms first appeared [4].

A further study investigated the sites of earliest change in AD using five pre-symptomatic AD subjects with two serial images. Hippocampi were segmented in native space but co-registered to allow for scaling changes. Serial hippocampi were segmented as a pair with the baseline hippocampi pasted onto repeat images and intensity thresholding applied in order to improve consistency. Mirror-image volumetry, where scans were flipped to allow left-sided hippocampi to be segmented on the right, was also utilised to reduce laterality bias. Averaged hippocampal and entorhinal cortex measures were 16.6% (95% CI 3.3 – 28.0%) lower than age- and gender matched controls at baseline whereas other structures measured: whole brain, temporal lobe and lateral ventricles were no different. Atrophy rates of all structures were different between controls and the familial AD subjects ( $p < 0.05$ ) with the averaged entorhinal cortex and hippocampal rates being 5.1% (95%CI 3.0 – 7.1%) greater in patients. These results allowed linear extrapolation backward to estimate the beginning of hippocampal and entorhinal cortex losses. This was suggested to be 3.5 years (95% CI, 0.7 – 7.5 years) before onset, when all patients were asymptomatic but is likely to be a underestimate owing to the small sample size and imprecision of measures [5].

A recent study of a larger dataset of familial AD subjects with a total of 41 serial scans (range of 3 – 8 per patient) at different clinical stages were assessed comparing these to 24 age- and gender-matched controls (54 scans 2 – 4 per patient). Using hierarchical statistical models, differences in hippocampal and whole brain volumes were assessed. Hippocampi were outlined registered to the MNI305 template and to each other in series (2 – 8 scans), in a random chronological order. This was performed by segmenting one hippocampus and then all further hippocampi were measured with the initial segmentation as a reference. Such delineation was always performed on the right side of the screen using mirror-image volumetry (flipping left hippocampal series to appear as right) in order to reduce laterality bias. Results here suggested changes in hippocampal rate of atrophy occurred 5.5 years prior to disease onset whereas changes in whole brain rate of atrophy occurred 3.5 years prior to onset. These atrophy rates in familial AD subjects accelerated with disease progression ( $p < 0.01$ ).

When considering volumes rather than rates, differences in mean hippocampal volume between familial AD subjects and controls became significant 3 years before clinical diagnosis, whereas differences in mean brain volumes became significant only 1 year before diagnosis [6]. These familial AD studies suggest that not only is hippocampal atrophy an early feature of disease as suggested by pathological studies [7] but that these changes can be measured in vivo using MRI and that these are suggestive of a window of opportunity for both diagnosis and treatment of AD. Furthermore, the latter study [6] suggests that atrophy rates in disease accelerate with progression and that longitudinal measures may be more useful than cross-sectional measures at detecting early differences of affected subjects from normal controls.

### 3 Sporadic AD and normal ageing

Familial AD subjects, although an important group to study, only make up a small proportion ( $< 1\%$ ) of the total number of people with AD. Most patients are older, may have co-existing disease, and owing to a lack of a positive diagnostic test, have lower certainty of the pathology causing disease. Manual hippocampal voluming and quantification of loss over time has also been used to answer specific questions regarding what is normal tissue loss with age, symmetry of structure in AD and differential involvement of the hippocampus in different neurodegenerative diseases.

*Normal ageing.* To understand what is happening in sporadic AD where disease onset is typically over 65 years, we have to establish what happens to normal brains with age. Therefore a number of studies have been performed using normal controls of different ages in order to model changes with age. We studied cross-sectional data from a group of subjects ( $n = 39$ ) ranging in age from 31 to 84 years. A number of different brain regions were studied including the hippocampus and both cross-sectional and longitudinal data was analysed. Hippocampal volumes were outlined in native space registered to each other in a random subject order. Follow-up hippocampal volumes were quantified by the cut-and-paste method: copying the baseline region onto the repeat scan and editing and intensity thresholding this region. We found that along with whole brain and temporal lobe, decreases in hippocampal volume with age were found. Increased atrophy rates with increasing age was also found with evidence for an acceleration in atrophy rates in all structures with the most marked changes occurring over 70 years which was most marked in the ventricles ( $p < 0.001$ ) and hippocampi ( $p < 0.01$ ). Such findings are important to consider when assessing neurodegenerative diseases as the older a subject group is, the more overlap between disease groups and normal controls may be seen [8].

*Asymmetry of hippocampal volumes in AD.* In response to some studies which suggested that changes in hippocampal asymmetry was a marker of AD [9] we used manual volumetric analyses to establish whether hippocampal asymmetry changed in AD. A mixed group of sporadic and familial AD subjects ( $n = 32$ ) were included with a large group of age- and gender-matched controls ( $n =$

50). Both cross-sectional and longitudinal manual hippocampal measures were included in the study as defined in the normal ageing study above. The results showed a non-significant trend for right > left ( $R > L$ ) asymmetry in controls at both time points. AD subjects showed a similar trend for  $R > L$  asymmetry at baseline, but not at repeat ( $p = 0.739$ ). Atrophy rates for left and right hippocampi were 1.2% (0.5–1.8%) and 1.1% (0.5–1.8%) for the controls, and 4.6% (3.3–6.0%) and 6.3% (4.9–7.8%) for AD subjects. There was no significant asymmetry in atrophy rates in controls ( $p = 0.9$ ), but borderline significantly higher atrophy rates in the right hippocampus of the AD group compared to the left ( $p = 0.05$ ). Although we found minor  $R > L$  asymmetry in hippocampal volumes in controls and some evidence to suggest that there was a change in asymmetry during the progression of AD, we did not find that such changes in asymmetry was a reliable marker of disease progression [10].

*Relative involvement of the hippocampus in neurodegenerative diseases.* The hippocampus is not specifically affected by AD pathology as other neurodegenerative diseases also cause apparent atrophy. We assessed hippocampal volume relative to other temporal lobe structures in two degenerative conditions characterised by temporal lobe pathology: AD and the temporal variant of frontotemporal lobar degeneration (FTLD), semantic dementia. We assessed volumes of amygdala, hippocampus, entorhinal cortex, parahippocampal gyrus, fusiform gyrus, and superior, middle, and inferior temporal gyri in ten subjects from each disease group and matched controls. Structures were measured in native space and mirror-image volumetry was used with scans loaded in a random order and with the rater blind to scan laterality and subject diagnosis. All volumes were corrected for head size using TIV. Semantic dementia showed asymmetrical temporal lobe atrophy, with greater left-sided and anterior damage. All left anterior temporal lobe structures were affected in semantic dementia, with the entorhinal cortex, amygdala, middle and inferior temporal gyri, and fusiform gyrus the most severely damaged. Asymmetrical, predominantly anterior hippocampal atrophy was also present. In AD, there was symmetrical atrophy of the entorhinal cortex, hippocampus, and amygdala, with no evidence of an anteroposterior gradient in the distribution of temporal lobe or hippocampal atrophy. Such a study shows the marked difference in the distribution of temporal lobe atrophy in semantic dementia and AD [11].

In another study we assessed the relative diagnostic power of the amygdala and hippocampus in FTLD and AD. With the amygdala being more anterior to the hippocampus, we considered it may be possible for this structure to better discriminate FTLD subjects from AD and controls. To be sure of disease diagnosis we used a group of post-mortem proven subjects ( $n = 10$  AD,  $n = 17$  FTLD and  $n = 10$  controls). Hippocampi and amygdala were measured on images normalised to the MNI305 template and using mirror-image volumetry with subjects scans loaded in random order and with the rater blind to scan laterality and subject diagnosis. Both structures were corrected for head size using TIV. Mean amygdala and hippocampal volumes were 15.0% (95% CI, 4.2%–24.5%) and 6.4% (95% CI, 5.9%–25.6%) respectively lower in the AD than in the control

group. In FTLN, the equivalent differences were 43.1% (95% CI, 31.9% – 52.6%) in the amygdala and 36.1% (95% CI, 27.5% – 43.7%) in the hippocampus. Volumes were significantly lower in the FTLN than in the AD group ( $p < 0.01$  in both regions). Within FTLN clinical subgroups, there was evidence of a difference in pattern of atrophy with greater asymmetry (left smaller than right) in semantic dementia compared with frontal variant FTLN ( $p < 0.001$ ). On average, the left hippocampus was 14% smaller in semantic dementia than in frontal variant FTLN, whereas the right hippocampus was 37% larger. On average, the left amygdala was 39% smaller in semantic dementia than in frontal variant FTLN, whereas the right amygdala was only 1% smaller [12].

Other parts of the limbic system are known to be affected in AD and FTLN including the cingulate gyrus. Therefore we compared rates of hippocampal atrophy with cingulate gyrus atrophy in post-mortem proven AD ( $n = 19$ ), FTLN ( $n = 8$ ) and controls ( $n = 11$ ) in order to be certain of disease diagnosis. Hippocampi and cingulate gyri were measured on scans normalised to the MNI305 template and co-registered together. Mirror-image volumetry was used to segment left and right structures and scans were loaded in random chronological order such that the rater blind to the time-point, scan laterality and subject diagnosis. One of the paired serial hippocampi was segmented initially and the remaining structure was segmented with reference to the original segmentation to ensure similar arbitrary anatomical decisions were made leading to greater precision of change. Cingulate changes were determined in a similar way. Mean (SD) annualised atrophy rates as a percentage per year in the cingulate in controls, AD and FTLN were  $-0.3$  (1.2),  $5.9$  (3.5), and  $8.6$  (4.1), respectively. Hippocampal atrophy rates in controls, AD and FTLN were  $-0.1$  (0.8),  $3.4$  (2.2), and  $5.2$  (5.4), respectively. Atrophy rates were significantly higher in the cingulate and hippocampi in AD and FTLN compared with controls ( $p < 0.01$ ). Significantly better discrimination between AD and controls was obtained by hippocampal rather than cingulate rates. We concluded that cingulate atrophy is as significant a feature of AD and FTLN as hippocampal atrophy [13].

These studies highlight the fact that hippocampal atrophy is not specific to AD or FTLN. However, measurement of this structure has been widely studied and attempts to automate its measurement have been made as a result.

## 4 Shorter interval atrophy rates

Most atrophy rates quantified in the literature are generated using scans with an interval of at least one year. Should atrophy rates be found to be more diagnostically useful than volumes, patients will have to return to clinic for follow-up examinations. The shorter this interval from baseline assessment, the better for the patient and their care. Such gains would also be useful for the design of clinical trials as the efficacy of treatment may be assessed earlier. However, with shorter intervals the signal-to-noise ratio reduces as the amount of change expected over the short intervals decreases. We assessed whether atrophy rates could be measured using shorter intervals. We measured hippocampi on baseline-, 6- and

12-month scans in a group of AD ( $n = 36$ ) and control subjects ( $n = 20$ ). Hippocampi were measured on N3 corrected scans normalised to the MNI305 template and co-registered together. Mirror-image volumetry was used for left and right hippocampi and scans were loaded in random chronological order with the rater blind to the time-point, scan laterality and subject diagnosis. We found that mean annualised atrophy rates using 6-month intervals were comparable at a group level to those generated from a 12-month interval. Higher variance was seen using shorter intervals, although this was only significant in the control group. This suggests rapid diagnosis, and tracking of disease progression in a clinical trial may be possible over these shorter intervals [14]. However, these measures are time-consuming and somewhat imprecise. Stability of acquisition is crucial and more precise methods of quantification of hippocampal change would make short interval assessment more realistic.

## 5 Application of manual measures to multi-centre clinical trials

Owing to the potential disease-specific effects of putative treatments for AD, serial assessment of hippocampal volumes have been performed in clinical trials. One phase II trial assessed the efficacy of active immunisation of one of the pathological components of AD: beta-amyloid (A $\beta$ ). The trial was stopped early due to encephalitis caused in some treated subjects. Whole brain, and hippocampal atrophy rates together with lateral ventricle expansion in both placebo and those subjects who responded to treatment as determined by an antibody titre anti-AN1792 IgG titer of  $>$  or  $= 1 : 2, 200$ . Hippocampi were measured by three raters, with inhomogeneity corrected images registered to MNI305 template and follow-up images registered to baseline. All hippocampi were measured using mirror-image volumetry for the left and right structures with raters blind to randomisation and chronological order of scans. Results focussed on the 45 responders and 57 placebo with paired serial MRI. Those subjects who responded to treatment had greater whole brain decrease ( $3.12 \pm 1.98$  vs  $2.04 \pm 1.74\%$ ;  $p = 0.007$ ) and ventricular expansion ( $1.10 \pm 0.75$  vs  $0.48 \pm 0.40\%$ ;  $p < 0.001$ ) over the scanning interval but a non-significant greater hippocampal volume decrease ( $3.78 \pm 2.63$  vs  $2.86 \pm 3.19\%$ ;  $p = 0.124$ ) than placebo patients. A dissociation between increased losses in brain volume and worsening cognitive performance was also found; a composite z score across a Neuropsychological Test Battery showed differences with antibody responders remaining unchanged and placebo worsening ( $0.03 \pm 0.39$  vs  $-0.24 \pm 0.45$  respectively;  $p = 0.008$ ). This study highlights the requirement of having reliable serial measures of hippocampal loss for clinical trials in order that disease-specific effects can be assessed. It may be that immunisation against A $\beta$  produced relatively less changes in hippocampal volume since hippocampal pathology in AD is mostly caused by neurofibrillary tangles. However it may be that our manual measures were not precise enough to detect the true changes that were occurring [15].

Further investigation into the MRI measures and neuropsychological data within the placebo arm of this trial showed that hippocampal measures did not correlate with any of the neuropsychological assessments performed: Mini-Mental State Examination, AD Assessment Scale-Cognitive Subscale, Disability Assessment for Dementia, AD Cooperative Study- Clinical Global Impression of Change and Clinical Dementia Rating. Correlations did exist between whole brain and ventricular atrophy rates and these scores. This is important for the planning of future trials as it may be that the lack of correlation between hippocampal measures and psychology reflects either the extensive damage of the hippocampus by the time the subjects had enrolled, the lack of precision of the hippocampal measures and the potential great influence of cortical degeneration [16].

## 6 Automation of hippocampal measures

Tracking disease-specific changes over time is important for prediction and diagnosis, and increasingly this is important for clinical trials as many time-points are often used to increase precision of changes. If hippocampal measures are to be used either in the clinic or for clinical trials, automation of these changes is essential. As a result a number of groups have been working on a number of ways of automating the detection of change over time in the hippocampus. We have achieved this by both using a manual baseline hippocampal measure as a starting region, and also by assessing ways of automatically generating this initial baseline hippocampal estimate.

A manual baseline hippocampus is the best estimate of initial hippocampal volume. With a combination of global registration of serial scans to this baseline and application of direct methods of atrophy detection assessing boundary shift [17], an estimate of change of volume of the hippocampus can be made. One study compared manually-derived atrophy rates with boundary shift integral (BSI) derived atrophy rates in a group of AD ( $n = 32$ ) and controls ( $n = 47$ ). Hippocampi were measured on scans co-registered together in native space. Mirror-image volumetry was used for left and right hippocampi. Segmentations were performed on baseline images and the repeat volumes were calculated by the cut-and-paste method. Raters were blind to scan laterality and subject diagnosis. A rigid registration was then performed over the hippocampus using the baseline hippocampal region to better align the serial hippocampi, and boundary shifts were quantified using the manual baseline region and the hippocampus-hippocampus registered images. Hippocampal BSI (HBSI) significantly reduced the mean rate ( $p < 0.01$ ) and variation in controls ( $p < 0.001$ ) and increased group separation between AD cases and controls. From logistic regression models, a 1% increase in HBSI atrophy rates was associated with an 11-fold (CI 3, 36) increase in the odds of a diagnosis of AD. For manually derived atrophy rates, the equivalent odds ratio was 3 (CI 2,4). This initial study using boundary shift in the hippocampus showed promise that this method may be

useful at automatically detecting change over time without the need for segmentation of serial regions [18].

Non-linear (fluid) registration has also been shown to be useful at detecting such hippocampal change [19] and may be more sensitive at quantifying change of hippocampal volume at the grey-white matter borders which may be missed by boundary shift measures. Fluid change can either be calculated by performing a fluid registration and inverting the deformation field and applying it to the baseline hippocampal region (propagation) or integrating the Jacobian determinants over the baseline hippocampal region. In an early study 15 normal controls, and 12 AD subjects were used with serial images. Manual atrophy rates were calculated in native space using registered scan pairs utilising the cut-and-paste method where repeat volumes generated by pasting the baseline hippocampal region and editing and intensity thresholding that region. Manual rates were compared with those generated using non-linear registration (both Jacobian and propagation). The scan-rescan volumetric consistency of propagation was shown to be superior to human serial segmentors (approximately 2%). The mean absolute volume difference between fluid propagation and manual segmentation was 0.7%.

As a result of the potential viability of the technique, it was applied to a larger group [20]. Thirty two AD subjects and 55 controls were assessed comparing both manual rates generated in a similar way to the smaller study and fluid (Jacobian and propagation) atrophy rates. Methodological changes such as adding a local hippocampal-hippocampal rigid registration step to better align hippocampi prior to fluid registration and adding an exit criterion were also described in this study. In AD patients, the mean (SD) atrophy rates for manual, fluidly-propagated, and Jacobian methods were 5.09 (3.59), 5.34 (3.43), and 3.55 (2.70) (% per year). In controls, atrophy rates were 1.31 (2.00), 0.89 (0.75), and 0.56 (1.12) (% per year). In this study we found fluid propagation methods to give results similar to the manual measures whereas Jacobian rates were smaller in mean and variance compared with manual measures. Both fluid measures were superior to manual in discriminating the two groups. Such methods have also been successfully applied to MCI populations showing greater reliability than manual measures [21].

We have also made attempts to automate the estimate of the baseline hippocampal region. One study used a single-person template (another subject from a study then excluded from the analysis) to transfer hippocampal labels to baseline images using a process of brain-to-brain affine and hippocampus-to-hippocampus rigid registration. This region was then used to estimate hippocampal losses using boundary shift measures [22]. Thirty six AD and 19 control subjects were investigated. Manually-derived hippocampi were measured on N3-corrected scans normalised to the MNI305 template and co-registered together. Mirror-image volumetry for left and right hippocampi was used and scans were loaded in random chronological order with the rater blind to the time-point, scan laterality and subject diagnosis. Automated methods were compared with entirely manual rates and semi-automated rates generated by BSI measures using

the manual baseline region. In controls, mean (S.D.) atrophy rates for manual, semi-automated, and automated methods were 18.1 (53.5), 15.3 (50.2) and 11.3 (50.4) mm<sup>3</sup> loss per year, respectively. In AD patients these rates were 174.6 (106.5) 159.4 (101.2) and 172.1 (123.1) mm<sup>3</sup> loss per year, respectively. The automated method was a significant predictor of disease ( $p = 0.001$ ) and gave similar group discrimination compared with semi-automated and manual methods.

These methods were then extended to a library of hippocampal templates and a leave-one-out experiment was performed using the same subjects [23]. Further to this, morphological operations were performed on these regions to improve accuracy of these baseline hippocampal estimates. A template library with morphological operations showed better accuracy of hippocampal segmentation compared with manual at baseline than single-person template methods (voxel similarity of 0.69 (0.05) and 0.72 (0.06) in controls and probable AD subjects) and using these automated baselines rates of atrophy were calculated using a) boundary shift, b) Jacobian change and (c) fluid propagation. Atrophy rates within these regions were most similar to the manual rates using the boundary shift integral (mean difference from manual rate 0.03% (1.29) in controls and 0.48% (2.44) in AD). This study showed that a template library segmentation approach, together with morphological operations, could provide a way of introducing variability into a single-person template method of hippocampal segmentation without losing resolution caused by averaging a number of subjects. The change over time can then be calculated automatically using boundary shift or fluid measures, with boundary shift giving most similar results to manual.

## 7 Context

Finally, we have performed a meta-analysis of existing longitudinal atrophy rates in AD in peer-reviewed literature [24]. We systematically reviewed and appraised studies which reported atrophy rates in the hippocampus in AD. However, cohorts and methods used to determine such rates are heterogeneous, leading to differences in reported annualised rates. We contacted all authors of studies we wanted to include to both confirm the results and provide missing data. We included nine studies from centres, with data from a total of 595 AD and 212 matched controls. Mean (95% CIs) annualised hippocampal atrophy rates were found to be 4.66% (95% CI 3.92, 5.40) for AD subjects and 1.41% (0.52, 2.30) for controls. The difference between AD and control subject in this rate was 3.33% (1.73, 4.94).

## 8 Conclusions

A large number of studies involving hippocampal measures have been performed which have revealed important clinical features of AD, investigated hippocampal

measures as a biomarker and improved methodology of hippocampal measurement. There are still many gains to be made, particularly with respect to hippocampal measurement and it may be that combining our methods with those from other groups can improve these measures for individual subjects in the clinic and groups of subjects in clinical trials.

## References

1. Watson, C., Andermann, F., Gloor, P., Jones-Gotman, M., Peters, T., Evans, A., Olivier, A., Melanson, D., Leroux, G.: Anatomic basis of amygdaloid and hippocampal volume measurement by magnetic resonance imaging. *Neurol* **42** (1992) 1743–1750
2. Sled, J., Zijdenbos, A., Evans, A.: A nonparametric method for automatic correction of intensity nonuniformity in MRI data. *IEEE Trans Med Imaging* **17** (1998) 87–97
3. Mazziotta, J., Toga, A., Evans, A., Fox, P., Lancaster, J.: A probabilistic atlas of the human brain: Theory and rationale for its development. *Neuroimage* **2** (1995) 89–101
4. Fox, N., Warrington, E., Freeborough, P., Hartikainen, P., Kennedy, A., Stevens, J., Rossor, M.: Presymptomatic hippocampal atrophy in Alzheimer's disease: a longitudinal MRI study. *Brain* **119** (1996) 2001–2007
5. Schott, J., Fox, N., Frost, C., Scahill, R., Janssen, J., Chan, D., Jenkins, R., Rossor, M.: Assessing the onset of structural change in familial Alzheimer's disease. *Ann Neurol* **53** (2003) 181–188
6. Ridha, B., Barnes, J., Bartlett, J., Godbolt, A., Pepple, T., Rossor, M., Fox, N.: Tracking atrophy progression in familial Alzheimer's disease: a serial MRI study. *Lancet Neurol* **5** (2006) 828–834
7. Braak, H., Braak, E.: Neuropathological staging of Alzheimer-related changes. *Acta Neuropathologica* **82** (1991) 239–259
8. Scahill, R., Frost, C., Jenkins, R., Whitwell, J., Rossor, M., Fox, N.: A longitudinal study of brain volume changes in normal aging using serial registered magnetic resonance imaging. *Arch Neurol* **60** (2003) 989–994
9. Geroldi, C., Laakso, M., DeCarli, C., Beltramello, A., Bianchetti, A., Soininen, H., Trabucchi, M., Frisoni, G.: Apolipoprotein E genotype and hippocampal asymmetry in Alzheimer's disease: a volumetric MRI study. *J Neurol Neurosurg Psychiatry* **68** (2000) 93–96
10. Barnes, J., Scahill, R., Schott, J., Frost, C., MN, M.R., Fox, N.: Does Alzheimer's disease affect hippocampal asymmetry? Evidence from a cross-sectional and longitudinal volumetric MRI study. *Dement Geriatr Cogn Disord* **19** (2005) 338–344
11. Chan, D., Fox, N., Scahill, R., Crum, W., Whitwell, J., Leschziner, G., Rossor, A., Stevens, J., Cicolotti, L., Rossor, M.: Patterns of temporal lobe atrophy in semantic dementia and Alzheimer's disease. *Ann Neurol* **49** (2001) 433–442
12. Barnes, J., Whitwell, J., Frost, C., Josephs, K., Rossor, M., Fox, N.: Measurements of the amygdala and hippocampus in pathologically confirmed Alzheimer disease and frontotemporal lobar degeneration. *Arch Neurol* **63** (2006) 1434–1439
13. Barnes, J., Godbolt, A., Frost, C., Boyes, R., Jones, B., Scahill, R., Rossor, M., Fox, N.: Atrophy rates of the cingulate gyrus and hippocampus in AD and FTLD. *Neurobiol Aging* **28** (2007) 20–28

14. Barnes, J., Scahill, R., Frost, C., Schott, J., Rossor, M., Fox, N.: Increased hippocampal atrophy rates in AD over six months using serial MR imaging. *Neurobiol Aging* **29** (2008) 1199–1203
15. Fox, N., Black, R., Gilman, S., Rossor, M., Griffith, S., Jenkins, L., Koller, M.: Effects of a beta immunization (AN1792) on MRI measures of cerebral volume in Alzheimer disease. *Neurol* **64** (2005) 1563–1572
16. Ridha, B., Anderson, V., Barnes, J., Boyes, R., Price, S., Rossor, M., Whitwell, J., Jenkins, L., Black, R., Grundman, M., Fox, N.: Volumetric MRI and cognitive measures in Alzheimer disease : comparison of markers of progression. *J Neurol* **255** (2008) 567–574
17. Freeborough, P., Fox, N.: The boundary shift integral: an accurate and robust measure of cerebral volume changes from registered repeat MRI. *IEEE Trans Med Imaging* **16** (1997) 623–629
18. Barnes, J., Scahill, R., Boyes, R., Frost, C., Lewis, E., Rossor, C., Rossor, M., Fox, N.: Differentiating AD from aging using semiautomated measurement of hippocampal atrophy rates. *Neuroimage* **23** (2004) 574–581
19. Crum, W., Scahill, R., Fox, N.: Automated hippocampal segmentation by regional fluid registration of serial MRI: validation and application in Alzheimer’s disease. *Neuroimage* **13** (2001) 847–855
20. Barnes, J., Lewis, E., Scahill, R., Bartlett, J., Frost, C., Schott, J., Rossor, M., Fox, N.: Automated measurement of hippocampal atrophy rates using fluid-registered serial MRI in AD and controls. *JCAT* **31** (2007) 581–587
21. van de Pol, L., Barnes, J., Scahill, R., Frost, C., Lewis, E., Boyes, R., van Schijndel, R., Scheltens, P., Fox, N., Barkhof, F.: Improved reliability of hippocampal atrophy rate measurement in mild cognitive impairment using fluid registration. *Neuroimage* **34** (2007) 1036–1041
22. Barnes, J., Boyes, R., Lewis, E., Schott, J., Frost, C., Scahill, R., Fox, N.: Automatic calculation of hippocampal atrophy rates using a hippocampal template and the boundary shift integral. *Neurobiol Aging* **28** (2007) 1657–1663
23. Barnes, J., Foster, J., Boyes, R., Pepple, T., Moore, E., Schott, J., Frost, C., Scahill, R., Fox, N.: A comparison of methods for the automated calculation of volumes and atrophy rates in the hippocampus. *Neuroimage* **40** (2008) 1655–1671
24. Barnes, J., Bartlett, J., van de Pol, L., Loy, C., Scahill, R., Frost, C., Thompson, P., Fox, N.: A meta-analysis of hippocampal atrophy rates in Alzheimer’s disease. *Neurobiol Aging* **In Press**