The association between cardiac physiology, acquired brain injury, and postnatal brain growth in critical congenital heart disease

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ABSTRACT

Objective: To assess the trajectory of perioperative brain growth in relationship to cardiac diagnosis and acquired brain injuries.

Methods: This was a cohort study of term neonates with hypoplastic left heart syndrome (HLHS) and d-transposition of the great arteries (d-TGA). Subjects underwent magnetic resonance imaging of the brain pre- and postoperatively to determine the severity of brain injury and total and regional brain volumes by the use of automated morphometry. Comparisons were made by cardiac lesion and injury status.

Results: A total of 79 subjects were included (49, d-TGA; 30, HLHS). Subjects with HLHS had more postoperative brain injury (55.6% vs 30.4%, $P = .03$) and more severe brain injury (moderate-to-severe white matter [WM] injury, $P = .01$). Total and regional perioperative brain growth was not different by brain injury status (either pre- or postoperative). However, subjects with moderate-to-severe WM injury had a slower rate of brain growth in WM and gray matter compared with those with no injury. Subjects with HLHS had a slower rate of growth globally and in WM and deep gray matter as compared with d-TGA (total brain volume: 12 cm³/wk vs 7 cm³; WM: 2.1 cm³/wk vs 0.6 cm³; deep gray matter: 1.5 cm³/wk vs 0.7 cm³; $P < .001$), after we adjusted for gestational age at scan and the presence of brain injury. This difference remained after excluding subjects with moderate-to-severe WM injury.

Conclusions: Neonates with HLHS have a slower rate of global and regional brain growth compared with d-TGA, likely related to inherent physiologic differences postoperatively. These findings demonstrate the complex interplay between cardiac lesion, brain injury, and brain growth. (J Thorac Cardiovasc Surg 2018;155:291-300)

T1-weighted magnetic resonance imaging. A, Automatic segmentation of brain structures to measure (B) total brain; C, Gray matter; D, White matter; and (E) deep gray matter volume.

Central Message

Neonates with hypoplastic left heart syndrome have a slower rate of perioperative brain growth than d-transposition of the great arteries. Brain injury has less influence on neonatal brain growth.

Perspective

Brain injury and delayed brain development are common in neonates with hypoplastic left heart syndrome and d-transposition of the great arteries. Despite similarities in brain health preoperatively, neonates with hypoplastic left heart syndrome have a slower rate of ongoing brain growth in the perioperative time period, a reflection of the postoperative physiologic differences between these 2 cardiac lesions.

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Abbreviations and Acronyms

BIS	= brain injury severity
CHD	= congenital heart disease
GA	= gestational age
GM	= gray matter
HLHS	= hypoplastic left heart syndrome
IVH	= intraventricular hemorrhage
IQR	= interquartile range
MRI	= magnetic resonance imaging
TBV	= total brain volume
d-TGA	= d-transposition of the great arteries
WM	= white matter
WMI	= white matter injury

Scanning this QR code will take you to supplemental figures and appendix for this article.

Advances in surgical techniques and perioperative care have led to improved survival of newborns with critical congenital heart disease (CHD), such as hypoplastic left heart syndrome (HLHS) and d-transposition of the great arteries (d-TGA).¹ Although there has been a decline in gross neurologic insults in these children, many experience behavioral, emotional, cognitive, and motor impairments, suggesting widespread brain dysfunction.^{2,3} Studies that have assessed neonates with d-TGA and HLHS as a single cohort have identified delayed brain development and a similar prevalence of preoperative brain injury despite the fact that these lesions are anatomically and physiologically distinct from one another.⁴⁻⁷ Interestingly, both lesions have evidence of altered fetal cerebral oxygen delivery and smaller total brain volumes (TBVs) as compared with the normal fetus, although the underlying mechanisms may differ in each lesion.⁸⁻¹⁰

Measurements of simple metrics of brain growth on magnetic resonance imaging (MRI) have identified smaller total and regional brain size at birth and infancy in CHD compared with controls^{11,12}; however, the rate of brain growth was similar and did not differ by cardiac lesion.¹¹ Cerebral MRI volumetry is a quantitative measure of total and regional brain volume providing an accurate means to assess brain volume and growth trajectory.¹³⁻¹⁵ Sophisticated automated volumetry techniques can model brain tissue characteristics during neonatal development and have identified decreased total and regional brain volumes in neonates with complex CHD previous to any corrective operation.¹⁶ In particular, studies have shown

associations between smaller total and regional brain volumes (white matter [WM] and cortical gray matter [GM]) with neurodevelopmental outcome in adolescents with CHD,¹⁷ as well as association between brain volume at birth and neonatal neurobehavior.¹⁸ These studies highlight the potential usefulness of this quantitative measure in identifying the patients at greatest risk for neurodevelopmental impairment.

We sought to assess the trajectory of perioperative brain growth in relationship to cardiac diagnosis and acquired brain injuries by using magnetic resonance morphometry in a large sample of well-characterized patients with standardized imaging time points in the neonatal period. We hypothesize that patients with HLHS exhibit slower rates of brain growth due to a palliative, staged surgical approach with persistent cyanosis and potential for ongoing postoperative brain injury. In contrast, newborns with d-TGA undergo definitive surgical correction with restoration of normal brain oxygen delivery, providing more favorable conditions for brain growth.

METHODS

Between 2001 and 2014, newborns with critical CHD at the University of California San Francisco Benioff Children's Hospital were invited consecutively to participate in a prospective protocol studying brain development and brain injury in CHD using MRI. Brain imaging findings from earlier versions of this cohort were reported previously.^{4,5} Patients who were born before 36 weeks' gestation, had a suspected congenital infection, had clinical evidence of a congenital malformation or syndrome, and/or had a suspected or confirmed genetic or chromosomal anomaly were excluded. Once written informed consent was received, patients underwent brain MRI before and after cardiac surgery. The institutional committee on human research approved the study protocol. This study was approved by the UCSF IRB study number 10-03749, original approval date October 2001.

Patients diagnosed as having d-TGA with or without a ventricular septal defect or HLHS were included in this current study. HLHS was defined as the presence of 1 functioning right ventricle with varying degrees of severe left heart hypoplasia requiring a palliative surgical intervention for survival (ie, Stage I operation) in the newborn period. All subjects had a Stage I (Norwood) operation with a RV-PA conduit (Sano) modification.

MRI Study

Preoperative MRI studies were performed as soon as the baby could be safely transported to the MRI scanner as determined by the clinical team. Postoperative studies were performed after completion of perioperative care and before discharge from the hospital. Imaging time points were separated by an average of 15 days in the entire cohort. Detailed MRI methods are listed in [Appendix E1](#). A neuroradiologist (A.J.B.) reviewed each MRI. Brain injury was characterized as stroke, white matter injury (WMI), intraventricular hemorrhage (IVH), and/or global hypoxic ischemic injury as previously described.^{5,6} Postoperative brain injuries are limited to newly acquired lesions not evident on the preoperative scan. WMI was classified as mild (1-3 foci each <2 mm), moderate (>3 foci or any foci >2 mm), or severe (>5% of white matter volume).⁵ IVH was characterized as grade I, II, III, or IV via the system of Papile and colleagues.¹⁹ In addition, brain injury severity (BIS) was determined for each patient as previously described.²⁰ The BIS was assigned as follows: 0 indicates normal (no injury); 1, minimal injury (minimal WMI

FIGURE 1. T1-weighted magnetic resonance imaging. A, Automatic segmentation of brain structures to measure (B) total brain; C, Gray matter; D, White matter; and (E) deep gray matter volumes.

and IVH grade I or II); 2, stroke (all stroke); and 3, moderate-to-severe injury (moderate and severe WMI, IVH grade III, or global hypoxic-ischemic injury).

Morphometry

With an automated approach, segmentation of the GM, WM, and deep GM structure (combining the thalamus, the basal ganglia, and the periventricular germinal zone) was performed, as previously described (Figure 1).²¹ Detailed methods are listed in Appendix E1.

Clinical Variables

Clinical data were collected prospectively from the medical records by a team of trained neonatal research nurses and reviewed by a pediatric intensivist (P.M.) who was blinded to all neuroimaging findings.

Statistical Analysis

Demographic characteristics, descriptors of brain injury, and clinical variables were compared between patients with HLHS and d-TGA by the use of standard descriptive statistics. To analyze brain structural volumes, we used general linear models that included gestational age (GA) at scan, type of cardiac lesion (d-TGA or HLHS), BIS, and sex as dependent variables, whereas TBV, GM, WM, or deep GM volumes were set as the independent variables. As most subjects were scanned twice (one before and one after the surgery), mixed-effect linear models were used to take into account multiple measurements per subject. To assess whether preoperative or postoperative brain injury influenced brain volumes, all subjects were first dichotomized into those with a cumulative BIS score > 0 and those with a BIS score $= 0$ based on the maximum value between preoperative and postoperative BIS scores.

In a separate analysis, an interaction term was included as $BIS \times GA$ at scan into the linear model to assess differences in brain growth rate depending on the presence of brain injury.

Finally, to assess difference by cardiac lesion, a linear model that contained a group term of cardiac lesion type (d-TGA or HLHS) and GA at scan as the main effect term was performed, including an interaction term (cardiac lesion $\times$ GA at scan). Adjustments were made for the presence of brain injury. The Bonferroni adjustment for multiple comparisons was used to obtain corrected P values, which after adjusting for 8 comparisons was $P = .006$.

RESULTS

A total of 79 infants were included in this study, 49 with d-TGA and 30 with HLHS. There was a total of 79 preoperative brain MRIs and 73 postoperative brain MRIs. The postoperative MRI was not analyzed in 5 subjects due to motion degradation. One subject did not have a postoperative MRI due to pacemaker placement. Demographics and clinical variables are listed in Tables 1 and 2. Both groups were similar in preoperative clinical characteristics including birth weight and birth head circumference. Postoperatively, subjects with HLHS had more frequent cardiac arrest events and a longer length of hospital stay compared with subjects with d-TGA. In addition, the postoperative MRI was performed on average 6.5 days later in the HLHS group (day-of-life MRI: median

TABLE 1. Demographics by cardiac lesion

	d-TGA (n = 49)	HLHS (n = 30)	P value
EGA delivery, wk (mean, 95% CI)	39.2 (38.8-39.6)	38.9 (38.4-39.3)	.35
Birth weight, g (mean, 95% CI)	3396.8 (3228.0-3565.6)	3211.9 (3030.6-3393.3)	.16
Male, n (%)	39 (79.6)	16 (53.3)	.02
Birth head circumference, cm (mean, 95% CI)	34.0 (33.7-34.3)	34.1 (33.6-34.6)	.78
Weight at MR2, g (mean, 95% CI)	3492.9 (3364.6-3621.4)	3308.5 (3183.1-3433.8)	.06

d-TGA, d-Transposition of the great arteries; HLHS, hypoplastic left heart syndrome; EGA, estimated gestational age; CI, confidence interval; MR2, postoperative magnetic resonance imaging.

24 days, interquartile range [IQR]: 20-30) as compared with the d-TGA group (day-of-life MRI: median 17.5 days, IQR: 15-25, $P = .001$). The average time between scans was 15 days (IQR 10-21) in the entire cohort, 12.5 days in d-TGA (IQR 9-17), and 19 days in HLHS (IQR 15-25). Weight at the time of the second MRI was similar in both groups (d-TGA: 3492.9 g, 95% confidence interval, 3364.6-3621.4; HLHS: 3308.5 g; 95% confidence interval, 3183.1-3433.8; $P = .06$) with a similar increase in weight from birth to the time of the second MRI in both groups (d-TGA: 96.1 g; HLHS: 96.6 g) (Table 1).

The frequency of preoperative brain injury was greatest in subjects with HLHS but not statistically significant (d-TGA = 14.3%, HLHS = 30%, $P = .09$). In contrast, subjects with HLHS had a greater prevalence of new postoperative injury (d-TGA = 30.4%, HLHS = 55.6%, $P = .03$), mostly in the form of stroke (Table 3). The cumulative BIS score, which represents the greatest score on either MRI, is represented in Figure 2. Subjects with HLHS had greater cumulative BIS scores than subjects with d-TGA (BIS = 2 and 3; $P = .01$).

Morphometry

Total and regional brain volumes increased significantly with increasing age in the entire cohort, even when we

accounted for the presence of brain injury, sex, and the type of cardiac lesion ($P < .0001$ for all regions). Subjects were dichotomized by cumulative BIS score (BIS = 0 vs BIS > 0). Overall, mean global and regional brain volumes (all pre- and postoperative data points combined) were greater in subjects without brain injury compared with those with brain injury, except in WM (TBV: 385 cm³ vs 359 cm³, $P < .0001$; GM: 147 cm³ vs 139 cm³, $P < .0001$; deep GM: 48 cm³ vs 46 cm³, $P < .004$; WM: 121 cm³ vs 116 cm³, $P = .007$). Although preoperative brain injury did not predict preoperative brain volumes, it was associated with significantly lower GM volumes on the postoperative MRI (147 cm³ vs 157 cm³, $P = .0001$). Similarly, postoperative brain injury was associated with lower GM volumes on the postoperative MRI (149 cm³ vs 159 cm³, $P = .0001$). In contrast, preoperative brain volumes were not associated with new postoperative brain injury ($P = .2$). Despite the difference in brain volumes at single points in time, the rate of global and regional brain growth did not correlate with the presence of brain injury noted on the pre- or postoperative MRI (rate of growth in those with no brain injury [cumulative BIS = 0] vs those with brain injury [cumulative BIS > 0]; TBV: 12.3 cm³ vs 12.4 cm³, $P > .3$; GM: 8.15 cm³ vs 8.14 cm³, $P > .5$; WM: 5.52 cm³ vs 5.46 cm³, $P > .4$; deep GM: 1.21 cm³ vs 1.18 cm³,

TABLE 2. Pre- and postoperative clinical variables by cardiac lesion

	d-TGA (n = 49)	HLHS (n = 30)	P value
Balloon atrial septostomy, n (%)	30 (61.2)	1 (3.3)	<.001
Lowest preoperative O ₂ sat, mean (95% CI)	56.9 (52.0-61.7)	80.1 (76.2-84.0)	<.001
Preoperative pH (first ABG), mean (95% CI)	7.27 (7.23-7.31)	7.29 (7.24-7.35)	.43
Preop base excess (first ABG), mean (95% CI)	-6.2 (-8.5 to -3.9)	-4.1 (-6.7 to -1.4)	.23
Preoperative cardiac arrest, n (%)	1 (2.0)	2 (6.7)	.55
Day-of-life operation median (IQR)	8 (5.5-11)	8 (6-11)	.99
Day-of-life MRI 1, median (IQR)	5 (3-6)	5 (3-6)	.96
Postoperative cardiac arrest, n (%)	0	5 (16.7)	.006
Postoperative ECLS, n (%)	2 (4.1)	4 (13.3)	.19
Postoperative duration of mechanical ventilation, median (IQR)	5 (3-7)	8 (6-10)	.0002
Day-of-life MRI 2, median (IQR)	17.5 (15-25)	24 (20-30)	.001
Hospital LOS, mean (95% CI)	23.8 (19.2-28.3)	40 (30.0-50.0)	.001

d-TGA, d-Transposition of the great arteries; HLHS, hypoplastic left heart syndrome; O₂ sat, oxygen saturation; CI, confidence interval; ABG, arterial blood gas; IQR, interquartile range; MRI 1, preoperative magnetic resonance imaging; ECLS, extracorporeal life support; MRI 2, postoperative magnetic resonance imaging; LOS, length of stay.

TABLE 3. Pre- and postoperative brain injury by cardiac lesion

	d-TGA (n = 49)	HLHS (n = 30)	P value
Preoperative			
Any injury	17/49 (34.7%)	13/30 (43.3%)	.44
WMI	7/49 (14.3%)	9/30 (30.0%)	.09
Stroke	12/49 (24.5%)	6/30 (20.0%)	.64
New postoperative*			
Any injury	14/46 (30.4%)	15/27 (55.6%)	.03
WMI	13/46 (28.3%)	9/27 (33.3%)	.65
Stroke	1/46 (2.2%)	7/27 (25.9%)	.003

d-TGA, d-Transposition of the great arteries; HLHS, hypoplastic left heart syndrome; WMI, white matter injury. *Postoperative magnetic resonance missing in 3 subjects with d-TGA and 3 subjects with HLHS due to death of subject.

$P > .3$; Figure 3, A, Figure E1, A). However, when we only evaluated patients with the most severe form of brain injury (moderate-to-severe WMI, cumulative BIS = 3), the rate of growth in WM and GM was significantly slower compared with those with no brain injury (GM: 6.7 cm³/wk vs 8.1 cm³/wk, $P = .001$; WM: 1.2 cm³/wk vs 1.8 cm³/wk, $P < .001$; Figure 3, B, Figure E1, B).

When we assessed differences by cardiac lesion, there was no difference in overall mean total and regional measures of brain volume (all pre- and postoperative data points combined and when assessing each time point separately) in subjects with d-TGA compared with HLHS (TBV: 375 cm³ vs 372 cm³, $P > .2$; GM: 142 cm³ vs

145 cm³, $P > .1$; WM: 120 cm³ vs 117 cm³, $P = .1$; deep GM: 47 cm³ vs 46 cm³, $P > .1$). However, subjects with d-TGA had a faster rate of increase in global and regional brain growth compared with subjects with HLHS, even when we adjusted for GA at scan and the presence of any injury (TBV: 12 cm³/wk vs 7 cm³/wk; WM volume: 2.1 cm³/wk vs 0.6 cm³/wk; deep GM volume: 1.5 cm³/wk vs 0.7 cm³/wk; $P < .001$; Figure 4, Figure E2). GM did not show a significant difference in growth by cardiac lesion ($P = .2$). Even when we excluded subjects with moderate-to-severe WMI (BIS = 3), those with d-TGA had a faster rate of increase in global and regional brain growth compared with subjects with HLHS ($P < .001$ for TBV, deep GM, and WM).

DISCUSSION

Our results demonstrate better perioperative brain growth in patients with d-TGA compared with HLHS. Specifically, there was a 40% increase in the rate of brain growth in WM, GM, and TBV in subjects with d-TGA in this short period from preoperative to postoperative imaging. Interestingly, although subjects with brain injury had overall smaller brain volumes at individual points in time compared with those without injury, the strongest predictor of brain growth trajectory was cardiac lesion. This is the first report, to our knowledge, directly comparing the perioperative patterns of brain growth between 2 infant groups with common, yet

FIGURE 2. Distribution of the cumulative BIS score by cardiac lesion. The cumulative BIS score is the worst score when combining the pre- and postoperative magnetic resonance imaging. A test for trends demonstrates less-severe cumulative BIS scores in the d-TGA group ($P = .01$). d-TGA, d-transposition of the great arteries; HLHS, hypoplastic left heart syndrome; BIS, brain injury severity.

Comparison between babies with BIS=0 and those with BIS=1-3

A

Comparison between babies with BIS=0 and those with BIS=3

B

FIGURE 3. A, Brain global/regional volume changes in the presence of brain injury (red line, cumulative BIS > 0) and without brain injury (blue line, cumulative BIS = 0). The x-axis represents the gestational age at the time of MRI, and the y-axis represents the volume in mm³. The plot includes pre- and postoperative brain MRI measures for each subject with a best-fitted line. Plots with paired observations for each subject are included in Figure E1. Overall, mean global and regional (gray matter and deep gray matter) brain volumes were lower in subjects with brain injury compared with those without brain injury. However, the rate of global and regional volume change over time was not associated with brain injury (all P values > .1). B, Brain regional volume changes in gray and white matter in the presence of no brain injury (blue line, cumulative BIS = 0) and in those with moderate-to-severe white matter injury (dark red line, cumulative BIS = 3). Those with moderate-to-severe white matter injury have significantly slower growth in gray matter and white matter as compared with those without injury ($P = .001$ and $P < .0001$, respectively). BIS, Brain injury severity.

physiologically distinct critical congenital cardiac defects using advanced magnetic resonance morphometry.

In the fetal time period, both d-TGA and HLHS fetuses have evidence of delayed brain development (TBV and

metabolic brain development),⁹ although the underlying mechanisms likely differ by cardiac physiology and anatomy. In particular, based on fetal lamb models, those with HLHS have abnormalities in both perfusion (due to

FIGURE 4. The rate of change in global and regional brain volumes by cardiac lesion after adjusting for the presence of brain injury. The x-axis represents the gestational age at the time of MRI, and the y-axis represents the volume in mm³. The plots include pre- and postoperative brain MRI measures for each subject with a best-fitted line. Plots with paired observations for each subject are included in Figure E2. Subjects with HLHS (purple line) have a slower rate of growth in total brain volume, white matter, and deep gray matter as compared with those with d-TGA (orange line). *Significant after Bonferroni correction, $P < .006$.

retrograde flow from the ductus arteriosus) and oxygenation of cerebral blood flow. In contrast, those with d-TGA are more likely to have abnormalities in oxygenation of cerebral blood flow.¹⁰ Ultimately, both lesions result in decreased cerebral oxygen delivery and consumption.⁸ Recently, Rudolph²² suggested that decreased substrate delivery to the brain such as glucose may be the primary cause for the abnormalities in brain development. Most studies find no differences by cardiac lesion in the degree of fetal or neonatal preoperative brain development.^{4,7} Some evidence suggests that within a particular group, specific anatomic features such as aortic atresia in HLHS lead to the greatest degree of delayed microstructural brain development.²³ Our study did not demonstrate significant differences preoperatively in measures of brain volumes when we compared HLHS with d-TGA. This demonstrates that although these lesions are physiologically different, brain growth is similar during fetal and up to the preoperative time period.

After birth and before their neonatal operation, patients with both d-TGA and HLHS are at risk for brain injury in the form of WMI and stroke.^{5,24,25} Indeed, both groups have a similar risk of brain injury, likely secondary to shared risk factors such as hypoxia, length of time to surgery, embolism, and preoperative cardiac arrest.²⁶⁻²⁹ New, postoperative brain injury is common and tends to be more prevalent in subjects with single-ventricle physiology.⁵ Similarly, we found that postoperative brain injury was more common in the HLHS group, with a greater number of small focal strokes, consistent with embolism. As opposed to patients with d-TGA, patients with HLHS after a Norwood procedure are vulnerable to embolic injury, which may be modifiable based on postoperative management and hospital length of stay.

The relationship between brain development and injury is complex and dependent on methods used to measure brain development. Several studies agree that brain immaturity, measured by the use of semiquantitative methods, is a risk

factor for preoperative brain injury.^{20,28,30} However, our measures of total and regional brain volumes preoperatively did not seem to predict the presence of new postoperative brain injury. The impact of brain injury on measures of ongoing brain development is less clear. For example, our group has shown that preoperative brain injury predicts delayed postoperative metabolic and microstructural brain development.²⁰ We found that preoperative brain injury is associated with smaller postoperative GM volumes and that overall brain volumes were smaller in those with injury compared with those without injury. However, we found that rate of brain growth did not differ by the presence of brain injury globally or regionally. Thus, although brain injury is associated with brain volume loss, there appears to be minimal influence on continued brain growth, at least in the perioperative time period. On the most severe end of the spectrum, subjects with moderate-to-severe WMI exhibit a slower rate of growth in WM and GM. We believe this result is driven by the complex interplay between cardiac lesion, brain injury, and brain growth. In our cohort, a larger percentage of subjects with HLHS had moderate-to-severe WMI, suggesting that cardiac lesion is the initial risk factor that influences both injury and brain growth. In fact, when subjects with moderate-to-severe WMI were removed from the analysis, those with d-TGA continued to have an even faster rate of global and regional brain growth as compared with HLHS.

Studies that have analyzed pyramidal tract maturation in newborns with CHD have demonstrated less rapid changes in fractional anisotropy over the perioperative time period in subjects with preoperative brain injury,³¹ similar to what is seen in premature infants with brain injury.³² This suggests that the effects of brain injury on brain development may differ at macroscopic and microscopic scales. The techniques used to measure these different aspects of brain development are unique and offer different perspectives on brain maturity. More sensitive measures of brain growth in regions corresponding to the location of acquired focal lesions may lead to a deeper understanding of the longer term effects of milder forms of brain injury on brain growth.

The relationship between brain volumes in particular and injury is inherently complicated. Brain injury may lead to swelling and increased volume acutely, followed by tissue loss, potential repair, and finally, in the immature brain, variable subsequent growth. Brain growth is one component of several factors that may impact long-term neurologic outcomes in patients with complex CHD. The experience with preterm infants has demonstrated the close interplay between injury and continued brain growth and their collective influence on outcomes. Specifically, studies in premature infants have demonstrated long-term effects of WMI or periventricular leukomalacia on

brain growth and function.³³ Although periventricular leukomalacia is localized to the WM, it can lead to disruption of cortical activity³⁴ and GM hypoplasia,³⁵ with important consequences for developing cortical circuits.³⁶⁻³⁹ Neuropathologic studies in nonsurvivors of critical CHD have shown abnormalities in the thalamus with thalamic neuronal loss and gliosis,⁴⁰ regions critical for working memory and attention, a common deficit noted in survivors of critical CHD. Our data demonstrate that moderate-to-severe WMI does appear to slow the rate of brain growth in WM and GM. Although this is likely largely driven by cardiac lesion, the impact of injury on brain growth and long-term neurodevelopmental outcomes warrants further study.

Our findings demonstrate a clear link between cardiac subgroup and ongoing perioperative brain growth. Given the high burden of acquired injury and delayed brain development early in life, the potential for repair and optimal brain growth is critical. Although patients with HLHS and d-TGA appear to share similar fetal and preoperative risk factors, they differ significantly postoperatively. Subjects with d-TGA undergo a corrective operation (arterial switch operation) that restores normal cardiovascular physiology, whereas those with HLHS undergo a series of palliative operations, never achieving normal circulation, with ongoing hypoxia and risk for diminished systemic perfusion. Patients with single-ventricle physiology rely on circulation in series that may result in diminished systemic perfusion with compromised overall cerebral blood flow. There are several other differences between these groups, including operative strategies (low- or full-flow bypass only vs circulatory arrest or regional cerebral perfusion) and postoperative management (length of stay, duration of mechanical ventilation) that influence our findings. Finally, somatic growth likely plays a role in brain development, particularly for those with single-ventricle physiology. However, in the short time period evaluated in our study, both groups had a similar rate of weight gain in the perioperative time period. In line with the cardiac physiology influences on brain development in CHD, we recently reported a relationship between the benefits of prenatal diagnosis and a healthier preoperative state with perioperative brain development.⁴¹

Ibuki and colleagues⁴² demonstrated normalization of total and frontal brain volumes at 3 years of age in subjects with d-TGA but not in single-ventricle patients. Their data suggested an association between hypoxia (oxygen saturation) and measures of neurodevelopmental outcome (psychomotor development index on the Bayley Scales of Infant and Toddler Development, Second Edition) in the single-ventricle group. Their study was limited due to a small sample size, considerable variability, and long intervals between imaging studies. In our study, the larger sample size, standardized imaging timepoints in the

neonatal perioperative period, and the short interval between MRI studies strengthens our conclusions and suggests that the differences in brain growth by cardiac lesion begin soon after the neonatal operation.

Our findings suggest improved perioperative brain growth in subjects with d-TGA; however, evidence suggests that these patients continue to have structural and functional neurologic abnormalities later in life. Adolescents with d-TGA have altered GM volume and thickness⁴³ as well as diminished WM microstructure^{17,44,45} compared with healthy controls. Neurodevelopmental assessments have revealed ongoing impairments in executive skills, visual-spatial skills, and the need for remedial services,⁴⁶ suggesting that abnormal fetal and preoperative physiology and acquired brain injury may have lasting effects. Indeed, patients with HLHS demonstrate the greatest deficits and neurodevelopmental morbidity.^{2,47-49}

Our findings are limited by not knowing the exact timing of acquired brain injury despite 2 imaging time points in the perioperative time period. Although we are invoking developmental etiologies (lack of oxygen and substrate delivery) as the primary influence on neonatal brain growth, the impact of injury as a destructive etiology on brain growth requires a larger sample size and more imaging timepoints, particularly in the transitional period after birth.

In conclusion, brain growth in the perioperative time period is influenced more by cardiac subgroup than by brain injury. Subjects with HLHS have a slower rate of perioperative brain growth globally and regionally in WM and deep GM. Further studies are needed to determine the predictive abilities of this measure of brain development on long-term neurodevelopmental outcomes, with a goal of identifying specific impairments as targets for early intervention.

Conflicts of Interest Statement

Authors have nothing to disclose with regard to commercial support.

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Key Words: congenital heart disease, neurodevelopment, brain injury, brain development, physiology

APPENDIX E1. DETAILED MAGNETIC RESONANCE IMAGING (MRI) METHODS

MRI studies were performed with pharmacologic sedation, as needed, on a 1.5- or 3-Tesla system (GE Healthcare Signa Echo-speed; Waukesha, Wis) with GE EXCITE 1.5T/3T software and included 4-mm-thickness T1-weighted sagittal and axial spin echo, 4-mm-thickness dual-echo T2-weighted spin echo, 1.5-mm-thickness coronal or sagittal volumetric 3-dimensional gradient echo with radiofrequency spoiling images, and a diffusion tensor sequence (repetition time: 7000 milliseconds; echo time: 99.5 milliseconds; 3-mm section thickness; no gap; 3 repetitions per image, with 18×36 cm field of view and 128×256 acquisition matrix), acquiring axial images through the whole brain with an in-plane resolution of 1.4×1.4 mm². Seven images per axial section were acquired, including a T2-weighted reference image ($b = 0$ s/mm²) and 6-15 diffusion-weighted images ($b = 700$ s/mm²) in noncollinear gradient directions.

MORPHOMETRY

T1-weighted MRI underwent intensity nonuniformity correction with N3.^{E1} These images were registered linearly to the spatiotemporal neonatal brain template from the Biomedical Image Analysis Group, Imperial College London (<http://biomedic.doc.ic.ac.uk/brain-development/index.php?n=Main.Neonatal2>). The individual images were first registered to the closest age template and further registered to the oldest brain template using the transformation matrix of the registration between the selected age template and the oldest template. We segmented the cerebrum and cerebellum using a patch-based brain extraction algorithm,^{E2} which subsequently were used to calculate the total brain volume once each mask was registered back to the native space where the MRI was acquired using the previously obtained transformation

matrix. MRI intensities were recorrected with N3 and normalized within the brain mask. The gray matter (GM), white matter, and cerebrospinal fluid were classified within the mask via a recently developed method^{E3} that integrated the nonlocal mean estimation of multiatlas texture patches with an advanced label fusion based on a joint probability between selected templates.^{E4} To overcome poor segmentation due to varying tissue contrast and varying cortical folding pattern during the postnatal period as well as heavy motion artifact, we constructed a patch library that contains individual atlases (image + manual segmentation; $n = 35$) scanned at various ages and under variable conditions of motion. The same segmentation method was used to segment deep GM and ventricles independently. Finally, we measured volumes of GM, white matter, and deep GM by summing the voxel-wise partial volume measurement for each structure, which was given as the posterior probability map resulting from the segmentation algorithm.

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Comparison between babies with BIS=0 and those with BIS=1-3

A

Comparison between babies with BIS=0 and those with BIS=3

B

FIGURE E1. A, Brain global/regional volume changes in the presence of brain injury (red line, cumulative BIS > 0) and without brain injury (blue line, cumulative BIS = 0). The x-axis represents the gestational age at the time of MRI, and the y-axis represents the volume in mm³. The plot includes pre- and postoperative brain MRI measures with paired observations for each subject (gray line) and a best-fit line. The rate of global and regional volume change over time was not associated with brain injury (all P values > .1). B, Brain regional volume changes in gray and white matter in the presence of no brain injury (blue line, cumulative BIS = 0) and in those with moderate-to-severe white matter injury (dark red line, cumulative BIS = 3). Those with moderate-to-severe WMI have significantly slower growth in gray matter and white matter compared with those without injury ($P = .001$ and $P < .00001$, respectively). BIS, Brain injury severity.

FIGURE E2. The rate of change in global and regional brain volumes by cardiac lesion after adjusting for the presence of brain injury. The x-axis represents the gestational age at the time of MRI, and the y-axis represents the volume in mm^3 . The plots include pre- and postoperative brain MRI measures with paired observations for each subject (*gray line*) and a best-fit line. Subjects with HLHS (*purple line*) have a slower rate of growth in total brain volume, white matter, and deep gray matter compared to those with d-TGA (*orange line*). *Significant after Bonferroni correction, $P < .006$.