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The role of diffusion tensor imaging in detecting hippocampal injury following neonatal hypoxic-ischemic encephalopathy

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Abstract

Background and Purpose: Neonatal hypoxic-ischemic injury of the brain and resultant encephalopathy (HIE) leads to major developmental impairments by school age. Conventional/anatomical MRI often fails to detect hippocampal injury in mild cases. We hypothesize that diffusion tensor imaging (DTI) has greater sensitivity for identifying subtle hippocampal injury.

Methods: We retrospectively analyzed DTI data collected from a cohort of neonates with HIE and controls. Conventional MRI sequences were classified qualitatively according to severity using a modified Barkovich scale. Using multivariate linear regression, we compared hippocampal DTI scalars of HIE patients and controls. Spearman correlation was used to test the association of DTI scalars in the hippocampal and thalamic regions. A multiple regression analysis tested the association of the DTI scalars with short-term outcomes.

Results: Fifty-five neonates with HIE (42% males) and thirteen controls (54% males) were included. Hippocampal DTI scalars were similar between HIE and control groups, even when restricting the HIE group to those with moderate-to-severe injury (8 subjects). DTI scalars of the thalamus were significantly lower in the moderate-to-severely affected patients compared to

controls (right FA 0.148 vs. 0.182, $p=0.01$; left FA 0.147 vs. 0.181, $p=0.03$). Hippocampal and thalamic DTI scalars were correlated ($p<0.001$). Hippocampal DTI scalars were not associated with short-term outcomes.

Conclusions: Quantitative DTI analysis of the hippocampus in neonates following HIE is a feasible technique to examine neuronal injury. Although DTI scalars were useful in identifying thalamic injury in our cohort, hippocampal DTI analysis did not provide additional information regarding hippocampal injury following HIE.

Keywords

Diffusion tensor imaging; hypoxic-ischemic encephalopathy; hippocampus; brain injury

INTRODUCTION

Neonatal hypoxic-ischemic encephalopathy (HIE) affects up to 3 per 1000 live births in developed countries.¹ HIE is associated with a high risk of cerebral palsy, developmental delay and mortality worldwide.^{2, 3} Therapeutic hypothermia (TH) is an effective therapy to improve motor domains of neurodevelopment. However, 25 to 75% of neonates still exhibit subsequent impairments, including deficits in cognition and memory, possibly due to incomplete protection of the hippocampus.^{4–7}

MRI is the gold standard for the evaluation of injury in patients with HIE, but conventional MRI techniques fail to detect likely microstructural injuries that explain the significant developmental delays seen in survivors of HIE.⁸ Diffusion tensor imaging (DTI) is able to examine subtle changes to the brain structure in HIE. DTI characterizes the diffusion of water molecules in three-dimensions in the brain and quantifies various components of diffusion. Powerful post-processing algorithms generate maps of isotropic versus anisotropic diffusion including diffusion weighted images, apparent diffusion coefficient (ADC), fractional anisotropy (FA), color-coded FA and radial/axial diffusivity maps. DTI scalars, such as FA, mean diffusivity (MD), radial diffusivity (RD) and axial diffusivity (AD), are quantifiable measures that can be extracted from a specific voxel or by averaging data over a region of interest. Each scalar imparts different information on the directional patterns of diffusion and correlates with injury in various types of neuropathology. For example, tissue edema and inflammation result in increased MD.⁹ FA is a summary measure of multiple factors affecting diffusion in the white matter. Lowered FA values are common in multiple disease states and thus FA is a sensitive but not specific marker.¹⁰ In neonates with HIE, low FA values in various regions including the corpus callosum and centrum semiovale are associated with worse developmental outcomes.^{11–15}

Hippocampal involvement is commonly found in pathological specimens from neonates who have died of HIE.¹⁶ Hippocampal injury is also observed on conventional MR imaging when other signs of severe injury are also apparent.^{17–20} However, limited data exist on the use of DTI to examine the degree of hippocampal injury in HIE and its subsequent implications for prognostication and clinical management. TH is known to be neuroprotective of hippocampal cells in the animal model in the short term.^{21, 22} DTI may assist in determining therapeutic efficacy of hypothermia and other neuroprotective therapies in humans.

In this study, we used DTI to identify and quantify various diffusion characteristics of the hippocampus, which may provide more detailed information about tissue integrity following HIE. We hypothesized that there will be a significant decrease in FA values and increase in MD values of the hippocampus in the infants affected by HIE compared to controls. We anticipated a significant difference in AD and RD values between these two groups as well. We also hypothesized that the degree of diffusion changes in the hippocampus will correlate with the degree of thalamic DTI scalars given the global nature of HIE, and with short-term outcomes.

METHODS

This study was approved by the Institutional Review Board and, given its retrospective nature, informed consent was waived.

Patients and Controls

Neonates included in this study met the following criteria: (1) a diagnosis of neonatal encephalopathy and treatment with TH, (2) MRI and DTI imaging without significant artifacts that allowed for high-quality processing, performed between day of life 4 and 18, and (3) gestational age greater than or equal to 36 weeks. Patients were excluded from the analysis if: (1) TH was discontinued prior to 72 hours, (2) they were diagnosed with potential causes of neonatal encephalopathy other than hypoxic-ischemic injury (3) infection was strongly suspected (bandemia greater than 10% for more than 3 days, treatment with antibiotics for more than 10 days or positive blood cultures present). A retrospective chart review of all patients who underwent TH between January 1, 2010 and January 31, 2016, in the Johns Hopkins Neuro-Intensive Care Nursery was performed and eligible neonates were identified. Controls for the study were identified from neonates who had studies performed for other clinical indications in our pediatric MRI database during the same epoch using the identical imaging approach/parameter. Controls met the following criteria: (1) MRI was performed in the first 28 days of life, (2) no anatomical abnormalities were identified, (3) no neurologic conditions were identified, (4) gestational age was between 36 and 41 weeks, and (5) MRI and DTI data were available without artifacts. Most studies were performed for extra-cranial head and neck pathologies or evaluations for genetic syndromes. No associated intracranial pathology was found.

Demographic and baseline clinical data, which included sex, gestational age, APGARs, arterial cord gas pH and base excess were collected retrospectively. Total number of days of admission in our level IV NICU, time to full oral feeds, and presence of clinical or treated electrographic seizures were recorded as short term outcomes.

Acquisition of MRI Data

A 1.5-Tesla clinical MRI scanner (Aera, Siemens, Erlangen, Germany) was used to obtain all brain MRI images using our previously described standard protocol which is optimized for neonatal brain imaging.²³ MR sequences included isotropic three-dimensional T1-weighted and axial T2-weighted imaging, susceptibility-weighted imaging (SWI), and a single-shot spin echo, echo-planar axial DTI sequence with diffusion gradients along 20

non-collinear directions (effective low and high b value of 0 respectively 800 s/mm²). Uniform DTI parameters were used to ensure consistency between patients and controls, and protocols were consistent throughout the study period. The DTI parameters were as follows: repetition time 8,500 ms, slice thickness 2.0 mm, echo time 86 ms, field of view 240 × 240 mm, and matrix size 192 × 192. A parallel-imaging integrated acquisition technique with generalized autocalibration was performed twice to improve the signal-to-noise ratio as well as balanced diffusion gradients to minimize eddy current artifacts. Neonatal imaging was typically performed in natural sleep; in rare cases rectal midazolam was used if motion artifacts impacted image quality.

Qualitative MRI Analysis

All patients were assessed for severity of HIE-related injury based on qualitative analysis of conventional, trace of diffusion and apparent diffusion coefficient maps. Patients were categorized (according to a modified Barkovich scale ²⁴) as follows: (0) normal, (1) mild injury, i.e., injury of the periventricular white matter (T2- hyperintense signal or restricted diffusion), (2) moderate injury, i.e., injury of the basal ganglia/thalamus or cortex, and (3) severe injury, i.e., injury of the basal ganglia/thalami and cortex. Image analysis was performed by board-certified pediatric neuroradiologists with 10–24 years of experience.

Quantitative DTI Analysis

DTI raw data were collected on patients meeting the inclusion criteria. DTI Studio software was utilized to co-register and correct for eddy current and motion artifacts using a 12-mode affine transformation.²⁵ Maps of FA, trace of diffusion and ADC were generated. DiffeoMap software was used to perform a rigid transformation to our atlas in order to normalize the orientation of the images.²⁶ Following this, regions of interest (ROI) were drawn manually using ROI Editor software in the bilateral hippocampi and thalami (Figure 1).²⁷ For each structure, ROI were drawn on 5 contiguous slices and mean FA, Eigenvalue 0 and trace of diffusion scalars were generated. ROI were drawn manually in consensus by two study team members (E.O. and J.S.). MD values were calculated with the following formula: MD = trace of diffusion value divided by 3.

Statistical Analysis

Paired t-tests were performed to assess for differences between the right and left sides in FA values for the hippocampus and thalami. There were significant differences thus the subsequent statistical analysis was performed without averaging the two sides. To compare the hippocampal DTI scalars of neonates with HIE and controls multiple linear regression analysis controlling for gestational age and age at time of MRI was performed. A sub-group analysis of the neonates with severely affected MRI studies as evidenced by qualitative analysis (categories 2 and 3 on Barkovich scale) and controls was performed. Using a Spearman rank correlation, partialling gestational age correlated hippocampal DTI scalars with thalamic DTI scalars. Multiple logistic regression analysis and proportional hazards regression analysis were used to test the association of the DTI scalars with short-term outcomes in the HIE patients. Analyses were performed using SAS software v9.3 (SAS Institute, Inc., Cary, NC, USA). All tests were two-sided, and significance was set at $p < 0.05$. A trained biostatistician (K.A.C.) performed all statistical analyses.

RESULTS

Patient and Controls

One hundred and fifty-five neonates with HIE underwent TH at our institution during the study period. One hundred neonates were excluded due to lack of high quality DTI data or significant artifacts ($n=79$), gestational age ($n=10$), premature discontinuation of TH ($n=5$), suspected infection ($n=4$), MRI obtained after 18 days of life ($n=1$) or concurrent severe congenital heart disease ($n=1$). Fifty-five neonates (23 male) met criteria for inclusion. Demographic and clinical characteristics of each population are presented in Table 1.

Qualitative MRI Analysis

MRI findings were analyzed qualitatively (Table 2). Forty-seven HIE patients were classified as normal (category 0) or mildly affected (category 1). Eight neonates were categorized as having moderate or severe findings by MRI criteria.

DTI Analysis

DTI scalars are shown in Tables 3–6. There was no significant difference in FA, MD, RD and AD values in the hippocampus between the patients and controls for the entire cohort (Table 3). A subset analysis was performed on the eight patients who had moderately or severely affected MRI studies. No significant difference in DTI scalars of the hippocampus were noted between this subset population and controls (Table 4).

As shown in Table 5, the right mean FA in the thalamus was lower in subjects than controls (0.164 vs. 0.182, $p=0.02$). In the subset analysis of moderate-to-severely affected patients in Table 6, FA values on both sides of the thalamus were lower in subjects than controls (right mean FA 0.148 vs. 0.182, left mean FA 0.147 vs. 0.181, $p=0.01$ and $p=0.03$, respectively). Scatterplots demonstrating the changes in hippocampal FA and MD values in relationship to thalamic scalars are displayed in Figure 2. Right- and left-sided FA scalars in the hippocampus were directly correlated with those in the right and left thalamus, ($r=0.56$, $p<0.001$ and $r=0.57$, $p<0.001$, respectively). Similarly, right- and left-sided MD scalars in the hippocampus were directly correlated with those in the right and left thalamus, ($r=0.57$, $p<0.001$ and $r=0.55$, $p<0.001$, respectively).

Associations between DTI scalars in the hippocampus and short term outcomes are shown in Table 7. There was no significant association between hippocampal DTI scalars and time to full oral intake, length of NICU stay, or the presence of seizures.

DISCUSSION

Our study is the first quantitative assessment of DTI scalars in the hippocampus of neonates with HIE treated with TH. We did not find a statistically significant difference in hippocampal DTI scalars between infants affected by HIE and those in control infants. However, FA values were statistically significantly lower in the thalami of severely affected neonates with HIE compared to controls, consistent with the literature.^{28, 29} The correlation between thalamic and hippocampal DTI scalars serves as a validation of our hippocampal measurements and is consistent with the evidence that both the hippocampus and deep gray

matter are injured, particularly when the hypoxic-ischemic insult is severe.^{19, 20, 30} Thus, injury associated with HIE does not occur independently in the hippocampus and the thalamus, but instead HIE causes global injury and disruption of extensive networks between these two brain regions.^{31–34}

Despite known hippocampal-thalamic connection, diffusion changes were noted in the thalami of severely-affected neonates in our study, but not in the hippocampi. The presence of persistent thalamic changes is significant for its potential developmental implications.³⁵ Our inability to identify hippocampal injury is either due to no injury being present, the attenuation of injury by TH more so than in the thalami or the inability of DTI to detect these changes. Given the global nature of HIE and the long term hippocampal atrophy seen in prior studies,^{36, 37} the likelihood of no injury to the hippocampus being present is low. Studies of the hippocampal volume in children who had HIE as neonates included subjects that were born prior to the global institution of TH. The effects of this therapy on hippocampal injury and its detection are unclear and may ameliorate the degree of injury significantly. The thalamus may be more severely affected by the hypoxic-ischemic event or may have less injury attenuation by TH, accounting for its persistent diffusional changes after TH in moderately-severely affected neonates. Microstructural changes and metabolic derangements are reduced by TH in the thalamus; however evidence of injury remains.³⁸ Our inability to identify thalamic injury in the normal to mildly affected group may be a reflection of these microstructural improvements. A comparison of pre- and post-hypothermia DTI studies of the hippocampus would be required to elucidate the effect of TH on the hippocampal injury. Our cohort also included comparatively fewer moderately and severely affected neonates which limited our ability to compare sub-groups. The degree of diffusional changes is likely continuum according to injury severity. The comparison of specific injury severity subtypes would better define where the changes are most significant and may reveal hippocampal differences that are not apparent in the current investigative model.

Hippocampal injury following HIE has been demonstrated in animal studies, showing injury of the CA1-CA3 regions of the hippocampus and dentate gyrus granule cells following hypoxic-ischemic insult.^{39–44} Diffusion changes correlate with hippocampal injury in a mouse model.⁴⁵ However, there is a dichotomy in the ADC changes of different regions within the hippocampus in mice.⁴⁶ CA1 and CA2 regions of the hippocampus showed reduced ADC values while ADC values were increased in the adjacent white matter.⁴⁶ DTI analysis of the hippocampal afferent fibers in the mouse model does not reveal a significant difference in FA or MD values following mild to moderate injury compared to controls, mirroring our findings.⁴⁷ However, despite a lack of recognized acute injury, there is a significant alteration in prefrontal-hippocampal networks.⁴⁷ Therefore, there may be undetected but clinically relevant injury present. These studies were performed on mice without TH. TH has been shown to reduce histologic evidence of injury in the hippocampus in sheep and mice, but it remains unclear to what extent this protection persists at later points.^{21, 22} To better answer these questions in humans, it is essential to find an imaging technique that has the sensitivity to detect subtle injury of the hippocampus.

Multiple descriptive studies examined short term outcomes including time to oral feeds and the frequency of seizures amongst neonates with HIE before and after the institution of TH.^{48, 49} A multi-center observational study demonstrated prolonged hospital stays, reduced likelihood of being discharged home on full oral feeds and increased seizure occurrence in patients who were more severely affected compared to those with mild HIE.⁵⁰ However, the prognostic significance of imaging findings, particularly in regards to individual structures, has yet to be elucidated. In our study, DTI scalars in the hippocampus do not predict short term outcomes of time to oral feeding, hospital length of stay, or seizure occurrence. However, our inability to detect subtle hippocampal injury limited our capacity to correlate any changes with outcomes. Further studies are required to determine the role of hippocampal injury in short term outcomes. The hippocampus is involved in complex cognitive mechanisms that may only manifest when children are challenged with higher-order learning and memory tasks at school age.⁵¹

Several studies have demonstrated hyperexcitability of hippocampal neurons following hypoxic injury, but the role in acute epileptogenesis remains unclear.^{52–55} Our study demonstrated no association between DTI scalars in the hippocampus and acute seizure occurrence. The lack of sensitivity for hippocampal injury in our study likely influenced our ability to detect an association with seizure occurrence. Interestingly, animal studies of neonatal HIE demonstrate a significant increase in hippocampal mossy fiber sprouting, which has a possible role in subsequent epileptogenesis.⁵⁶ Additional studies are needed to evaluate if DTI changes in the hippocampus are associated with seizure presence or persistence beyond the neonatal period.

Given its implications in multiple cognitive processes and memory, the hippocampus, when even subtly injured, may have significant, under recognized effects on patients affected by HIE. Hypoxic injury to the hippocampus in the perinatal period, recognized as hippocampal volume loss in school age years, has been correlated with memory deficits.^{36, 37, 57–59} However, injury was only discovered retrospectively. Recent evidence suggests that hypoxic-ischemic injury disrupts the structure and function of global hippocampal networks, resulting in impaired long-term recognition memory.⁶⁰ Identification of subtle injuries and the ability to stratify the degree of damage is therefore essential. This will facilitate the identification of prognostic markers and identify at risk populations that may benefit from additional follow-up and adjunct therapies.

Although DTI scalars allow quantification of injury, this technique is dependent on acquisition of a diagnostic MRI study. Due to clinical instability, more severely affected newborns are less likely to get an MRI study in the optimal time frame or to stay still and stable in the MRI scanner for the duration of the study. Therefore, for some severely affected neonates, MRI cannot be obtained until they are closer to the chronic phase of injury (months of age) which would not be appropriate for comparison. There is increased attention in the literature to administering TH to late preterm neonates.⁶¹ DTI values change with white matter maturation, thus the study population was restricted to term or near-term neonates.⁶² Pseudo-normalization is noted to occur at approximately 8–10 days post-injury for ADC changes.⁶³ We plotted FA values according to day of MRI and found no difference between studies occurring prior to or after ten days of life. Therefore, pseudo-normalization

did not appear to have a role in the lack of difference found between hippocampal scalars in the HIE versus control groups.

Observed injury severity has decreased following the advent of TH. Thus, the proportion of patients with moderately to severely affected imaging findings among our cohort was relatively small, likely limiting our ability to detect subtle hippocampal injury. In addition, ROI determinations are observer dependent, resulting in possible inter-observer variability. In our study, this variability was minimized by requiring agreement between two observers. The strong correlation between DTI scalars of the hippocampus and thalamus also argues for the validity of the chosen ROI and confirms the global effect of HIE on the brain. In terms of the short term outcomes, it is important to note that the admission length is dependent on multiple factors including current census and unit acuity. It does not account for time in other levels of care prior to discharge; therefore, this may be an inadequate measure of the length of necessary rehabilitation for the patient population.

Quantitative analysis of DTI is an appealing potential non-invasive marker for brain injury in the hippocampus and may be applicable to other regions of the brain as well. Given the crucial role of the hippocampus during development, hippocampal injury should be carefully examined to determine its possible prognostic significance. In our study, quantitative DTI analysis of the hippocampus is a suboptimal measurement of subacute injury. Restricting the region of interest to the fornix may provide additional insight. Analysis of this relatively homogeneous white matter tract from the hippocampus could be superior in identifying injury using DTI. Higher resolution 3T MRI scanning may help to better define the hippocampus and even smaller areas of interest such as the white matter tracts.⁶⁴ DTI white matter mapping is increasingly being used to define areas of interest and may provide a consistent and rapid assessment of this region.⁶⁵ In addition, comparison of pre- and post-hypothermia DTI studies, when feasible, would elucidate whether acute injury of the hippocampus is attenuated by hypothermia therapy.

The lack of a distinct lesion in the hippocampus does not preclude the risk of cognitive and memory impairment long term.⁶⁰ Therefore, it is essential that future studies determine the correlation of DTI measures with neurodevelopmental outcomes even without the detection of subacute injury. The thalamic involvement, as demonstrated in moderately-to-severely affected neonates, is a risk factor for neurodevelopmental impairment.³⁵ It is unclear which outcomes are most affected by co-existing injury in both areas. Additional studies should also be performed with a larger cohort of more severely affected patients to assess a potential difference between the hippocampus in HIE and control populations, with continued comparison to thalamic DTI measures.

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Figure 1. Regions of Interest Delineation, Sample positioning of regions of interest for bilateral thalamus (row A) outlined in pink and green on fractional anisotropy maps and bilateral hippocampus (row B) outlined in orange and pink on apparent diffusion coefficient maps. Axial images are ordered from superior (left) to inferior (right) on contiguous slices.

Figure 2.

Correlation of hippocampal and thalamic FA and MD values by laterality. Scatterplots demonstrate the relationship between thalamic and hippocampal DTI values on the right (red) and left (blue) sides of the brain. Quadratic regression lines are superimposed, indicating the correlation between thalamic and hippocampal FA values in (A) and MD values in (B). Spearman correlation coefficients (r values) are displayed. Analysis corrected for gestational age and age at time of MRI. FA, fractional anisotropy; MD, mean diffusivity; DTI, diffusion tensor imaging.

Table 1.

Demographic and Patient Characteristics of Neonates with HIE and Controls

Characteristic	HIE Group (n=55)	Control (n=13)
Male, n (%)	23 (42)	7 (54)
Gestational Age (weeks)	39 (36–41)	39 (36–41)
APGAR at 5 min	1 (0–9)	9 (4–9)
APGAR at 10 min	4 (3–10)	n/a
Cord pH	6.95 (6.6–7.44)	n/a
Base Deficit, mmol/L	15 (4.5–30)	n/a
Age at MRI (days)	8 (4–16)	6 (1–15)
Presence of Seizures, n (%)	25 (45)	n/a

Values denote median (range) unless otherwise indicated. n/a, not applicable; n, number of subjects; HIE, hypoxic-ischemic encephalopathy.

Table 2.

MRI Classification by Qualitative Analysis

Classification	HIE Group (n=55)	Control (n=13)
0	15 (27%)	13 (100%)
1	32 (58%)	0
2	4 (7%)	0
3	4 (7%)	0

Classification by the modified Barkovich score. HIE, hypoxic ischemic encephalopathy; n, number of subjects.

Table 3.

Differences between hippocampal DTI measures for the entire cohort of HIE patients and controls

Measure	HIE patients (n=55) Mean (95% CI)	Controls (n=13) Mean (95% CI)	P-value
Right FA	0.137 (0.127–0.147)	0.145 (0.125–0.165)	0.49
Right MD	1.227 (1.193–1.260)	1.213 (1.144–1.282)	0.73
Right AD	1.401 (1.357–1.446)	1.389 (1.296–1.481)	0.81
Right RD	1.139 (1.110–1.168)	1.125 (1.064–1.186)	0.69
Left FA	0.132 (0.124–0.141)	0.141 (0.123–0.159)	0.38
Left MD	1.221 (1.186–1.256)	1.195 (1.121–1.269)	0.53
Left AD	1.388 (1.343–1.434)	1.367 (1.272–1.461)	0.68
Left RD	1.137 (1.105–1.169)	1.109 (1.042–1.175)	0.44

DTI scalars have been adjusted for age at time of MRI and gestational age by linear regression with least squares means displayed with 95% confidence interval (CI) in parentheses. FA, fractional anisotropy; MD, mean diffusivity; AD, axial diffusivity; RD, radial diffusivity; DTI, diffusion tensor imaging; HIE, hypoxic-ischemic encephalopathy; n, number of subjects.

Table 4.

Differences between hippocampal DTI measures for moderately-to-severely affected HIE patients and controls

Measure	HIE patients (n=8) Mean (95% CI)	Controls (n=13) Mean (95% CI)	P-value
Right FA	0.125 (0.101–0.148)	0.144 (0.125–0.162)	0.20
Right MD	1.243 (1.145–1.341)	1.204 (1.127–1.280)	0.52
Right AD	1.406 (1.283–1.529)	1.377 (1.281–1.473)	0.70
Right RD	1.161 (1.073–1.250)	1.117 (1.048–1.186)	0.42
Left FA	0.120 (0.100–0.140)	0.141 (0.126–0.157)	0.09
Left MD	1.252 (1.161–1.343)	1.185 (1.114–1.256)	0.24
Left AD	1.406 (1.297–1.515)	1.356 (1.271–1.441)	0.46
Left RD	1.175 (1.090–1.259)	1.010 (1.033–1.166)	0.16

DTI scalars have been adjusted for age at time of MRI and gestational age by linear regression with least squares means displayed with 95% confidence interval (CI) in parentheses. FA, fractional anisotropy; MD, mean diffusivity; AD, axial diffusivity; RD, radial diffusivity; DTI, diffusion tensor imaging; HIE, hypoxic-ischemic encephalopathy; n, number of subjects.

Table 5.

Differences between thalamic DTI measures for the entire cohort of HIE patients and controls

Measure	HIE patients (n=55) Mean (95% CI)	Controls (n=13) Mean (95% CI)	P-value
Right FA	0.164 (0.158–0.171)	0.182 (0.169–0.195)	0.02 [*]
Right MD	1.067 (1.040–1.094)	1.061 (1.005–1.118)	0.86
Right AD	1.234 (1.204–1.264)	1.246 (1.182–1.309)	0.75
Right RD	0.983 (0.960–1.009)	0.969 (0.914–1.023)	0.64
Left FA	0.166 (0.159–0.173)	0.181 (0.166–0.196)	0.08
Left MD	1.061 (1.034–1.087)	1.059 (1.003–1.114)	0.94
Left AD	1.230 (1.200–1.261)	1.245 (1.182–1.308)	0.68
Left RD	0.976 (0.950–1.002)	0.965 (0.912–1.019)	0.72

DTI scalars have been adjusted for age at time of MRI and gestational age by linear regression with least squares means displayed with 95% confidence interval (CI) in parentheses. FA, fractional anisotropy; MD, mean diffusivity; AD, axial diffusivity; RD, radial diffusivity; DTI, diffusion tensor imaging; HIE, hypoxic-ischemic encephalopathy; n, number of subjects

^{*} denotes statistical significance with $p < 0.05$.

Table 6.

Differences between thalamic DTI measures for moderately-to-severely affected HIE patients and controls

Measure	HIE patients (n=8) Mean (95% CI)	Controls (n=13) Mean (95% CI)	P-value
Right FA	0.148 (0.127–0.168)	0.182 (0.166–0.198)	0.01 *
Right MD	1.118 (1.023–1.213)	1.055 (0.981–1.130)	0.29
Right AD	1.275 (1.172–1.379)	1.239 (1.158–1.320)	0.57
Right RD	1.039 (0.946–1.132)	0.963 (0.891–1.036)	0.20
Left FA	0.147 (0.124–0.170)	0.181 (0.163–0.199)	0.03 *
Left MD	1.112 (1.018–1.205)	1.053 (0.980–1.126)	0.31
Left AD	1.270 (1.165–1.374)	1.238 (1.156–1.320)	0.63
Left RD	1.033 (0.942–1.123)	0.960 (0.890–1.031)	0.20

DTI scalars have been adjusted for age at time of MRI and gestational age by linear regression with least squares means displayed with 95% confidence interval (CI) in parentheses. FA, fractional anisotropy; MD, mean diffusivity; AD, axial diffusivity; RD, radial diffusivity; DTI, diffusion tensor imaging; HIE, hypoxic-ischemic encephalopathy; n, number of subjects

* denotes statistical significance with $p < 0.05$.

Table 7.

Association of DTI scalars in the hippocampus with short term outcomes.

DTI Scalar ^a	Outcome					
	Length of Hospital Stay		Time to Oral Intake		Presence of Seizures	
	Hazard ratio ^b (95% CI)	p-value	Hazard ratio ^b (95% CI)	p-value	Odds ratio ^c (95% CI)	p-value
Right FA	0.97 (0.91–1.04)	0.36	1.00 (0.93–1.07)	0.98	1.01 (0.87–1.17)	0.88
Right MD	1.00 (0.98–1.02)	0.93	1.00 (0.98–1.03)	0.73	1.03 (0.98–1.08)	0.24
Left FA	0.96 (0.89–1.04)	0.36	1.00 (0.92–1.09)	0.92	0.96 (0.82–1.13)	0.64
Left MD	1.00 (0.98–1.02)	0.91	1.00 (0.98–1.02)	0.83	1.04 (0.99–1.09)	0.11

^a Each DTI scalar was analyzed in a separate model. All models were adjusted for gestational age and age at time of MRI.

^b Hazard ratio for a 0.01 unit increase in the DTI scalar measure.

^c Odds ratio for a 0.01 unit increase in the DTI scalar measure.

DTI, diffusion tensor imaging; FA, fractional anisotropy; MD, mean diffusivity; CI, confidence interval.