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The hippocampal-to-ventricle ratio (HVR): Presentation of a manual segmentation protocol and preliminary evidence

Dorothee Schoemaker [a,b], Claudia Buss [c], Sandra Pietrantonio [b], Larah Maunder [d], Silka Dawn Freiesleben [b], Johanna Hartmann [e], D. Louis Collins [f], Sonia Lupien [g], Jens C. Pruessner [b,e,*]

[a] Massachusetts General Hospital, Harvard Medical School, Boston, USA
[b] McGill Centre for Studies in Aging & Douglas Mental Health Institute, Faculty of Medicine, McGill University, Montreal, Canada
[c] Department of Psychology, Charite Berlin, Berlin, Germany
[d] Department of Psychology, Queen's University, Kingston, Canada
[e] Department of Psychology, University of Constance, Constance, Germany
[f] McConnell Brain Imaging Center, Montreal Neurological Institute, McGill University, Montreal, Canada
[g] Centre for Studies on Human Stress, Montreal Mental Health University Institute, Faculty of Medicine, Université de Montréal, Montreal, Canada

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ABSTRACT

Disentangling age-related changes from developmental variations in hippocampal volume has proven challenging. This article presents a manual segmentation protocol for the hippocampal-to-ventricle ratio (HVR), a measure combining the assessment of hippocampal volume with surrounding ventricular volume. By providing in a single measure both a standard volumetric assessment of the hippocampus and an approximation of volume loss, based on ventricular enlargement, we believe the HVR provides a superior cross-sectional estimation of hippocampal structural integrity.

In a first attempt to validate this measure, we contrasted the HVR and standard hippocampal volume in their associations with age and memory performance in two independent cohorts of healthy aging individuals. The first cohort consisted in 50 cognitively normal subjects (mean age: 66.8 years, SD: 4.96, range: 60–75 years), while the second cohort included 88 cognitively normal subjects (mean age: 65.06 years, SD: 6.42, range: 55–80 years). We showed that the manual segmentation protocol for the HVR can be implemented with high reliability. In both cohorts, the HVR showed stronger negative associations with age than standard hippocampal volume. Correlations with memory performance were also numerically superior with the HVR than standard hippocampal volume, across the two cohorts.

These findings support an added benefit of using the HVR over standard hippocampal volume when examining relationships with age or memory function in aging individuals. Although further validation is required, we propose that the computation of the HVR is a promising method to improve the evaluation of hippocampal integrity from cross-sectional MR images.

1. Introduction

The hippocampus, a medial temporal lobe structure involved in the formation of memory (Eichenbaum, 2004), is affected by various normal and pathological processes associated with advancing age. Studies investigating age-related neurodegeneration often use hippocampal volume estimates from structural Magnetic Resonance Imaging (MRI) as a proxy for structural and functional integrity (Bilgel et al., 2019; Estévez-Santé and Jiménez-Huete, 2019; Khlif et al., 2019). However, evidence demonstrates that hippocampal volume is highly variable in the population. A study on a large and healthy adult cohort showed a similar variability in hippocampal volumes in young than in older adults (Lupien et al., 2007). These results revealed that 25% of subjects aged 18–24 years presented hippocampal volumes as small as those observed in the average older adult aged 60–75 years. The large inter-individual variability in hippocampal volumes in young adulthood is indicative of a significant impact of developmental factors that are distinct from age-related changes. Several human and animal studies provide

* Corresponding author. Box 905, Department of Psychology University of Constance University Street 10, 78457 Constance, Germany.
E-mail address: jens.pruessner@uni-konstanz.de (J.C. Pruessner).

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empirical support for this idea, identifying factors such as genetic susceptibility (Sullivan et al., 2001), environment enrichment (Kempermann et al., 1997, 1998; Mlynarik et al., 2004), nutrition (Zainuddin and Thuret, 2012), and stress (Gould and Tanapat, 1999; Mirescu et al., 2004; Pruessner et al., 2005) as significant determinants of hippocampal volume.

The important inter-individual variation in hippocampal volume across subjects limits the validity and sensitivity of absolute volume as an indicator of hippocampal integrity in cross-sectional studies investigating age-related changes. In other words, a small hippocampal volume is not necessarily representative of a loss of integrity, but could rather reflect developmental predispositions. Consistently, a linear association between hippocampal volume and cognition has not been consistently observed. In a meta-analysis examining the evidence for a structure-function relationship between hippocampal volume and memory ability across the lifespan, Van Petten (2004) found little support for a "bigger is better" hypothesis. Instead, this review highlighted a pattern of substantial variability in the association between hippocampus size and memory performance in healthy older adults. In contrast, studies have repeatedly reported a linear relationship between memory function and hippocampal volume in pathological aging, especially in the presence of more severe forms of neurodegeneration, such as Alzheimer's Disease (Köhler et al., 1998; de Toledo-Morrell et al., 2000; Mizuno et al., 2000; Barber et al., 2001; Mungas et al., 2002; Jack et al., 2004) and Mild Cognitive Impairment (MCI) (e.g. Jack et al., 2000). Refining volumetric measures of hippocampal integrity might allow reducing discrepancies in results across studies.

It can be assumed that, when hippocampal volume is at its peak, surrounding cerebrospinal fluid (CSF) space is minimal. Conversely, hippocampal atrophy is accompanied by an increase in the volume of surrounding CSF (i.e. perihippocampal CSF; as an illustration see Fig. 1). Ventricular enlargement can therefore serve as an indicator of volume loss. The use of ventricular imaging to detect the presence of atrophy is not new: believed to be secondary to shrinkage of parenchymal brain tissue, ventricular expansion is commonly present in neurodegenerative disorders (Dalaker et al., 2011; Apostolova et al., 2012a,b; Mak et al., 2017; Seif et al., 2018). Passive enlargement of the lateral ventricles, in particular, has been suggested as a potential biomarker of CNS disease progression, including Alzheimer's Disease and MCI (De Leon et al., 1993; Thompson et al., 2004; Apostolova et al., 2012a,b; Apostolova et al., 2013; Bartos et al., 2019). A study by Chou et al. (2009) showed that measures of ventricular enlargement can accurately differentiate control subjects from MCI or Alzheimer's Disease patients. Similarly, De Leon et al. (1993) demonstrated that dilation of the perihippocampal fissure, secondary to hippocampal atrophy, was a superior predictor of impending AD than ratings of cortical atrophy. Taken together, these findings support the relevance of ventricular enlargement as a marker of brain atrophy associated with normal or pathological aging processes.

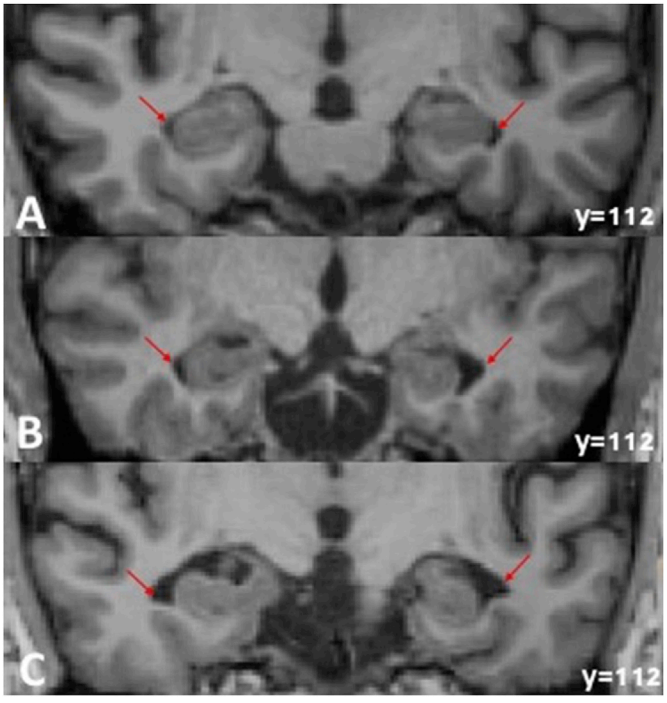

Fig. 1. Illustration of perihippocampal ventricular enlargement in normal aging and mild cognitive impairment

Fig. 1. *MR images (MPRAGE) showing coronal sections of the medial temporal lobe in 3 subjects. Subject A is a healthy 33 year-old male; subject B is a cognitively normal 70 year-old male; subject C is a 71 year-old male presenting with Mild Cognitive Impairments. The red arrows bring attention to the notable difference in perihippocampal ventricular space across subjects.*

To overcome the previously outlined limitations associated with the use of absolute hippocampal volume measurements as a proxy for hippocampal structural integrity, computing a ratio of prehippocampal CSF to hippocampal volume might provide valuable information about volume change from its maximal value. It is expected that such ratio would convey a superior indication of volume change when relying on cross-sectional assessments. In support of this notion, a recent study looking at various ratios combining brain volumes computed with FreeSurfer determined the "hippocampal on inferior lateral ventricle" ratio was the most sensitive and specific in differentiating AD patients from normal controls (Bartos A et al., 2019). In accordance, we hypothesize that such ratio would allow for a clearer distinction between normally occurring variations in absolute hippocampal volume across individuals and changes in hippocampus volume associated with normal or pathological aging.

In this article, we present and describe a manual segmentation protocol to compute a *hippocampal-to-ventricle ratio* (HVR). In contrast to absolute volume, this measure combines both an estimation of structural volumes (i.e. hippocampus volume) and of ventricular enlargement in relevant areas (i.e. perihippocampal ventricular space). We hypothesized that this ratio represents a superior predictor of age-related structural changes and cognitive performance than the absolute hippocampal volume. To provide preliminary evidence for the relevance of this measure, we contrasted the HVR and absolute hippocampal volume in their associations with age and memory performance in two datasets from distinct cohorts of healthy elderly subjects.

2. Materials and methods

2.1. Description of study samples and procedures

2.1.1. Cohort 1

Subjects: The first cohort comprised 50 cognitively normal subjects (mean age, 66.8 years [SD = 4.96]; range 60–75 years), including 25 males and 25 females, enrolled in a study conducted at the Douglas Mental Health University Institute looking at the effects of basal cortisol levels on memory. Informed written consent as well as demographic and clinical data were obtained from every subject. The majority of subjects self-identified as right-handed, with the exception of three left-handed and one ambidextrous subjects. Exclusion criteria for study participation were: 1) severe physical illness or trauma; 2) concomitant drug use including psychotropic drugs, glucocorticoid medication, anticonvulsants, or sedatives and 3) dementia, Parkinson's Disease, or any major Axis I psychiatric disorder.

MRI Acquisition: Subjects were scanned in a 3-T Siemens Magnetom TrioTim (Siemens Healthcare, Germany) at the Montreal Neurological Institute (Montreal, Canada). The structural MRI scan consisted of a standard three-dimension magnetization prepared rapid gradient echo (3D MP-RAGE) sequence acquired with the following parameters: field of view (FOV) = 256 × 256 mm^2, repetition time (TR) = 2200 msec, echo

time (TE) = 2.9 msec, flip angle = 30°, 176 slices (interleaved acquisition), voxel size = 1 mm isotropic.

Memory assessment: Memory was assessed using a previously described cued recall procedure (CRP) (Lupien et al., 1994). This type of associative memory task has been shown to be sensitive to hippocampus function in several lesion and functional MRI studies (Mayes et al., 2007). The task was administered to participants through a laptop computer running the software E-Prime (Psychology Software Tools, Pittsburgh, PA). Participants were presented with associated and non-associated word pairs, which they were asked to read out loud. Word pairs were presented on the computer screen, in random order, for 5 seconds. The encoding phase was followed by an immediate recall task; for this purpose, one word from the previously learnt word pairs was presented and participants had to recall the other word from the pair. 18 words from the previously presented associated word pairs and 18 words from previously presented non-associated word pairs were presented, for a maximum score of 36 on the immediate recall task. Twenty minutes after completion of the immediate recall, participants were presented with the same 36 words from the previously learnt word pairs one at a time. Again, for each word, they were asked to indicate which word it had been presented with during the encoding phase. The delayed recall score thus consisted of maximum score of 36. The delayed recall score was used as an index of memory performance in our analyses.

2.1.2. Cohort 2

Subjects: This sample consisted of 88 cognitively normal aging individuals enrolled in a longitudinal study at the McGill University Research Centre for Studies in Aging (Montreal, QC, Canada). Informed written consent was obtained from every subject. Inclusion criteria for participation in the longitudinal study were: 1) aged between 55 and 80 years; 2) absence of current or past neurological, psychiatric or severe medical conditions; 3) no contra-indication for MRI; 4) no current or past history of substance abuse; 4) no current use of medication known to cross the blood brain barrier or alter cognitive functioning and 5) a score in the normal range on standard depression questionnaires. Only participants who completed a structural MRI scan were included in the analyses. The sample included 45 males and 43 females (mean age, 65.06 years [SD = 6.42]; range 55–80 years). All participants identified as right-handed, as confirmed by the short form of the Edinburgh Handedness Inventory (Veale, 2014).

MRI Acquisition: MRI scans were performed on a 3-T Siemens Magnetom TrioTim scanner (Siemens Healthcare, Germany) at the Douglas Institute Brain Imaging Center (Montreal, Canada). Structural images were collected for each participant using a sagittal 3D MP-RAGE sequence and the following acquisition parameters: FOV = 256 × 256 mm^2, TR = 2300 msec, TE = 2.98 msec, flip angle = 9°, 176 slices (interleaved acquisition), voxel size = 1 mm isotropic.

Memory Assessment: Memory was assessed using the Rey Auditory Verbal Learning Task (Schmidt, 1996). Briefly, this standard task involves the learning of a list of 15 words (List A) presented over five subsequent trials, each followed by an immediate free recall (learning phase). The learning phase is followed by the presentation of an interference list (List B) that participants are immediately asked to freely recall (interference phase). After the interference phase, participants are asked to freely recall words from the main list (List A). Finally, after a delay of 25–30 minutes, participants are again asked to freely recall words from the main list (i.e. delayed recall of List A). The delayed free recall score was used as index of memory performance in our analyses.

2.2. MR image analyses

2.2.1. MRI preprocessing

Prior to manual segmentation, acquired anatomical images were denoised (Coupé et al., 2008, 2010) and corrected for non-uniformity (Sled et al., 1998). Images were subsequently linearly registered to the ICBM 152 normalized brain template (Collins et al., 1994). At this point, a visual quality control was performed to ensure quality of the image preprocessing.

2.2.2. Volumetric assessment of the hippocampus and surrounding cerebrospinal fluid

The hippocampus and perihippocampal CSF volumes were defined using manual segmentation, which was performed by trained experts in neuroanatomy. The segmentation was done employing the open source software DISPLAY which is part of the minc toolkit package developed at the Brain Imaging Centre of the Montreal Neurological Institute (https://bic-mni.github.io/). This software allows for simultaneous viewing of the structure in all three orientations. The segmentation of hippocampus and perihippocampal CSF volume was mainly performed on the coronal plane, although horizontal and sagittal planes were employed when providing complementary information for the identification of boundaries not available from coronal planes. Segmentation time for hippocampus and perihippocampal CSF space depended on the anatomical complexity of the individual subject, but took on average 90 minutes per subject (45 minutes per hemisphere), ranging from 40 to 150 minutes per subject.

The anatomical boundaries for hippocampal volume assessment are derived from our previously published protocol (Pruessner et al., 2000) and are only briefly described here. For the creation of the HVR, the following segmentation guidelines were applied: The most posterior part of the hippocampus was defined as the first appearance of an ovoid mass of gray matter inferomedial to the trigone of the lateral ventricle (TLV). The lateral border of the hippocampus at this point was the TLV, whereas medially, the border of the hippocampus was identified by the white matter located between the hippocampus proper and the quadrogeminal cistern. Superior and medial to the hippocampus proper, the gray matter spaces of Andreas Retzius gyrus, fasciolar gyrus, and crus of the fornix were excluded through visual inspection, or using arbitrary landmarks when ambiguous.

Segmentation of the perihippocampal CSF began with the appearance of the hippocampus. On the lateral side, the hippocampus is typically bordering with the inferior horn of the lateral ventricle. For the hippocampus tail, any CSF space that was located lateral to the hippocampus was thus included in the CSF assessment (Fig. 2). For the superior border, a horizontal line was drawn from the medial superior edge of the hippocampus in both medial and lateral direction and marked the superior border of the perihippocampal CSF space to be included (Fig. 2). On the medial side, a vertical line was drawn superior-inferior from the most medial part of the hippocampus. Any CSF space that was located adjacent to the medial portion of the hippocampus was included in the ventricle assessment (Fig. 2). Since the inferior horn of the lateral ventricle is naturally surrounded by white matter, no further arbitrary border was needed to determine the lateral border of ventricle space.

For segmentation of the hippocampus body (beginning with the separation of the inferior horn of the lateral ventricle from the lateral ventricle in the coronal view), the most visible superolateral layer of white matter was included as hippocampus volume, assuming that it represents the fimbria. If gray matter was found superior to the fimbria, the first row of gray matter was also included, assuming it represents hippocampus gray matter surrounding the fimbria. In contrast, the white matter band at the inferomedial level of the hippocampus body was excluded, assuming it represents white matter that separates the hippocampus proper from the parahippocampal cortex. On the medial side, the superior and inferior borders of the hippocampus body are also white matter areas, facilitating the identification of hippocampus gray matter in this area. All subcomponents of the hippocampus, i.e. the dentate gyrus, located in between the four CA regions in the hippocampal formation, together with the CA regions themselves and part of the subiculum, the parasubiculum, were included. The parasubiculum was divided from the presubiculum by drawing an arbitrary border - a straight line with an angle of approximately 45° from the white matter separating the hippocampus proper from the parahippocampal cortex

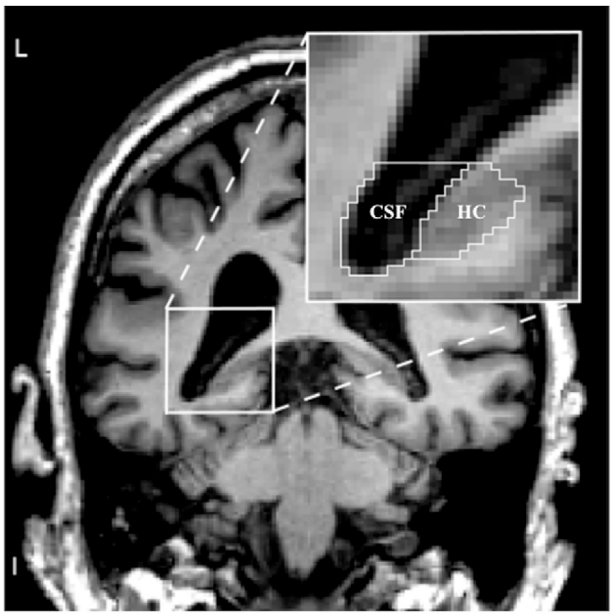

Fig. 2. Illustration of segmentation boundaries for the HVR in the hippocampus tail

Fig. 2. Coronal section of the brain with a focus on the hippocampus. Illustration of boundaries derived from the HVR segmentation protocol for the posterior part of the hippocampus (HC) and adjacent cerebrospinal fluid (CSF).

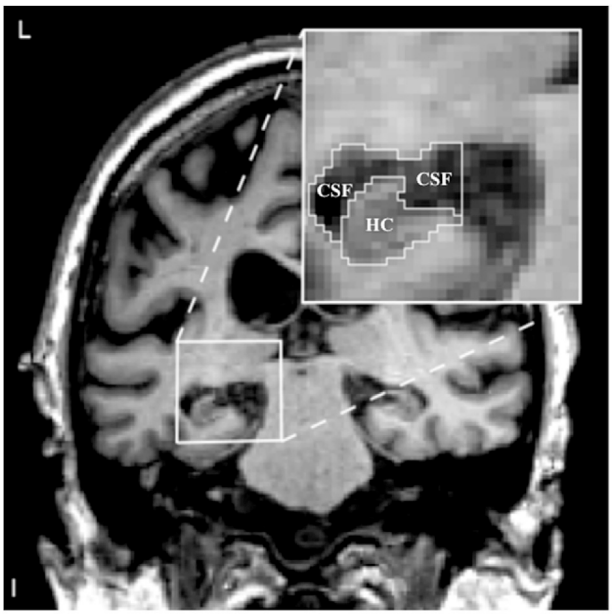

Fig. 3. Illustration of segmentation boundaries for the HVR in the hippocampus body

Fig. 3. Coronal section of the brain with a focus on the hippocampus. Illustration of boundaries derived from the HVR segmentation protocol for hippocampus body (HC) and adjacent cerebrospinal fluid (CSF).

superomedially to the quadrogeminal cistern. The lateral border at this point was identified by the inferior horn of the lateral ventricle. If the inferior horn was invisible due to partial volume effects, the most lateral layer of visible hippocampus gray matter was defined as ventricle mass, and employed as hippocampus border. The quadrogeminal cistern defined the medial, and superomedial border of the hippocampus at this point.

In the hippocampus body on the lateral side, all CSF space that was visible laterally adjacent to the hippocampus was included in the perihippocampal CSF segmentation, with the most lateral layer of hippocampus gray being defined as CSF space if none was otherwise visible, in line with the original protocol (Pruessner et al., 2000, Fig. 3). On the medial side, an arbitrary border was employed by drawing a vertical line in superior-inferior extent from the most medial aspect of the hippocampus (Fig. 3). All CSF space lateral to the vertical line, appearing either inferior or superior of the hippocampus proper, was then included in the segmentation. This way, a distinction between perihippocampal CSF space secondary to hippocampal atrophy versus CSF space of the quadrogeminal cistern was attempted.

The appearance of the hippocampus head was defined by the emergence of the gyrus intralimbicus (appearance of gray matter as a protuberance in the superomedial region of the hippocampus, replacing the CSF space of the quadrogeminal cistern). The most important structures for identification of lateral, anterior and superior borders of the hippocampus head in this region were the inferior horn, the uncal recess of the inferior horn, and the alveus. Besides the coronal view, the sagittal and horizontal views were employed for identification of the anterior and superior border of the hippocampus. Also, the uncal cleft could often serve as marker of the inferior border of the hippocampus.

In the hippocampus head, all CSF space that appeared laterally, together with any appearing CSF space directly adjacent superiorly or inferiorly to the hippocampus, was included (Fig. 4). On the medial side, the vertical line as arbitrary border of hippocampus CSF space was removed, and replaced with two horizontal lines. For the superior border of the hippocampus, a horizontal line was placed on top of the uncal recess of the inferior horn of the lateral ventricle, or the inferior border of the AG, or the superior border of the hippocampus proper, depending on

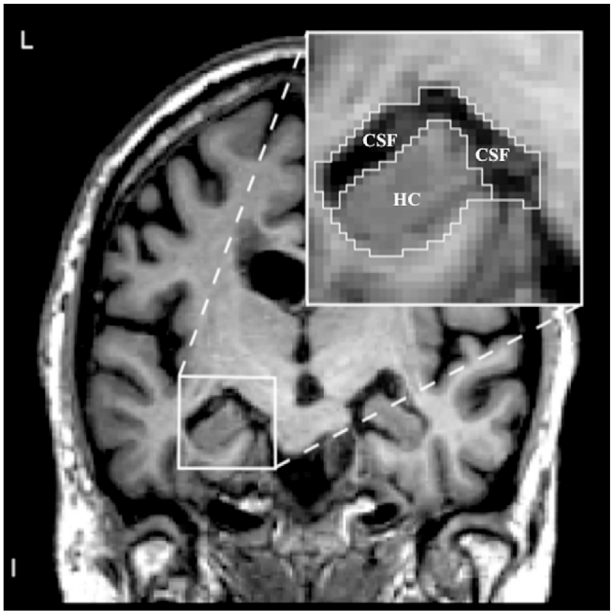

Fig. 4. Illustration of segmentation boundaries for the HVR in the hippocampus head

Fig. 4. Coronal section of the brain with a focus on the hippocampus. Illustration of boundaries derived from the HVR segmentation protocol for hippocampus head (HC) and adjacent cerebrospinal fluid (CSF).

what was visible. For the inferior border of the hippocampus, a horizontal line was either placed at the inferior border of the hippocampus directly, or the inferior horn, or the subiculum, again depending on visibility. Medially, all CSF space directly adjacent to hippocampus gray matter between the two lines was then included in the perihippocampal CSF segmentation (Fig. 4).

Finally, within the hippocampus, if CSF space appeared continuously in the area of the hippocampal fissure for at least three consecutive slices

with at least two voxels each, it was included as CSF space as well.

2.2.3. Calculation of hippocampal volumes and HVR

A voxel count from manually segmented labels was performed to estimate the total perihippocampal CSF (CSFvol) and hippocampus (HCvol) volume. To account for variations in head size, all calculations were performed in the standard stereotaxic space, using the MNI ICBM 152 template (Collins et al., 1994). To compute the HVR, we were guided by the assumption that the ventricle space directly surrounding the hippocampus increases as a function of atrophy or neurodegeneration of the hippocampus proper. Calculating a ratio combining a structural estimation of the target structure together with the ventricle space surrounding the target structure could provide an *integrity index,* which will indicate the preservation of the given structure. The following formula was derived for the computation of the HVR:

$$HVR = \frac{HCvol}{(HCvol + CSFvol)} \quad (1)$$

HVR = hippocampal-to-ventricle ratio, HCvol = Volume of the hippocampus, CSFvol = Volume of perihippocampal CSF.

In this formula, HCvol is divided by the sum of HCvol plus its surrounding CSF (i.e. the sum of temporal horn CSF, uncal recess of the temporal horn, hippocampal fissure, and portions of the quadrigeminal cistern) that are assumed to have expanded as a consequence of hippocampal atrophy or loss of integrity. The resulting value can be interpreted by itself. This can be exemplified by considering very small versus very large ventricle space surrounding the target structure – in the case of very small ventricles (close to '0'), the ratio will be close to '1' – as the target structure then gets divided by itself. This can be imagined as the ideal case in which almost no hippocampus volume decline or ventricle enlargement has taken place. In the case of very large ventricles, the HVR will be considerably smaller than 1, as the ventricular enlargement now will have a stronger impact on the final index. In case of a value of 0.5, it can be estimated that as much as half of the presumed original hippocampus volume has been lost. Thus, the closer the number is to 0, the stronger the (hypothesized) atrophy.

2.3. Statistical analyses

To evaluate the reliability of the established segmentation guidelines, we computed both inter- and intra-rater reliability. To assess intra-rater reliability (test-retest), one of the raters segmented the MR images from the same 14 subjects twice. To assess inter-rater reliability, three independent raters segmented the MR images from the same 20 subjects. Agreement within (intra-rater) and between raters (inter-rater) was estimated using Intraclass Correlation Coefficients (ICC), using a two-way random model (Bartko, 1966). The ICCs were calculated separately for the hippocampus (left/right hippocampus) and perihippocampal CSF space (left/right CSF).

For each cohort, the average HCvol, CSFvol and HVR were computed. A ratio of CSFvol on HCvol (CSFvol/HCvol x 100%), representing the volume occupied by CSFvol relative to HCvol, was also calculated. Associations between age, delayed recall scores and either the CSFvol, HCvol or the HVR were explored in both cohorts using simple Pearson correlations. Correlations were computed for the left hemisphere, right hemisphere and for both hemispheres combined (i.e. total volume). To test our hypothesis that the HVR is a superior index of hippocampal integrity than absolute volume, we examined differences in the magnitude of correlations obtained with either the HCvol or the HVR. Differences in obtained correlations were assessed using the Steiger's *Z* test, which allows the comparison of overlapping correlations (Steiger, 1980). To limit the number of comparisons, and because of the aim of this study, we only contrasted correlation coefficients obtained with the total HCVol or total HVR.

Finally, the association between the total HCvol or total HVR and memory performance was further investigated in two separate linear regression analyses. Because of their potential effects on memory performance, age, education and sex/gender were entered in the model as covariates. All statistical analyses were performed using SPSS 20 (SPSS, Chicago, IL).

3. Results

The ICCs for intra-rater and inter-rater reliability are presented in Table 1. Overall, the inter- and intra-rater reliability was high for both the hippocampus and perihippocampal CSF segmentation. The reliability coefficients obtained for the hippocampus volume were consistent with those reported as part of the original segmentation protocol (Pruessner et al., 2000).

3.1. Cohort 1

The average values of HCvol, CSFvol, CSF/HC %, and HVR for Cohort 1 are presented in Table 2. The HVR showed a strong negative correlation with the total CSFvol ($r = -0.91$, $p < .01$) and a positive correlation with the total HCvol ($r = 0.78$, $p < .01$). The results of correlations with age and memory performance for Cohort 1 are presented in Table 3. Correlations between age and the left, right or total HVR were all significant ($p < .05$). In contrast, none of the correlations between age and CSFvol or HCvol reached statistical significance. Correlation with age was significantly superior when computed with the total HVR than the total HCvol (Steiger's $Z = 1.98$, $p < .05$). Correlations with memory performance were significant for the left, right and total HVR ($p < .01$). Correlation with memory scores were also significant for the left, right and total CSFvol ($p < .01$). While a significant correlation was found between memory performance and the right HCvol ($p < .05$), correlations with either the left or total HCvol did not reach statistical significance. Subsequent analysis showed that the correlation between memory performance and the total HVR was marginally superior to the correlation computed with the total HCvol (Steiger's $Z = 1.7$, $p < .10$). The results of linear regression analyses evaluating the value of either the HCvol or HVR in predicting memory performance in Cohort 1 are summarized in Table 4. The model including the total HCvol explained 32% ($p < .01$) of the variance in memory scores, with education being the only significant predictor of memory performance. When computing an equivalent model with the total HVR, both education and the total HVR significantly contributed to the model and the proportion of explained variance in memory scores was of 40% ($p < .01$).

3.2. Cohort 2

The average values of HCvol, CSFvol, CSF/HC %, and HVR for Cohort 2 are presented in Table 2. The HVR was significantly and negatively correlated with the total CSFvol ($r = -0.95$, $p < .01$) and positively correlated with the HCvol ($r = 0.68$, $p < .01$). The results of correlations with age and memory performance for Cohort 2 are presented in Table 3. Correlations between age and the left, right or total HVR were all significant ($p < .01$). Correlations between age and the left, right or total HCvol ($p < .05$) and CSFvol ($p < .01$) were also all significant. The

Table 1
Results of reliability analyses.

	Intra-rater reliability		Inter-rater reliability	
	Left	*Right*	*Left*	*Right*
HC	.92	90	.84	.86
CSF	.91	.92	.88	.88

Intraclass Correlation coefficients (ICC) quantifying intra- and inter-rater reliability for the manual segmentation of the hippocampus (HC) and perihippocampal CSF (CFS) volumes necessary to compute the HVR, for the left and right hemisphere.

Table 2
Summary of volumetric measures.

	HCvol *Mean (SD)*		CSFvol *Mean (SD)*		CSF/HC % *Mean (SD)*		HVR *Mean (SD)*	
	Left	*Right*	*Left*	*Right*	*Left*	*Right*	*Left*	*Right*
Cohort 1	4913.0	4993.2	1909.0	1921.1	40.4	39.9	0.72	0.73
	(618.0)	(567.9)	(819.8)	(771.2)	(20.6)	(19.6)	(0.09)	(0.09)
Cohort 2	4768.7	4819.0	2378.3	2213.9	53.1	49.5	0.67	0.68
	(784.4)	(750.7)	(657.7)	(607.7)	(25.4)	(22.6)	(0.09)	(0.08)

Average values for the hippocampus volume (HCvol), the perihippocampal CSF volume (CSFvol), the CSFvol/HCvol ratio, and the HVR across the two studied cohorts. Volumetric measures (i.e. HCvol, CSFvol) are presented in cubic millimeters (mm^3).

Table 3
Summary of correlation analyses.

	Total volume			Right hemisphere			Left hemisphere		
	CSFvol	*HCvol*	*HRV*	*CSFvol*	*HCvol*	*HRV*	*CSFvol*	*HCvol*	*HRV*
Cohort 1									
Age	.27†	-.07	-.31*	.24	-.04	-.28*	.28†	-.09	-.33*
Delayed recall (CRP)	-.40**	.30†	.49**	-.42**	.35*	.51**	-.38**	.23	.46**
Cohort 2									
Age	.38**	-.27*	-.41**	.33**	-.25*	-.38**	.39**	-.28**	-.42**
Delayed recall (RAVLT)	-.46**	.46**	.54**	-.42**	.44**	.51**	-.47**	.47**	.54**

Pearson correlations between the left hemisphere, right hemisphere and combined volume of the perihippocampal CSF (CSFvol), hippocampus (HCvol) or hippocampal-ventricule ratio (HVR), together with age and memory performance across the two studied cohorts (Cohort 1 and Cohort 2). **p < 0.01, *p < 0.05, † Marginal significance (p < .10).

Table 4
Predictors of memory performance across both study samples.

	Cohort 1		Cohort 2	
Model 1	R^2 = .32 (p<.01)		R^2 = .28 (p<.001)	
	Beta	*t (sig)*	*Beta*	*t (sig)*
Total HCvol	.133	0.80 (ns)	**.390**	**3.89 (p<.001)**
Age	−.208	−1.48 (ns)	**−.209**	**−2.15 (p<.05)**
Education	**.332**	**2.37 (p<.05)**	.125	1.32 (ns)
Sex/Gender	.270	1.59 (ns)	.165	1.70 (ns)
Model 2	**R^2 = .40 (p<.01)**		**R^2 = .32 (p<.001)**	
	Beta	*t (sig)*	*Beta*	*t (sig)*
Total HVR	**.417**	**2.33 (p<.05)**	**.479**	**4.54 (p<.001)**
Age	−.044	−0.29 (ns)	−.118	1.17 (ns)
Education	**.375**	**2.82 (p<.01)**	.130	1.40 (ns)
Sex/Gender	.085	0.49 (ns)	.117	1.21 (ns)

Summary of linear regression models evaluating how the combined left and right standardized hippocampal volume (Total HCvol - Model 1) or hippocampal-ventricule ratio (Total HVR - Model 2), age, education and sex/gender predict memory performance across both study cohorts (Cohort 1 and Cohort 2). ns = not significant (p > .05).

Steiger's Z test showed that correlations between age and the total HVR were significantly superior than correlations obtained with the total HCvol (Steiger's Z = 2.05, p < .05). Correlations with memory scores were significant for all HVR, CSFvol and HCvol measures (p < .01). Although the correlation between memory performance and total HVR was numerically superior, it did not significantly differ from the correlation obtained with the total HCvol (Steiger's Z = 1.30, p > .05). The results of linear regression analyses evaluating the value of either the HCvol or the HVR in predicting memory performance in Cohort 2 are summarized in Table 4. The model including total HCvol explained 28% (p < .001) of the variance in memory scores, and both the total HCvol (p < .001) and age (p < .05) significantly contributing to the overall model fit. When an equivalent model was computed with the total HVR, the proportion of explained variance in memory scores was 32% (p < .001) and the total HVR was found to be the only significant predictor of memory performance.

4. Discussion

In this article, we introduced the manual segmentation protocol for the HVR, a novel index to estimate the structural integrity of the hippocampus. By combining a measure of hippocampal volume and adjacent perihippocampal ventricular enlargement, we hypothesized that the HVR would provide a more sensitive estimation of hippocampal structural integrity, particularly when relying on cross-sectional data.

We demonstrated that the manual segmentation protocol for the HVR can be implemented with high reliability, with intra-rater ICCs ranging from 0.90 to 0.92 and inter-rater ICCs ranging from 0.84 to 0.88. In two separate cohorts of healthy aging individuals, we further evaluated the relevance of this index via correlation and regression analyses.

The HVR showed significantly stronger negative associations with age than the HCvol across the two cohorts. In one of the two studied samples (Cohort 1), we even failed to find a significant association between age and HCvol, while this correlation strongly increased and reached significance when using the HVR. The discrepancy in correlations between HCvol and age obtained across the two cohorts is possibly due to difference in demographics. The smaller sample size, more restricted age range, and younger age of participants included in Cohort 1 possibly leads to less variation and thus less power to reveal significant correlations. Nonetheless, the lack of association between hippocampus volume and age has been previously described in cross-sectional studies investigating non-pathological populations, although inconsistently. While some studies found a global preservation in hippocampal volume with advancing age (Fjell et al., 2009; Lim et al., 1990; Laakso et al., 1995; Sullivan et al., 1995; Good et al., 2001) others report significant volume reduction (Coffey et al., 1992; Jack et al., 1992, 1997; Walhovd et al., 2011). On the other hand, longitudinal studies in normal aging individuals have consistently observed a reduction of hippocampal volume over time, with an atrophy rate ranging from 0.3 to 2.2% per year (for reviews, see: Fox and Schott, 2004; Frisoni et al., 2010). Evidence from longitudinal studies thus strongly suggests that hippocampus volume loss does occur as a consequence of normal aging. It is has been argued that cross-sectional studies using absolute volume measurements likely underestimate age-related changes (Raz et al., 2005; Fjell et al., 2014). Using the HVR, our results align with those of longitudinal studies and confirm a negative association between age and hippocampal structural integrity.

When examining associations with delayed recall memory performance, correlations obtained with the HVR were numerically superior to correlations obtained with the HCvol or CSFvol across both cohorts, although these differences did not reach statistical significance. In linear regression models, the HVR contributed to the prediction of memory performance across the two study samples. On the other hand, the HCvol was found to be a significant predictor of memory performance in only one of the two cohorts (Cohort 2). Again, these discrepancies in results across the two studied samples could be explained by differences in group demographics. However, inconsistencies in the relationship between hippocampus volume and memory are frequently found in the literature. As highlighted in a literature review by Van Petten (2004), there is an important variability in results of studies assessing relationships between hippocampal size and memory in non-pathological aging, with some studies reporting significant positive associations (Reiman et al., 1998; Convit et al., 2003; Rosen et al., 2003) and others reporting null (de Toledo-Morrell et al., 2000; Laakso et al., 2000; Petersen et al., 2000) or even negative associations (Sullivan et al., 1995; Köhler et al., 1998; Chantôme et al., 1999). Overall, and due to the important heterogeneity in results across studies, the evidence for an association between hippocampal volume and memory function in normal aging is weak (Van Petten, 2004). Refining volumetric measures of the hippocampus could allow improving consistency in findings across studies.

A key factor likely contributing to the heterogeneity in results across studies consists in the large variation in hippocampal volumes in the general population (Lupien et al., 2007). In other words, a small hippocampal volume could result from developmental predispositions rather than underlying pathological processes or atrophy. The important variation in hippocampal volumes in the general population hinders the assessment of hippocampal integrity from cross-sectional volumetric MRI data. By incorporating a measure of adjacent ventricular space to standard volumetric measure of the hippocampus, we believe that the HVR allows better distinguishing volumetric changes from developmental variations in hippocampal volume. Via stronger associations with age and memory scores, our preliminary results support this hypothesis and indicate that the HVR might be a more sensitive marker of changes in hippocampus integrity. Overall, our results show that the examined structural markers vary in the strength of their associations with age or memory scores, with the hippocampus showing the weakest correlations, followed by correlations obtained with perihippocampal CSF volume, and the HVR showing the strongest correlations. As such, taken in isolation, standard hippocampal volume does not represent the most informative volumetric marker for age-related structural and functional changes. Measures of ventricular enlargement appear to be of significant value in that regards, particularly when combined to additional volumetric assessment. In both studied cohorts, perihippocampal CSF was highly related to the HVR and seemed to drive the improvement in correlations with age or memory scores obtained with the HVR. These results are in agreement with those from Wang et al. (2002) and Bartos et al. (2019) demonstrating an improved discrimination between Alzheimer's Disease patients and age-matched controls when combining rates of cortical atrophy and ventricular enlargement. For instance, Bartos et al. (2019) investigated the ability of various ratios of volumetric measures obtained with FreeSurfer (v. 6.0) to differentiate Alzheimer's Disease patients from healthy aging individuals. They found that the ratio providing the best discrimination between groups was the hippocampus-to-inferior lateral ventricle ratio. While direct comparison with the HVR cannot be validly performed due to differences in group demographics and in the definition of anatomical boundaries, the hippocampus-to-inferior lateral ventricle ratios (left: 0.77 ± 0.15; right: 0.78 ± 0.18) reported by Bartos et al. (2019) in healthy aging individuals are globally consistent with the HVR obtained in the present manuscript (Cohort 1 - left: 0.72 ± 0.09; right: 0.73 ± 0.09, Cohort 2 - left: 0.67 ± 0.09; right: 0.68 ± 0.08). This consistency in findings across studies provides additional support for the validity of such ratio. Our results also align with those from a previous study computing a hippocampal volumetric integrity (HVI) index using the fraction of brain matter (non-CSF) encompassing the region expected to represent the hippocampus, based on a standardized probabilistic atlas derived from control subjects (Ardekani et al., 2016, 2017). The authors showed that the HVI efficiently differentiates Alzheimer's Disease patients from normal controls (2016) and predicts conversion from MCI to Alzheimer's Disease with high accuracy (2017).

Although these preliminary results are promising, the HVR requires further validation. Future studies should aim to replicate the present findings in different samples and populations. For example, future work could evaluate whether the HVR allows a better discrimination between pathological and non-pathological populations (e.g. Alzheimer's Disease or MCI patients versus normal aging). The ultimate proof for the relevance of the HVR would come from longitudinal data in cognitively normal subjects who subsequently develop cognitive impairments due to neurodegeneration – this data would allow establishing whether the HVR is more sensitive than previous MRI structural markers in predicting cognitive decline over time. Further validation studies on larger and more inclusive samples of healthy/non-pathological individuals should also be conducted with the aim of establishing normative data for the HVR. Such normative data would provide valuable information regarding normal variations in HVR. The availability of normative data for the HVR would allow detecting abnormal levels of hippocampal atrophy at the subject-level and would add to the potential clinical utility of this measure. Because the HVR heavily rely on the assumption that when hippocampal volume is at its peak, surrounding CSF space is at its minimum, the examination of the HVR in a developmental population would allow corroborating the association between perihippocampal CSF volume and normal and pathological aging processes. To conclude, while the manual segmentation of the hippocampus is labor intensive, the additional segmentation of the adjacent CSF space is relatively simple and rapid. If future studies support validity and relevance of this index, the anatomical segmentation protocol for the HVR could become semi- or fully automated and made available as part of computerized pipelines (e.g. MAGET; Chakravarty et al., 2013). This would facilitate its application and use in large datasets and across laboratories.

5. Conclusion

In this article, we describe and present the segmentation protocol for the HVR, a novel index aiming to improve the estimation hippocampal integrity from cross-sectional data. We also provide preliminary evidence for the validity of this index. The strong increase in association with age and memory scores suggests that this marker is relevant to the assessment of age-related changes in hippocampal volume. Although the HVR requires further validation, these preliminary results indicate that this index could become a useful marker of hippocampal integrity and be particularly valuable to studies investigating the clinical or functional significance of hippocampal integrity using cross-sectional MRI data. A refined assessment of hippocampal integrity is central to our understanding of how this region is affected by normal and pathological aging processes as well as the functional repercussions of such changes.

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References

Apostolova, L.G., Alves, G., Hwang, K.S., Babakchanian, S., Bronnick, K.S., Larsen, J.P., Thompson, P.M., Chou, Y.-Y., Tysnes, O.B., Vefring, H.K., 2012a. Hippocampal and ventricular changes in Parkinson's disease mild cognitive impairment. Neurobiol. Aging 33, 2113–2124.

Apostolova, L.G., Babakchanian, S., Hwang, K.S., Green, A.E., Zlatev, D., Chou, Y.-Y., DeCarli, C., Jack Jr., C.R., Petersen, R.C., Aisen, P.S., 2013. Ventricular atrophy and its clinical correlates in the imaging cohort from the ADCS MCI Donepezil/Vitamin E study. Alzheimers Dis. Assoc. Disord. 27, 174–181.

Apostolova, L.G., Green, A.E., Babakchanian, S., Hwang, K.S., Chou, Y.-Y., Toga, A.W., Thompson, P.M., 2012b. Hippocampal atrophy and ventricular enlargement in normal aging, mild cognitive impairment and Alzheimer's disease. Alzheimers Dis. Assoc. Disord. 26, 17–27.

Ardekani, B.A., Bermudez, E., Mubeen, A.M., Bachman, A.H., 2017. Prediction of incipient Alzheimer's disease dementia in patients with mild cognitive impairment. J. Alzheimer's Dis. 55, 269–281.

Ardekani, B.A., Convit, A., Bachman, A.H., 2016. Analysis of the MIRIAD data shows sex differences in hippocampal atrophy progression. J. Alzheimer's Dis. 50, 847–857.

Barber, R., McKeith, I., Ballard, C., Gholkar, A., O'brien, J., 2001. A comparison of medial and lateral temporal lobe atrophy in dementia with Lewy bodies and Alzheimer's disease: magnetic resonance imaging volumetric study. Dement. Geriatr. Cognit. Disord. 12, 198–205.

Bartko, J.J., 1966. The intraclass correlation coefficient as a measure of reliability. Psychol. Rep. 19, 3–11.

Bartos, A., Gregus, D., Ibrahim, I., Tintěra, J., 2019. Brain volumes and their ratios in Alzheimer' s disease on magnetic resonance imaging segmented using Freesurfer 6.0. Psychiatry Res. Neuroimaging (in press).

Bilgel, M., Jedynak, B.M., Alzheimer's Disease Neuroimaging Initiative, 2019. Predicting time to dementia using a quantitative template of disease progression. Alzheimer's Dementia: Diagn. Assess. Dis. Monit. 11, 205–215.

Chakravarty, M.M., Steadman, P., van Eede, M.C., Calcott, R.D., Gu, V., Shaw, P., Raznahan, A., Collins, D.L., Lerch, J.P., 2013. Performing label-fusion-based segmentation using multiple automatically generated templates. Hum. Brain Mapp. 34, 2635–2654.

Chantôme, M., Perruchet, P., Hasboun, D., Dormont, D., Sahel, M., Sourour, N., Zouaoui, A., Marsault, C., Duyme, M., 1999. Is there a negative correlation between explicit memory and hippocampal volume? Neuroimage 10, 589–595.

Chou, Y.-Y., Leporé, N., Avedissian, C., Madsen, S.K., Parikshak, N., Hua, X., Shaw, L.M., Trojanowski, J.Q., Weiner, M.W., Toga, A.W., 2009. Mapping correlations between ventricular expansion and CSF amyloid and tau biomarkers in 240 subjects with Alzheimer's disease, mild cognitive impairment and elderly controls. Neuroimage 46, 394–410.

Coffey, C., Wilkinson, W., Parashos, L., Soady, S., Sullivan, R., Patterson, L., Figiel, G., Webb, M., Spritzer, C., Djang, W., 1992. Quantitative cerebral anatomy of the aging human brain A cross-sectional study using magnetic resonance imaging. Neurology 42, 527-527.

Collins, D.L., Neelin, P., Peters, T.M., Evans, A.C., 1994. Automatic 3D intersubject registration of MR volumetric data in standardized Talairach space. J. Comput. Assist. Tomogr. 18, 192–205.

Convit, A., Wolf, O.T., Tarshish, C., De Leon, M.J., 2003. Reduced glucose tolerance is associated with poor memory performance and hippocampal atrophy among normal elderly. Proc. Natl. Acad. Sci. 100, 2019–2022.

Coupé, P., Manjón, J.V., Fonov, V., Pruessner, J., Robles, M., Collins, D.L., 2010. Nonlocal patch-based label fusion for Hippocampus segmentation. Med. Image Comput. Comput. Assist. Interv. 13, 129–136.

Coupé, P., Yger, P., Prima, S., Hellier, P., Kervrann, C., Barillot, C., 2008. An optimized blockwise nonlocal means denoising filter for 3-D magnetic resonance images. Medical Imaging. IEEE Trans. 27, 425–441.

Dalaker, T.O., Zivadinov, R., Ramasamy, D.P., Beyer, M.K., Alves, G., Bronnick, K.S., Tysnes, O.B., Aarsland, D., Larsen, J.P., 2011. Ventricular enlargement and mild cognitive impairment in early Parkinson's disease. Mov. Disord. 26, 297–301.

De Leon, M., Golomb, J., George, A., Convit, A., Tarshish, C., McRae, T., De Santi, S., Smith, G., Ferris, S., Noz, M., 1993. The radiologic prediction of Alzheimer disease: the atrophic hippocampal formation. Am. J. Neuroradiol. 14, 897–906.

de Toledo-Morrell, L., Dickerson, B., Sullivan, M., Spanovic, C., Wilson, R., Bennett, D., 2000. Hemispheric differences in hippocampal volume predict verbal and spatial memory performance in patients with Alzheimer's disease. Hippocampus 10, 136–142.

Eichenbaum, H., 2004. Hippocampus: cognitive processes and neural representations that underlie declarative memory. Neuron 44, 109–120.

Estévez-Santé, S., Jiménez-Huete, A., 2019. Comparative analysis of methods of volume adjustment in hippocampal volumetry for the diagnosis of Alzheimer disease. J. Neuroradiol. (in press).

Fjell, A.M., McEvoy, L., Holland, D., Dale, A.M., Walhovd, K.B., Initiative AsDN, 2014. What is normal in normal aging? Effects of aging, amyloid and Alzheimer's disease on the cerebral cortex and the hippocampus. Prog. Neurobiol. 117, 20–40.

Fjell, A.M., et al., 2009. One-year brain atrophy evident in healthy aging. J. Neurosci. 29 (48), 15223–15231.

Fox, N.C., Schott, J.M., 2004. Imaging cerebral atrophy: normal ageing to Alzheimer's disease. The Lancet 363, 392–394.

Frisoni, G.B., Fox, N.C., Jack Jr., C.R., Scheltens, P., Thompson, P.M., 2010. The clinical use of structural MRI in Alzheimer disease. Nat. Rev. Neurol. 6, 67–77.

Good, C.D., Johnsrude, I.S., Ashburner, J., Henson, R.N.A., Friston, K.J., Frackowiak, R.S.J., 2001. A voxel-based morphometric study of ageing in 465 normal adult human brains. Neuroimage 14, 21–36.

Gould, E., Tanapat, P., 1999. Stress and hippocampal neurogenesis. Biol. Psychiatry 46, 1472–1479.

Jack, C., Shiung, M., Gunter, J., O'brien, P., Weigand, S., Knopman, D., Boeve, B., Ivnik, R., Smith, G., Cha, R., 2004. Comparison of different MRI brain atrophy rate measures with clinical disease progression in AD. Neurology 62, 591–600.

Jack, C.R., Petersen, R., Xu, Y., O'brien, P., Smith, G., Ivnik, R., Boeve, B., Tangalos, E., Kokmen, E., 2000. Rates of hippocampal atrophy correlate with change in clinical status in aging and AD. Neurology 55, 484–490.

Jack, C.R., Petersen, R.C., O'brien, P.C., Tangalos, E.G., 1992. MR-based hippocampal volumetry in the diagnosis of Alzheimer's disease. Neurology 42, 183-183.

Jack, C.R., Petersen, R.C., Xu, Y.C., Waring, S.C., O'brien, P.C., Tangalos, E.G., Smith, G.E., Ivnik, R.J., Kokmen, E., 1997. Medial temporal atrophy on MRI in normal aging and very mild Alzheimer's disease. Neurology 49, 786–794.

Kempermann, G., Kuhn, H.G., Gage, F.H., 1997. More hippocampal neurons in adult mice living in an enriched environment. Nature 386, 493.

Kempermann, G., Kuhn, H.G., Gage, F.H., 1998. Experience-induced neurogenesis in the senescent dentate gyrus. J. Neurosci. 18, 3206–3212.

Khlif, M.S., Egorova, N., Werden, E., Redolfi, A., Boccardi, M., DeCarli, C.S., Brodtmann, A., 2019. A comparison of automated segmentation and manual tracing in estimating hippocampal volume in ischemic stroke and healthy control participants. Neuroimage: Clin. 21, 101581.

Köhler, S., Black, S., Sinden, M., Szekely, C., Kidron, D., Parker, J., Foster, J., Moscovitch, M., Wincour, G., Szalai, J., 1998. Memory impairments associated with hippocampal versus parahippocampal-gyrus atrophy: an MR volumetry study in Alzheimer's disease. Neuropsychologia 36, 901–914.

Laakso, M.P., Hallikainen, M., Hänninen, T., Partanen, K., Soininen, H., 2000. Diagnosis of Alzheimer's disease: MRI of the hippocampus vs delayed recall. Neuropsychologia 38, 579–584.

Laakso, M.P., Soininen, H., Partanen, K., Helkala, E.L., Hartikainen, P., Vainio, P., Hallikainen, M., Hänninen, T., Riekkinen Sr., P.J., 1995. Volumes of hippocampus, amygdala and frontal lobes in the MRI-based diagnosis of early Alzheimer's disease: correlation with memory functions. J. Neural Transm. - Parkinson's Dis. Dementia Sect. (JNT) 9, 73–86.

Lim, K.O., Zipursky, R.B., Murphy, G.M., Pfefferbaum, A., 1990. In vivo quantification of the limbic system using MRI: effects of normal aging. Psychiatry Res. Neuroimaging 35, 15–26.

Lupien, S., Lecours, A.R., Lussier, I., Schwartz, G., Nair, N., Meaney, M.J., 1994. Basal cortisol levels and cognitive deficits in human aging. J. Neurosci. 14, 2893–2903.

Lupien, S.J., Evans, A., Lord, C., Miles, J., Pruessner, M., Pike, B., Pruessner, J.C., 2007. Hippocampal volume is as variable in young as in older adults: implications for the notion of hippocampal atrophy in humans. Neuroimage 34, 479–485.

Mak, E., Su, L., Williams, G.B., Firbank, M.J., Lawson, R.A., Yarnall, A.J., Duncan, G.W., Mollenhauer, B., Owen, A.M., Khoo, T.K., Brooks, D.J., Rowe, J.B., Barker, R.A., Burn, D.J., O'Brien, J.T., 2017. Longitudinal whole-brain atrophy and ventricular enlargement in nondemented Parkinson's disease. Neurobiol. Aging 55, 78–90.

Mayes, A., Montaldi, D., Migo, E., 2007. Associative memory and the medial temporal lobes. Trends Cogn. Sci. 11, 126–135.

Mirescu, C., Peters, J.D., Gould, E., 2004. Early life experience alters response of adult neurogenesis to stress. Nat. Neurosci. 7, 841–846.

Mizuno, K., Wakai, M., Takeda, A., Sobue, G., 2000. Medial temporal atrophy and memory impairment in early stage of Alzheimer's disease: an MRI volumetric and memory assessment study. J. Neurol. Sci. 173, 18–24.

Mlynarik, M., Johansson, B.B., Jezova, D., 2004. Enriched environment influences adrenocortical response to immune challenge and glutamate receptor gene expression in rat hippocampus. Ann. N. Y. Acad. Sci. 1018, 273–280.

Mungas, D., Reed, B., Jagust, W., DeCarli, C., Mack, W., Kramer, J., Weiner, M., Schuff, N., Chui, H., 2002. Volumetric MRI predicts rate of cognitive decline related to AD and cerebrovascular disease. Neurology 59, 867–873.

Petersen, R.C., Jack, C., Xu, Y.-C., Waring, S., O'brien, P., Smith, G., Ivnik, R., Tangalos, E., Boeve, B.F., Kokmen, E., 2000. Memory and MRI-based hippocampal volumes in aging and AD. Neurology 54, 581.

Pruessner, J.C., Li, L.M., Serles, W., Pruessner, M., Collins, D.L., Kabani, N., Lupien, S., Evans, A.C., 2000. Volumetry of hippocampus and amygdala with high-resolution MRI and three-dimensional analysis software: minimizing the discrepancies between laboratories. Cerebr. Cortex 10, 433–442.

Pruessner, J.C., Baldwin, M.W., Dedovic, K., Renwick, R., Mahani, N.K., Lord, C., Meaney, M., Lupien, S., 2005. Self-esteem, locus of control, hippocampal volume, and cortisol regulation in young and old adulthood. Neuroimage 28, 815–826.

Raz, N., Lindenberger, U., Rodrigue, K.M., Kennedy, K.M., Head, D., Williamson, A., Dahle, C., Gerstorf, D., Acker, J.D., 2005. Regional brain changes in aging healthy adults: general trends, individual differences and modifiers. Cerebr. Cortex 15, 1676–1689.

Reiman, E.M., Uecker, A., Caselli, R.J., Lewis, S., Bandy, D., De Leon, M.J., De Santi, S., Convit, A., Osborne, D., Weaver, A., 1998. Hippocampal volumes in cognitively normal persons at genetic risk for Alzheimer's disease. Ann. Neurol.: Off. J. Am. Neurol. Assoc. Child Neurol. Soc. 44, 288–291.

Rosen, A.C., Prull, M.W., Gabrieli, J.D., Stoub, T., O'hara, R., Friedman, L., Yesavage, J.A., deToledo-Morrell, L., 2003. Differential associations between entorhinal and hippocampal volumes and memory performance in older adults. Behav. Neurosci. 117, 1150–1160.

Schmidt, M., 1996. Rey Auditory Verbal Learning Test: A Handbook. Western Psychological Services, Los Angeles, CA.

Seif, M., Ziegler, G., Freund, P., 2018. Progressive ventricles enlargement and cerebrospinal fluid volume increases as a marker of neurodegeneration in patients with spinal cord injury: a longitudinal magnetic resonance imaging study. J. Neurotrauma 35, 2941–2946.

Sled, J.G., Zijdenbos, A.P., Evans, A.C., 1998. A nonparametric method for automatic correction of intensity nonuniformity in MRI data. Medical Imaging. IEEE Trans. 17, 87–97.

Steiger, J.H., 1980. Tests for comparing elements of a correlation matrix. Psychol. Bull. 87, 245.

Sullivan, E.V., Marsh, L., Mathalon, D.H., Lim, K.O., Pfefferbaum, A., 1995. Age-related decline in MRI volumes of temporal lobe gray matter but not hippocampus. Neurobiol. Aging 16, 591–606.

Sullivan, E.V., Pfefferbaum, A., Swan, G.E., Carmelli, D., 2001. Heritability of hippocampal size in elderly twin men: equivalent influence from genes and environment. Hippocampus 11, 754–762.

Thompson, P.M., Hayashi, K.M., de Zubicaray, G.I., Janke, A.L., Rose, S.E., Semple, J., Hong, M.S., Herman, D.H., Gravano, D., Doddrell, D.M., 2004. Mapping hippocampal and ventricular change in Alzheimer disease. Neuroimage 22, 1754–1766.

Van Petten, C., 2004. Relationship between hippocampal volume and memory ability in healthy individuals across the lifespan: review and meta-analysis. Neuropsychologia 42, 1394–1413.

Veale, J.F., 2014. Edinburgh handedness inventory–short form: a revised version based on confirmatory factor analysis. Laterality: asymmetries of Body. Brain Cogn. 19, 164–177.

Walhovd, K.B., Westlye, L.T., Amlien, I., Espeseth, T., Reinvang, I., Raz, N., Agartz, I., Salat, D.H., Greve, D.N., Fischl, B., 2011. Consistent neuroanatomical age-related volume differences across multiple samples. Neurobiol. Aging 32, 916–932.

Wang, D., Chalk, J.B., Rose, S.E., de Zubicaray, G., Cowin, G., Galloway, G.J., Barnes, D., Spooner, D., Doddrell, D.M., Semple, J., 2002. MR image-based measurement of rates of change in volumes of brain structures. Part II: application to a study of Alzheimer's disease and normal aging. Magn. Reson. Imag. 20, 41–48.

Zainuddin, M.S.A., Thuret, S., 2012. Nutrition, adult hippocampal neurogenesis and mental health. Br. Med. Bull. 103, 89–115.