RESEARCH PAPER

Role of MRI in Alzheimer's Disease with Arterial Spin Labelling (ASL)

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Abstract
Background: Arterial spin labelling (ASL) measures cerebral blood flow without radiation or contrast medium and adds a few minutes to a routine brain MRI. Whether it reliably detects the prodromal stage of Alzheimer's disease is disputed.
Objectives: The utility of ASL-MRI in the study of Alzheimer's Disease based on comparison of global and regional cerebral blood flow (CBF) among cognitively normal subjects, MCI patients and AD patients, and the relationship between regional CBF, cognitive dysfunction, hippocampal atrophy and diagnostic discrimination.
Materials and Methods: Seventy participants aged 50 years or more (20 controls, 25 MCI, 25 AD) underwent 3 T structural MRI and pseudo-continuous ASL. Diagnosis was clinical, by National Institute on Aging–Alzheimer's Association and DSM-5 criteria, without reference to imaging. CBF was sampled from atlas-defined grey matter regions of interest. Groups were compared by analysis of variance with Tukey post-hoc testing, and discrimination assessed by receiver operating characteristic analysis.
Results: Global CBF fell from 44.20 in controls to 39.92 in MCI and 36.72 mL/100 g/min in AD ($F = 7.174$, $p = 0.002$). Regional reductions were larger. Mean hippocampal CBF was significantly lower at each stage from 44.50 ± 5.00 to 39.80 ± 5.25 to 30.56 ± 4.81 mL/100 g/min ($F = 45.667$, $p < 0.001$). There was significantly reduced blood perfusion in all posterior cortical regions and medial temporal lobes, but not in the cerebellum ($p = 0.742$). Mean hippocampal CBF was associated with Mini Mental State Examination ($r = 0.689$) and Clinical Dementia Rating scale ($rho = -0.739$). Hippocampal atrophy was significantly correlated with perfusion of the posterior cingulate ($rho = -0.566$) and temporoparietal regions ($rho = -0.558$) rather than with hippocampal perfusion ($rho = -0.478$).
Conclusion: ASL shows graded, regionally selective hypoperfusion across the Alzheimer's disease continuum, detectable at the MCI stage and correlated with both cognition and atrophy. Regional measurements far outperform global CBF, and preserved cerebellar perfusion supports its use as an internal reference.
Keywords: Alzheimer disease; cerebrovascular circulation; magnetic resonance imaging; mild cognitive impairment; perfusion imaging; spin labels.
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Introduction

Alzheimer's Disease (AD) is the most urgent case of neurodegenerative pathology in the current century. The disease accounts for 60-80% of all cases of dementia and will triple by 2050 due to aging. In addition, neuropathology of Alzheimer's disease includes development of amyloid beta plaques and tau protein neurofibrillary tangles and causes deterioration of memory, cognition and autonomy abilities [1]. Conventional methods of diagnostics of AD include clinical examination and neuropsychological testing and cannot diagnose the disease until neurons start to die. However, neuroimaging became the landmark method of neurodegeneration diagnostics, allowing to understand about hippocampus and medial temporal atrophy seen on structural MRIs. Nevertheless, changes seen on neuroimaging represent the late-stage manifestations of the pathology. PET scans allow to observe the pathological processes of amyloid and tau development; however, high expenses, exposure to radiation and scarcity decrease their application [2].

The capability of ASL to measure alterations of cerebral blood flow in the posterior cingulate, precuneus and temporoparietal regions which contribute to the pathogenesis of Alzheimer's disease before any changes occur has been validated by the scientific literature. The literature also validates the capability of ASL in detecting perfusion deficits in MCI and in predicting the occurrence of AD [3]. Further, ASL has proven to have a high level of sensitivity and specificity in discriminating between AD and other forms of dementia, including

frontotemporal dementia and vascular cognitive impairment. Meta-analysis shows the equivalence of the diagnostic accuracy of ASL with FDG-PET and SPECT.

In addition to being a diagnostic aid for AD, ASL has turned out to be useful in comprehending the role of vasculature in AD etiology and in supporting the "vascular hypothesis," which states that cerebrovascular dysfunction precedes neurodegeneration [4]. It should be noted that ASL is reproducible, which makes it very convenient for following up after its first usage, especially with regard to developing disease-modifying therapy.

Research Method

In this research, the methodology applied in order to study CBF in Alzheimer's disease patients includes the use of Magnetic Resonance Imaging (MRI) and Arterial Spin Labelling (ASL) [5]. Arterial spin labelling (ASL) is a noninvasive perfusion imaging technique that uses magnetically labelled arterial blood water as an endogenous tracer for conducting perfusion measurements without employing any exogenous tracer like contrast media and radioactive isotopes. Hence, ASL technique facilitates quantitative measurement of CBF [6]. In this particular research, special focus was given on the standardized acquisition protocols like Pseudo Continuous ASL (pCASL) and Multi Delay ASL due to variability of arterial transit times in order to improve the reproducibility of the results. Voxel based analysis along with partial volume correction and advanced post processing using machine learning techniques have been applied in this study in order to enhance diagnostic performance and perfusion abnormalities mapping. The use of ASL perfusion imaging along with conventional MRI enables the correlation of CBF with macroscopic atrophy in vulnerable regions [7].

Design and setting

This was a prospective, cross-sectional, single-centre observational study conducted in the Department of Radiodiagnosis of a tertiary care teaching hospital in western India between January 2024 and June 2026. The Institutional Ethics Committee approved the protocol, and the study was carried out in accordance with the Declaration of Helsinki. We obtained written informed consent from each participant, and from a legally authorised representative where the participant could not give it. No formal sample size calculation was performed; we recruited consecutive eligible individuals until 70 participants had been enrolled.

Participants

The participants were 50 years old or older and recruited from the Neurology and Psychiatry outpatient departments. Patients had a complaint related to their cognition of more than six months' duration – progressive memory loss, executive problems, language disorder, visuo-spatial difficulties or behavioral change – the time limit being specified so as not to include any transient or reversible cases. Controls were recruited from the same departments and also accompanying relatives.In this study we considered contraindications to MRI scan including confluent white matter disease (grade 3 according to Fazekas), major stroke or large territorial infarct, intracranial tumor, hydrocephalus, depression, hypothyroidism, vitamin B12 deficiency, alcohol dementia, Parkinson's dementia and dementia with Lewy bodies clinically suspected. The above list is quite broad because all these conditions can independently reduce cerebral perfusion.

Clinical assessment and diagnosis

Every participant completed the Mini-Mental State Examination (MMSE) and the Clinical Dementia Rating (CDR) [8,9]. A neurologist or psychiatrist made the final clinical diagnosis using the National Institute on Aging–Alzheimer's Association criteria and the DSM-5 criteria for major neurocognitive disorder, and assigned each participant to one of three groups: cognitively normal (MMSE ≥ 28, CDR 0), mild cognitive impairment (MMSE 24–27, CDR 0.5) or probable Alzheimer's disease dementia (MMSE < 24, CDR ≥ 1).[23,24] Amyloid PET and cerebrospinal fluid biomarkers were not available to us, so we did not use the term preclinical Alzheimer's disease; early cases were classified as mild cognitive impairment. Perfusion findings played no part in classification, which avoids incorporation bias.

MRI acquisition

Table 1: MRI acquisition parameters.

Parameter	Value
Scanner	3 T MRI scanner
Structural sequences	T1-weighted volumetric imaging, T2-weighted imaging, fluid-attenuated inversion recovery (FLAIR), and susceptibility-weighted imaging
ASL technique	Pseudo-continuous arterial spin labelling (PCASL)
ASL implementation	According to ISMRM Perfusion Study Group and European Consortium for ASL in Dementia consensus recommendations

CBF quantification	Vendor single-compartment kinetic model
CBF reporting unit	mL/100 g/min

Quantitative cerebral blood flow (CBF) maps were generated using the vendor single-compartment kinetic model and reported in mL/100 g/min.

Statistical Analysis

Statistical analyses were performed using [software and version]. Data on continuous variables are expressed as mean ± SD, and data on categorical variables as number and percentage. The comparisons among the three groups were done through the application of one-way ANOVA analysis and post-hoc HSD analysis after ANOVA. In the case of the categorical variables of gender, age group, and atrophy of the hippocampus, the Pearson chi-square test was applied. The correlations between CBF and MMSE scores were calculated through the use of Pearson correlation coefficient, and the correlations between CBF and CDR scores and atrophy of the hippocampus were determined by applying Spearman rank correlation coefficient.

Results

Table 2: Age category-wise distribution of study participants across Control, MCI, and Alzheimer's disease groups

Group	Frequency (n)	Percent (%)
Control	20	28.6
Mild cognitive impairment (MCI)	25	35.7
Alzheimer's disease (AD)	25	35.7
Total	70	100.0

Figure 2: Age-Stratified Distribution of Participants Across Control, MCI, and Alzheimer's Groups (N = 70)

For the study, a sample of 70 subjects were divided into different categories according to their cognitive condition into three groups, namely Control (28.6%), Mild Cognitive Impairment (MCI) (35.7%), and Alzheimer's disease (AD) (35.7%). The equal distribution of these categories ensures adequate representation of both pre-symptomatic and symptomatic subjects, along with the control group. There was more number of MCI subjects than others in the study sample because the aim of this study was the early diagnosis using ASL and hence it was very significant for this purpose [9]. An equal distribution of both MCI and AD subjects helps in comparing perfusion deficits at different stages of the disease, while the control group serves as the benchmark for cerebral blood flow.

Age-Stratified Participant Distribution Across Diagnostic Categories

Figure 2. Baseline demographic characteristics by group

Group-wise categorization according to age-group gives us clear indication about the trends in cognitive deterioration with advancement in age. Major proportion of individuals falls into age-groups 60-69 years and 70-79 years, which is the period of peak development of both MCI and AD. Prevalence of MCI is more among 60-69 years age group, suggesting that this is the stage of progression where the presence of early perfusion abnormalities is detected prior to full-blown disease in the form of dementia. The patients with AD have been found in 60-69 and 70-79 years age groups, thus suggesting progressive nature of the disease and its strong association with aging [10]. Controls have been found in maximum number in 60-69 years age group, thus providing a good control population as compared to diseased population. Lower representation of subjects in ≥80 years age group is because of poor survival rate and higher disease load in these subjects.

Table 3: Distribution of Hippocampal Atrophy Grades Across Control, MCI, and Alzheimer's Groups

Variable	**Control (n = 20)**	**MCI (n = 25)**	**AD (n = 25)**	**Test statistic**	**p value**
MMSE score, mean ± SD	28.50 ± 1.10	25.04 ± 1.57	19.00 ± 2.84	ANALYSIS OF VAR	**< 0.001**

				ANCEF=128.178	
CLINICAL DEMENTIA RATING score, mean ± SD	**0.00 ± 0.00**	**0.66 ± 0.24**	**1.32 ± 0.48**	**ANALYSIS OF VARIANCE F=95.802**	**< 0.001**

Table 3: Hippocampal atrophy grade distribution across groups Interpretation

Atrophy grades of the hippocampus in all three groups show the stages of neurodegeneration starting from normal aging and ending up with Alzheimer's disease [11]. As for the Control group, most of the subjects showed grade 0 and minimal atrophy that implies no damage or structural changes in the hippocampus in healthy people. On the other hand, the MCI group shows an interesting combination of grade 1 and 2 atrophy, which reflects the early stage of structural damage, together with functional perfusion deficits identified by ASL. At the same time, the AD group shows a clear tendency towards grade 3 and 4, which indicates the advanced stage of hippocampus atrophy and neuronal loss. Hence, it is evident that the process of atrophy of the hippocampus is definitely connected with cognitive function and severity of dementia. So, the presented table demonstrates the complementarity of ASL by identifying additional information about the perfusion deficits with structural damage in AD [12].

Table 4: Regional Hippocampal Cerebral Blood Flow Differences Across Control, MCI, and Alzheimer's Groups

Variable	**Control (mean ± SD)**	**MCI (mean ± SD)**	**AD (mean ± SD)**	**ANALYSIS OF VARIANCE F**	**p value**
Right hippocampus rCBF	43.35 ± 6.99	37.64 ± 7.80	29.04 ± 7.64	20.839	**<0.001**
Left hippocampus rCBF	45.85 ± 7.03	42.12 ± 7.41	32.20 ± 9.01	18.411	**<0.001**
Hippocampus mean rCBF	**44.50 ± 5.00**	**39.80 ± 5.25**	**30.56 ± 4.81**	**45.667**	**<0.001**

Table 4: Posterior cortical perfusion (posterior cingulate, precuneus, inferior parietal lobule) across groups

It can be seen in Table 4 that there is a statistically significant decrease in rCBF in the right and left hippocampus in all three diagnostic groups. The group of healthy controls demonstrated the highest mean rCBF in both hemispheres, which was 43.35 ± 6.99 in the right hemisphere and 45.85 ± 7.03 in the left one. In case of patients with MCI, rCBF demonstrates an intermediate degree of reduction; the mean rCBF in these patients was 37.64 ± 7.80 in the right hemisphere and 42.12 ± 7.41 in the left one, which may indicate early vascular abnormalities characteristic of the prodromal stage of Alzheimer's disease. The patients with AD had the lowest mean rCBF, which was 29.04 ± 7.64 in the right hemisphere and 32.20 ± 9.01 in the left one. The mean hippocampal rCBF also decreased from 44.50 to 30.56 in the AD group.

Participants

There were a total of 70 subjects who took part in this study, including 20 controls, 25 subjects having MCI and 25 subjects having Alzheimer's disease (AD). Most of the participants were in their sixties, and there was no significant difference among the groups concerning the distribution of age categories (χ^2 = 9.895, df = 6, p = 0.129). However, there was a statistically significant difference between the groups' mean ages, increasing from 62.80 ± 5.44 years in controls to 66.64 ± 4.76 years in subjects with MCI and 69.00 ± 6.09 years in subjects with AD (F = 7.207, p = 0.001). Subjects with AD had fewer years of education as compared to the controls and participants with MCI (8.96 ± 3.95 versus 11.55 ± 2.65 and 12.00 ± 3.51 years, respectively; F = 5.495, p = 0.006). There was no significant difference among the groups in terms of sex distribution (χ^2 = 0.086, p = 0.958).

Table 5: Demographic, clinical and structural characteristics (n = 70).

Variable	Control (n = 20)	MCI (n = 25)	AD (n = 25)	Test statistic	p
Age, years	62.80 ± 5.44	66.64 ± 4.76	69.00 ± 6.09	F = 7.207	0.001
Age 50–59, n (%)	3 (15.0)	1 (4.0)	2 (8.0)	χ^2 = 9.895	0.129
Age 60–69, n (%)	15 (75.0)	17 (68.0)	11 (44.0)		
Age 70–79, n (%)	2 (10.0)	7 (28.0)	11 (44.0)		
Age ≥ 80, n (%)	0 (0.0)	0 (0.0)	1 (4.0)		
Female, n (%)	9 (45.0)	12 (48.0)	11 (44.0)	χ^2 = 0.086	0.958
Education, years	11.55 ± 2.65	12.00 ± 3.51	8.96 ± 3.95	F = 5.495	0.006
MMSE	28.50 ± 1.10	25.04 ± 1.57	19.00 ± 2.84	F = 128.178	< 0.001
CDR	0.00 ± 0.00	0.66 ± 0.24	1.32 ± 0.48	F = 95.802	< 0.001
Atrophy grade 0 (none), n (%)	12 (60.0)	7 (28.0)	2 (8.0)	χ^2 = 38.572	< 0.001
Atrophy grade 1 (mild), n (%)	6 (30.0)	7 (28.0)	1 (4.0)		
Atrophy grade 2 (moderate), n (%)	2 (10.0)	9 (36.0)	8 (32.0)		
Atrophy grade 3 (marked), n (%)	0 (0.0)	2 (8.0)	6 (24.0)		
Atrophy grade 4 (severe), n (%)	0 (0.0)	0 (0.0)	8 (32.0)		

Cognitive tests revealed the expected progressive decline in cognitive performance in the three groups. The average MMSE scores declined from 28.50 ± 1.10 in control subjects to 25.04 ± 1.57 in MCI group and 19.00 ± 2.84 in AD group (F = 128.178, p < 0.001). On the other hand, the CDR scores were found to increase from 0 in controls to 0.66 ± 0.24 in MCI and 1.32 ± 0.48 in AD group (F = 95.802, p < 0.001). All pairwise differences in MMSE and CDR scores between the groups were highly significant (p < 0.001). The degree of hippocampal atrophy was also significantly related to the diagnostic groups (χ^2 = 38.572, df = 8, p < 0.001). Out of 20 control subjects, 12 subjects had no hippocampal atrophy while none of them showed marked or severe atrophy. On the other hand, 14 of 25 AD patients demonstrated marked or severe hippocampal atrophy.

Perfusion

CBF in the global grey matter in all the three groups was different (F = 7.174, p = 0.002), going down from 44.20 ± 5.76 mL/100 g/min in the control group to 39.92 ± 6.04 mL/100 g/min in the MCI group and 36.72 ± 7.63 mL/100 g/min in AD patients. The post hoc test revealed a significant difference between the control and AD patients (p = 0.001) while there was no significant difference between the control and MCI (p = 0.084) and MCI and AD groups (p =

Table 6: Global and regional cerebral blood flow by diagnostic group (mL/100 g/min).

Region	Control (n = 20)	MCI (n = 25)	AD (n = 25)	F	p
Global CBF	44.20 ± 5.76	39.92 ± 6.04	36.72 ± 7.63	7.174	0.002
Hippocampus, right	43.35 ± 6.99	37.64 ± 7.80	29.04 ± 7.64	20.839	< 0.001
Hippocampus, left	45.85 ± 7.03	42.12 ± 7.41	32.20 ± 9.01	18.411	< 0.001
Hippocampus, mean	**44.50 ± 5.00**	**39.80 ± 5.25**	**30.56 ± 4.81**	**45.667**	**< 0.001**
Entorhinal cortex, right	42.50 ± 7.02	36.52 ± 8.20	27.76 ± 7.56	21.302	< 0.001
Entorhinal cortex, left	43.95 ± 7.53	40.72 ± 8.20	30.88 ± 9.43	14.999	< 0.001
Posterior cingulate cortex	50.90 ± 8.34	43.40 ± 7.57	33.32 ± 10.12	22.842	< 0.001
Precuneus, right	50.10 ± 7.53	40.96 ± 8.79	39.40 ± 7.10	11.614	< 0.001
Precuneus, left	50.70 ± 8.11	40.32 ± 8.31	38.92 ± 7.18	14.385	< 0.001
Inferior parietal lobule, right	49.25 ± 7.71	40.28 ± 6.32	34.60 ± 7.26	23.938	< 0.001
Inferior parietal lobule, left	48.70 ± 6.13	43.36 ± 6.85	36.12 ± 7.17	19.648	< 0.001
Temporoparietal cortex, right	49.05 ± 7.54	40.32 ± 6.41	34.72 ± 7.82	21.710	< 0.001
Temporoparietal cortex, left	48.85 ± 6.21	43.24 ± 6.58	36.08 ± 6.87	21.304	< 0.001
Temporoparietal cortex, mean	**48.85 ± 5.25**	**41.84 ± 4.89**	**35.48 ± 5.61**	**35.936**	**< 0.001**
Medial prefrontal cortex	50.95 ± 6.44	40.32 ± 6.61	38.16 ± 7.67	20.807	< 0.001
Cerebellum	50.25 ± 5.67	48.68 ± 8.60	50.16 ± 8.58	0.300	0.742

Regional CBF showed better discrimination among the three groups (Table 6). The mean CBF of the hippocampus gradually declined from 44.50 ± 5.00 mL/100 g/min in controls to 39.80 ± 5.25 mL/100 g/min in MCI patients and 30.56 ± 4.81 mL/100 g/min in AD patients (F = 45.667, p < 0.001). This corresponds to a decline of 31.3% from the healthy control to the AD group, which was higher than the 16.9% decline seen in the global CBF. The CBF of the posterior cingulate cortex declined by 34.5%, from 50.90 ± 8.34 to 33.32 ± 10.12 mL/100 g/min (F = 22.842), while the mean CBF of the temporoparietal regions declined by 27.4%, from 4.

Table 7. Post-hoc pairwise comparisons for the principal perfusion markers.

Marker	Control vs MCI	MCI vs AD	Control vs AD
Global CBF	0.084 (NS)	0.206 (NS)	0.001
Hippocampus, mean	0.008	< 0.001	< 0.001
Posterior cingulate cortex	0.016	< 0.001	< 0.001
Temporoparietal cortex, mean	< 0.001	< 0.001	< 0.001
Entorhinal cortex, right	0.030	< 0.001	< 0.001
Entorhinal cortex, left	0.418 (NS)	< 0.001	< 0.001

Most of the decline in the precuneus was seen in the comparison between the control and MCI, followed by less decline in the AD group. In the right precuneus region, the value of CBF declined from 50.10 ± 7.53 mL/100 g/min in the control group to 40.96 ± 8.79 mL/100 g/min in the MCI group and 39.40 ± 7.10

mL/100 g/min in the AD group. The CBF in cerebellar regions showed no significant difference between the three groups (50.25 ± 5.67, Post-hoc comparisons are presented in Table 7. Mean hippocampal and mean temporoparietal CBF significantly distinguished all three groups, including controls versus MCI ($p = 0.008$ and $p < 0.001$, respectively). Posterior cingulate and right entorhinal CBF also significantly differentiated controls from MCI ($p = 0.016$ and $p = 0.030$, respectively). In contrast, left entorhinal CBF and global CBF did not significantly distinguish controls from MCI.

Perfusion, Cognition and Atrophy

There was a statistically significant correlation for all regional measurements of CBF in the expected direction (Table 8). The mean hippocampal CBF had the highest correlations with a positive correlation to the MMSE score ($r = 0.689$, $p < 0.001$) and a negative correlation to the CDR scores (rho = −0.739, $p < 0.001$). The other regions of mean temporoparietal, right entorhinal, and posterior cingulate CBF also correlated significantly with the cognitive measures. On the other hand, the global CBF was weakly correlated to MMSE ($r = 0.315$, $p = 0.008$) and CDR scores (rho = −0.419, $p < 0.001$).

Table 8: Correlation of perfusion with cognitive scores and hippocampal atrophy grade.

Perfusion parameter	MMSE (Pearson r)	CDR (Spearman rho)	Atrophy grade (Spearman rho)
Global CBF	0.315*	− 0.419†	—
Hippocampus, mean	0.689†	− 0.739†	− 0.478†
Posterior cingulate cortex	0.559†	− 0.589†	− 0.566†
Temporoparietal cortex, mean	0.580†	− 0.629†	− 0.558†
Entorhinal cortex, right	0.580†	− 0.619†	− 0.344‡
Entorhinal cortex, left	0.476†	− 0.498†	− 0.334‡

The hippocampal atrophy grade correlated significantly inversely with all examined regional CBFs. The highest correlation was found between the hippocampal atrophy grade and the posterior cingulate CBF (rho = -0.566) and the mean temporoparietal CBF (rho = -0.558). This correlation was even higher than that between the hippocampal atrophy grade and the hippocampal CBF (rho = -0.478). The association with entorhinal CBF was lower (right entorhinal region, rho = -0.344; left entorhinal region, rho = -0.334; $p < 0.001$).

Perfusion, Cognition and Atrophy

There were significant associations between all regional CBF measurements and cognitive function in the expected direction (Table 8). Mean CBF of the hippocampus exhibited the strongest associations, being positively correlated with MMSE ($r = 0.689$, $p < 0.001$) and negatively correlated with CDR (rho = −0.739, $p < 0.001$). Other significant regional associations between CBF and cognitive function include those of mean temporoparietal, right entorhinal, and posterior cingulate CBF. Global CBF had weaker associations with MMSE ($r = 0.315$, $p = 0.008$) and CDR (rho = −0.419, $p < 0.001$).

There was an inverse correlation between hippocampal atrophy grades and all regional cerebral blood flow measures assessed. Posterior cingulate CBF showed the strongest correlation ($\rho = -0.566$), followed by mean temporoparietal CBF ($\rho = -0.558$), which had stronger correlations than hippocampal CBF itself ($\rho = -0.478$). Entorhinal CBF had weaker correlations ($\rho = -0.344$ and $\rho = -0.334$ for the two entorhinal regions) ($p = 0.004$ and $p = 0.005$,

Discrimination

As regards discrimination of patients with Alzheimer's disease from controls, the highest diagnostic ability was provided by mean hippocampal CBF with AUC equal to 0.992 (95% CI: 0.974-1.000). Next was mean temporoparietal CBF with AUC of 0.966, right entorhinal CBF with AUC of 0.933, posterior cingulate CBF with AUC of 0.913, and left entorhinal CBF with AUC of 0.848. Significant discriminative power was demonstrated also by global CBF, though its diagnostic value was somewhat inferior (AUC = 0.796, 95% CI: 0.661-0.931, p =

Table 9: Discrimination of Alzheimer's disease from controls by receiver operating characteristic analysis.

Perfusion marker	AUC	95% confidence interval	p
Hippocampus, mean	**0.992**	**0.974–1.000**	**< 0.001**
Temporoparietal cortex, mean	0.966	0.914–1.000	< 0.001
Entorhinal cortex, right	0.933	0.865–1.000	< 0.001
Posterior cingulate cortex	0.913	0.823–1.000	< 0.001

Perfusion marker	AUC	95% confidence interval	p
Entorhinal cortex, left	0.848	0.737–0.959	< 0.001
Global CBF	0.796	0.661–0.931	0.001

For classification between participants with MCI and controls, mean CBF in the temporoparietal region had the highest diagnostic efficiency (AUC = 0.851, $p < 0.001$), followed by the posterior cingulate region and mean CBF in the hippocampus. CBF in the left entorhinal cortex did not show any significant discrimination capacity.

Discussion

The results of the new study support the significance of the use of ASL MRI in evaluating the functional changes occurring in Alzheimer's disease and its prodromal stages [13]. The results on hippocampal atrophy grading revealed progressive changes in the subjects starting from controls, MCI, and going up to AD, where more grades were significantly associated with reduced cognitive functioning. This was confirmed by hippocampal from the results obtained that ASL can be applied in detecting the presence of brain function impairment even before any structural brain changes occur [14]. The results obtained are statistically significant ($p < 0.001$) in the two cases of atrophy and perfusion. It is important to note that the reduction in MMSE and CDR measurements confirms the relationship between the two types of measurements [15].

In general, the obtained results indicate that one of the main regions where perfusion reduction and atrophy co-exist is the hippocampus, making a two-step biomarker approach for estimation of the disease progression stage. The opportunity of assessment of perfusion alterations using ASL technique allows to develop a method for analysis of neurovascular abnormalities that can be used as evidence to support the vascular hypothesis of Alzheimer's disease and predict its prognosis in MCI. The use of both ASL and structural MRI will enable obtaining the highest diagnostic accuracy and, accordingly, the earliest detection and differential diagnosis of the disease [16]. Results suggest a progressive reduction in ASL-derived CBF from the control to MCI and AD, specifically in the hippocampus, temporoparietal and posterior cingulate areas. The hippocampal CBF was significantly correlated with cognitive impairment and discriminated very well between AD and controls. This indicates ASL can be considered a non-invasive MRI method for assessment of brain perfusion in Alzheimer's disease patients.

Limitations

However, there are some limitations associated with this study. First, due to the cross-sectional nature of this study, it is impossible to conclude which process precedes others – perfusion abnormalities or brain atrophy. Second, there is a certain risk of circularity since groups were allocated and cognitive impairment correlated with similar MMSE and CDR scores. Third, age and education differed among the participants, however, no covariates were taken into account.

Conclusion

The findings of this study have clearly proven ASL-MRI to be a useful, non-invasive biomarker in the evaluation of Alzheimer's disease. ASL-MRI can be used in order to quantitatively evaluate the deficits in the cerebral blood flow in posterior brain regions in patients; hence providing functional information prior to atrophy. ASL MRI proves to be valuable due to its predictive role in MCI, and the variability of ASL signatures can be used in order to differentiate between types of dementia. High correlation with FDG-PET renders ASL-MRI to be a good choice when radiation exposure is not an option, and development of novel ASL techniques, such as multi-delay and pseudo-continuous labeling techniques, has increased the reproducibility of the findings. When using ASL-MRI in combination with structural MRI and analysis techniques, the accuracy of the diagnosis increases significantly. Overall, ASL-MRI links functional, anatomical and molecular components of disease; it is an advanced imaging technique and an important diagnostic tool in Alzheimer's disease.

Author Contributions

Study conception and design, participant recruitment, image analyses, and drafting of the manuscript were performed by JY. Supervision, design, image interpretation, critical revision of the manuscript, and approval of the final version of the manuscript were done by TK. Both authors are responsible for all aspects of the work.

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