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ORIGINAL PAPER

Age-associated hippocampal volume changes in childhood arterial ischemic stroke

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Abstract

Purpose Recent evidence suggests that recovery from secondary neurodegeneration following arterial ischemic stroke (AIS) may be related to age at injury and site of occlusion. We conducted a study of hippocampal volume (HCV) in a cohort of pediatric patients with middle cerebral artery (MCA) territory AIS to determine whether HCV would be preserved in younger children as compared to older children.

Methods This single-center, HIPAA-compliant retrospective study was approved by the institutional review board. The medical records of 149 children treated for AIS between 2000 and 2016 were reviewed for inclusion criteria: unilateral MCA territory AIS and availability of high-resolution T1-weighted MR imaging at both acute and chronic time periods. Manual segmentation was utilized to measure stroke-side HCV, contralateral HCV, hemispheric volumes, and stroke volume on each scan. To correct for variable brain size, HCV measurements were ratio normalized. Patients were divided into two age-at-stroke groups: younger (30 days–9 years old) and older (> 9–18 years old). Analysis was performed using Fisher's test or Student's *t* test.

Results The MR imaging of 19 children (9 younger, 10 older) was analyzed. At follow-up, the average stroke-side HCV increased by 10.9% in the younger group and decreased by 6.3% in the older group ($P = 0.010$); this between-group difference remained significant even when ratio normalized ($P = 0.003$). The total brain volume-adjusted acute stroke size between groups was not statistically different ($P = 0.649$).

Conclusions In children with AIS, younger age is associated with the relative preservation of HCV, which could reflect differences in age-related plasticity.

Keywords Magnetic resonance imaging · Arterial ischemic stroke · Pediatric stroke · Hippocampal volume

Introduction

The hippocampus, although primarily supplied by the posterior cerebral artery, is highly vulnerable to secondary neurodegeneration following middle cerebral artery (MCA) occlusion [1, 2]. In children with arterial ischemic stroke (AIS), the capacity for plasticity is likely influenced by age at the time of injury [3, 4]. To what extent these age-dependent mechanisms protect neurons, particularly in the hippocampus, remains unclear.

Using an MCA occlusion mouse model, we obtained data that suggest that post-stroke hippocampal recovery is largely dependent on age at stroke [5]. In this model, both juvenile and adult mice exhibited synaptic dysfunction in the hippocampus early in recovery (24 h and 7 days post-occlusion). While this dysfunction persisted in adult mice at later recovery (30 days post-occlusion), juvenile mice showed significant improvement in hippocampal synaptic activity during this same time period.

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To translate the clinical relevance of the mouse model findings, we conducted a study of hippocampal volume (HCV) in a cohort of pediatric patients with MCA territory AIS to determine whether HCV would be preserved in younger children as compared to older children.

Methods

This single-center, HIPAA-compliant retrospective study was approved by the institutional review board (COMIRB 14-0443). The medical records of 149 children (> 30 days and < 19 years old at symptom onset) treated for AIS at a regional children's hospital between 2000 and 2016 were retrospectively reviewed. Inclusion criteria were unilateral MCA territory stroke and availability of 3D T1-weighted isotropic (1-mm voxel size) MR scans suitable for manual hippocampal volumetry at both acute (< 72 h since diagnosis) and chronic (> 90 days since diagnosis) time periods. A pediatric neuroradiologist and a pediatric neurologist independently confirmed the diagnosis of AIS in each patient and classified causes of stroke using the CASCADE criteria [6]. Cases with any diagnostic uncertainty or poor imaging resolution were excluded. Demographic and clinical information, including age at stroke, sex, race, ethnicity, presenting signs and symptoms, and treatment, were recorded via chart review.

Imaging protocol

The 3D T1-weighted isotropic images were acquired on one of three platforms at our institution, including (1) 1.5T Siemens Avanto with 3D T1-weighted MPRAGE (TR = 1700–1800 ms; TE = 2.92 ms; section thickness = 1.0 mm; field of view = 250 mm, matrix = 246 × 256, reconstructed voxel size 1 mm × 1 mm × 1 mm), (2) 1.5T Philips Ingenia with 3D T1-weighted TFE (TR = 8–9 ms; TE = 4.6 ms; section thickness = 1.0 mm; field of view = 256 mm, matrix = 256 × 256, reconstructed voxel size 1 mm × 1 mm × 1 mm), and (3) 3T Philips Ingenia with 3D T1-weighted TFE (TR = 8–9 ms; TE = 3.8 ms; section thickness = 1.0 mm; field of view = 240 mm, matrix = 220 × 240, reconstructed voxel size 1 mm × 1 mm × 1 mm).

Data collection

MRI-based manual segmentation software (Aquarius iNtuition, TeraRecon, Forest City, CA) was used to measure stroke-side HCV, contralateral HCV, hemispheric volumes, and stroke volume on each scan. Hippocampal measurements were performed on 3D T1-weighted images by a trained medical student and confirmed by a blinded pediatric neuroradiologist. All hippocampi were measured using a three-plane approach, which involved initial manual segmentation in the coronal plane followed by boundary corrections in the axial and sagittal planes. Hippocampal boundaries were conventionally defined by landmarks detailed in the harmonized protocol for hippocampal segmentation [7]. Each HCV measurement was performed at least twice and only deemed reliable for study inclusion if the two latest trials varied < 10%.

Hemispheric volumes and acute stroke volumes were measured by a blinded pediatric neuroradiologist. Acute stroke volumes were segmented from areas of hyperintensity on diffusion-weighted images indicative of cytotoxic edema using a combination of the planimetric method [8] and volume thresholding in Aquarius iNtuition. Acute and chronic scans were evaluated for signs of hippocampal sclerosis by a pediatric neuroradiologist.

Statistical analysis

Patients were categorized into two age-at-stroke groups: younger (30 days old to 9 years old) and older (greater than 9 years old to 18 years old). The cutoff at age 9 was designated a priori to divide the pediatric age spectrum in half yielding two reasonably sized age-at-stroke groups, although this categorization approach is also supported by findings that suggest hippocampal growth increases until 9–11 years of age [9].

To correct for variable brain size and brain growth, raw HCV measurements were normalized to HCV ratios (HCVR; stroke-side HCV/contralateral HCV). The difference between acute and chronic HCVR was calculated and compared between groups. All data analysis was performed by Fisher's exact test or Student's *t* test using GraphPad software accessed at http://www.graphpad.com/quickcalcs. $P < 0.05$ was deemed statistically significant. The datasets generated during the current study are available from the corresponding author on request.

Results

Seventy children without isolated unilateral MCA territory AIS and 60 children without available high-resolution T1-weighted imaging at acute and chronic time points were excluded. Nineteen children met inclusion criteria; nine children in the younger age-at-stroke group and ten in the older group, with an average age at stroke of 9.1 years (Table 1). Gender mix and stroke laterality were not significantly different between the two groups.

The most common presenting signs and symptoms included hemiparesis ($n = 18/19$), speech deficit ($n = 10/19$), reduced level of consciousness ($n = 5/19$), headache ($n = 5/19$), and seizure ($n = 5/19$). There were two cases with persistent seizures (> 1 month post-AIS) documented in the younger group.

CASCADE criteria classification included unilateral focal cerebral arteriopathy of childhood (seven), other/idiopathic (six), cardioembolic (four), bilateral cerebral arteriopathy of

Table 1 Comparison of clinical characteristics

	All ($n = 19$)	Younger ($n = 9$)	Older ($n = 10$)	P value
Total, *n*	19	9	10	–
Average age at AIS, y	9.1	4.8	13.0	*< 0.001*
Gender, *n*				0.070
Female	10	7	3	
Male	9	2	7	
Stroke side, *n*				1.000
Right	7	3	4	
Left	12	6	6	
Race/ethnicity, *n*				0.582
Hispanic white	4	1	3	
Non-Hispanic black	1	1	0	
Non-Hispanic white	13	6	7	
More than one race	1	1	0	
Presenting signs and symptoms, *n*				0.801
Headache	5	1	4	
Hemiparesis	18	9	9	
Reduced consciousness	5	2	3	
Seizure	5	3	2	
Speech deficit	10	5	5	
Vomiting	4	1	3	
Seizure activity > 1 month post-AIS, *n*	2	2	0	0.211
Average time between scans, y	1.3	1.8	0.8	*0.002*
Treatment at diagnosis, *n*				0.347
ASA	11	5	6	
UFH	4	3	1	
TPA	2	0	2	
None	1	0	1	
Uncertain	1	1	0	
Treatment at discharge, *n*				1.000
ASA	14	7	7	
LMWH	4	2	2	
None	1	0	1	

Statistically significant differences between groups defined as p values less than 0.05 are italicized

AIS arterial ischemic stroke, *ASA* aspirin, *LMWH* low-molecular-weight heparin, *n* number, *TPA* tissue plasminogen activator, *UFH* unfractionated heparin, *y* years

childhood (one), and aortic/cervical arteriopathy (one). The underlying diagnoses were typical of what is commonly seen in childhood stroke cohorts.

Following diagnosis, most patients in the cohort received either aspirin ($n = 11/19$) or unfractionated heparin ($n = 4/19$); two children in the older group were given tissue plasminogen activator (Table 1).

The average time between acute and chronic MRI scans was 1.3 years, with the interval between scans longer in the younger group (1.8 years) than the older group (0.8 years). Total brain volume-adjusted acute stroke size between the younger (5.4%) and older (7.6%) groups were not statistically different ($P = 0.649$; Table 2). In no cases were new areas of infarction detected on chronic imaging that were not originally present on acute imaging. No findings of hippocampal sclerosis were identified on any acute or chronic imaging.

Comparing stroke-side HCV change over time, the younger group exhibited 10.9% growth, while the older group showed 6.3% loss ($P = 0.010$; Figs. 1 and 2; Table 2). The between-group difference remained significant even when HCV was ratio normalized ($P = 0.003$). There was no significant difference in contralateral HCV volume change between the younger (5.5%) and older groups (− 0.4%; $P = 0.357$).

Discussion

In our cohort of children with AIS, the younger age-at-stroke group exhibited a trend towards hippocampal growth ipsilateral to the MCA infarction, while the older group experienced ipsilateral HCV loss. Further, the HCVR increased in the younger group compared to the older group, providing an internal control to account for the overall hippocampal growth expected in the younger group. The results suggest that part of the mechanisms for plasticity and stroke recovery in the younger age group could result in compensatory enlargement of structures remote from the area of infarction, such as the hippocampus. This finding may also reflect differences in age-related plasticity analogous to that seen in a juvenile mouse model of AIS [5]. Other animal studies have shown that increased ischemic sensitivity is correlated with maturation of excitatory synapses and maximal synaptogenesis [10, 11]. Therefore, the age-associated trends in HCV change that we observed may be, in part, explained by variances in synaptic maturity at stroke onset.

To our knowledge, this is the first report of HCV changes occurring across the age spectrum in childhood AIS. Only one other study, led by Gold and Trauner, has previously described HCV changes in children with perinatal stroke [12]. These researchers observed that perinatal stroke patients exhibited significant HCV loss ipsilateral to the side of stroke when compared to age-matched controls. They also found that post-stroke HCV loss was correlated with seizures and strongly associated with poor performance on IQ and memory assessments. Since we compared to a different aged stroke cohort and not controls, these discrepant results may not be directly applicable to our study. Additionally, perinatal stroke is a notably different entity than childhood stroke, with different risk factors and outcomes than childhood stroke in general.

Limitations of this study included its small sample size and requirement of high-resolution MRI sequences, with the latter primarily contributing to the former. In addition, some variability between the older group and younger group may have

Table 2 Comparison of volume measurements

	All ($n = 19$)	Younger ($n = 9$)	Older ($n = 10$)	*P* value
Average HCV, cc				
Acute stroke-side HCV	2.93	2.48	3.34	*0.003*
Acute contralateral HCV	2.97	2.58	3.32	*0.012*
Chronic stroke-side HCV	2.96	2.74	3.16	*0.050*
Chronic contralateral HCV	3.03	2.71	3.31	*0.010*
Stroke-side HCV acute-chronic difference	0.03	0.27	− 0.18	*0.010*
Stroke-side HCV acute-chronic difference (as %[a])	1.9%	10.9%	− 6.3%	*0.010*
Contralateral HCV acute-chronic difference	0.06	0.13	0.00	0.412
Contralateral HCV acute-chronic difference (as %[a])	2.4%	5.5%	− 0.4%	0.357
HCVR[b]				
Acute ratio	1.00	0.96	1.01	0.077
Chronic ratio	0.98	1.02	0.95	0.069
Ratio acute-chronic difference	0.00	0.05	− 0.05	*0.003*
Average hemispheric volume, cc				
Acute stroke-side hemispheric volume	557	479	627	*0.004*
Acute contralateral hemispheric volume	545	468	615	*0.003*
Chronic stroke-side hemispheric volume	498	441	549	*0.018*
Chronic contralateral hemispheric volume	549	503	591	*0.018*
Stroke-side hemispheric volume acute-chronic difference	− 60	− 39	− 78	0.219
Contralateral hemispheric volume acute-chronic difference	4	35	− 24	*0.012*
Average stroke volume, cc				
Acute stroke volume	72.0	47.6	94.0	0.428
Acute stroke volume (as % of TBV[c])	6.6%	5.4%	7.6%	0.649
Chronic stroke volume	37.0	14.5	58.0	0.292
Chronic stroke volume (as % of TBV[c])	3.3%	1.7%	4.7%	0.371
Stroke V acute-chronic difference	− 35.0	− 33.1	− 36.0	0.879

cc cubic centimeters, *HCV* hippocampal volume, *HCVR* hippocampal volume ratio, *TBV* total brain volume

[a] HCV % change was calculated by the following formula: 1 − (acute HCV / chronic HCV)

[b] HCVR was calculated by the following formula: stroke-side HCV / contralateral HCV

[c] TBV was calculated by adding stroke-side and contralateral hemispheric volume

acted as a confounder in our study. Especially, the longer follow-up time in the younger group may have allowed more time for stroke recovery, although initial volume loss followed by volume gain seems unlikely during this timeframe. Another possibility is that the slightly larger stroke size in the older group may have contributed to greater HCV loss in

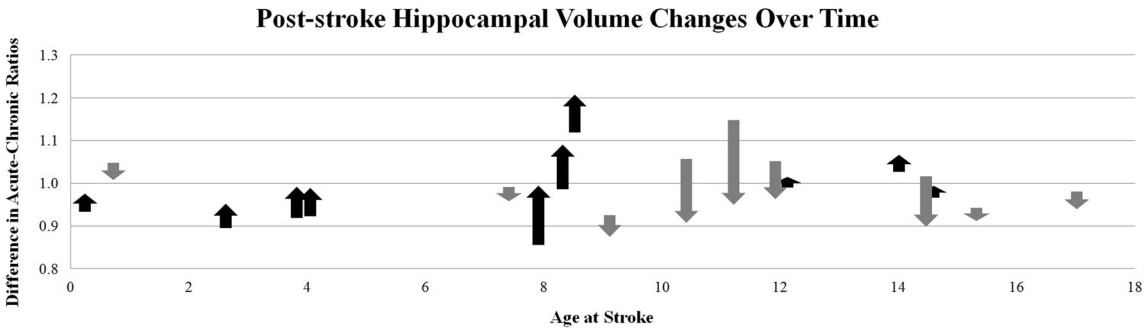

Fig. 1 Post-stroke hippocampal changes across the pediatric age spectrum. Acute and chronic HCVR are represented by the arrow base and the arrow tip, respectively. Comparing relative stroke-side HCV, children with AIS occurring before age 9 demonstrated a trend towards growth (black arrows), while older children with stroke exhibited a trend towards loss (gray arrows)

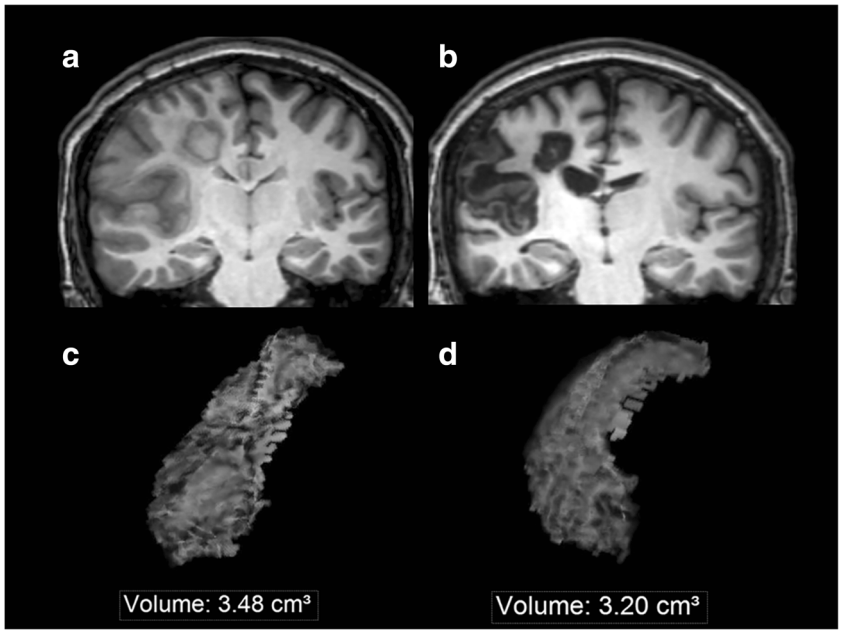

Fig. 2 Representative images from a 17-year-old male who suffered a right MCA territory AIS. Acute imaging performed 2 days after presentation demonstrates gyral edema on coronal T1 images (**a**) that matured into cystic encephalomalacia on MRI performed at 3-month follow-up (**b**). Three-dimensional volume rendered images of the right hippocampal segmentation performed on the acute (**c**) and follow-up (**d**) exams show loss of stroke-side HCV over time

this population. However, this is doubtful considering that the between-group difference would remain statistically significant even if we excluded the two older age-at-stroke patients with the greatest stroke volumes. The age threshold of 9 years was convenient as it resulted in evenly divided groups, although other data demonstrating hippocampal growth ends around this age provides further justification for it. While a well-defined clinical or physiologic rationale to use this age threshold is otherwise lacking, the fact that hippocampal volumes matured differently in these cohorts is felt to still support the preliminary mouse data. Acute and chronic imaging was sometimes performed on different MRI platforms, which could introduce systematic errors into the measurement of hippocampal volumes; however, all the isotropic 3D T1-weighted sequences used were Alzheimer's Disease Neuroimaging Initiative (ADNI) compatible, and cross-platform comparisons of these types of sequences are common in other literature.

Although the sample size is not large enough to formally assess the potential confounders of follow-up time and stroke size, we believe that our observation of younger-onset hippocampal growth and older-onset hippocampal loss correlates with our preliminary animal data and suggests that children in the younger group are demonstrating a meaningful degree of stroke recovery manifested as compensatory hippocampal enlargement ipsilateral to the infarction. As such, these findings need to be confirmed in larger multi-center cohorts, as well as validated using neuropsychological testing.

In summary, stroke-side HCV in younger children with AIS is relatively preserved and relatively increased to the contralateral HCV compared to older children, and this finding may reflect differences in age-related plasticity analogous to that seen in a juvenile mouse model of MCA occlusion. Future studies are needed in larger cohorts with correlative neuropsychological testing.

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Compliance with ethical standards

Conflict of interest On behalf of all authors, the corresponding author states that there is no conflict of interest.

Ethical approval For this type of study, formal consent was not required.

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