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Late failure of cerebral autoregulation in hypoxic-ischemic encephalopathy is associated with brain injury: A pilot study

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Abstract

Background: Post-resuscitation reperfusion following hypoxic-ischemia (HIE) is associated with secondary brain injury in neonates.

Objective: To quantify the association between perfusion exceeding autoregulatory limits and brain injury.

Methods: Continuous mean arterial blood pressure (MABP) and cerebral near-infrared spectroscopy (NIRS) data were prospectively collected from infants with HIE. Cerebral oximetry index (COx) was calculated as a moving correlation coefficient between MABP and NIRS. Upper and lower limits of autoregulation were identified by transition from negative to positive correlation. The proportion of time MABP was above (hyperperfusion) and below (hypoperfusion) autoregulatory limits was calculated during therapeutic hypothermia (days 1–3).

Results: Sixteen infants were included; injury was noted in 7/16. There was no significance in hyperperfusion burden between injured and uninjured infants during day one, (7% vs. 10%, $p=0.88$) or two (4% vs. 2%, $p=0.88$), but there was a marked increase for injured infants on day three (54% vs 14%, $p=0.02$). There was a corollary decrease in hypoperfusion for injured vs. uninjured infants on day 3 (6% vs 24%, $p=0.05$).

Conclusion: HIE infants with brain injury have a late failure of cerebral autoregulation, manifested as a hyperperfusion burden, suggesting pathologic events are active on day 3 of hypothermia. This finding may help to identify infants which might need additional neuroprotection.

Keywords

autoregulation; blood pressure; hypoxic-ischemic encephalopathy; MRI; near-infrared spectroscopy

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INTRODUCTION

Hypoxic-ischemic encephalopathy (HIE) is a clinically defined syndrome of neurologic impairment following a discrete perinatal ischemic event (1). The initial hypoxic-ischemic event causes neurons to outstrip their supply of ATP, resulting in mitochondrial failure and cell death. While the affected area may initially be limited, this process sets off a cascade of injury in surrounding tissue, driven by oxidative stress, excitotoxicity, inflammation, and cell death. The cerebrovascular autoregulatory system, which regulates blood flow to the brain, is impaired by hypoxic-ischemia, potentially allowing excessive cerebral blood flow during reperfusion (2) and further exacerbates existing injury by bringing in a fresh supply of potentially harmful reactive oxygen species (3).

Although early initiation of therapeutic hypothermia (TH) is known to reduce morbidity in moderately encephalopathic infants, a significant number of infants have injury despite undergoing TH (4). One possible explanation for this incomplete protection is a late failure of the cerebrovascular autoregulatory system to provide smooth, consistent blood flow to the brain during recovery. Hyperperfusion during recovery, resulting from failure of autoregulation, sets off a cascade of oxidative stress and may result in secondary injury. Older studies in the pre-TH era noted elevated cerebral blood flow velocity, a marker of impaired autoregulation, in the setting of severe injury when compared to those with mild or no injury (5,6), but it is unclear if TH alters the timing or magnitude of this phenomenon nor has there been continuous evaluation of autoregulation after HIE.

In this study, we examine changes in cerebral perfusion during TH to identify the timing and magnitude of hyperemia and the association with brain injury. We utilize continuous near-infrared spectroscopy (NIRS) and invasive arterial blood pressure monitoring to first identify the autoregulatory window, measure the percentage of time the blood pressure exceeds this window, and then associate this hyperperfusion burden with brain injury on subsequent MRI.

MATERIAL AND METHODS

Patient selection

Infants admitted to the NICU for therapeutic hypothermia treatment were recruited for a prospective monitoring study as soon after birth as possible, and no later than 24 hours of age. Inclusion criteria were gestational age ≥ 36 weeks, moderate or severe encephalopathy (identified using modified Sarnat criteria (7)), therapeutic hypothermia treatment, and presence of an umbilical arterial line. Exclusion criteria were prematurity and moribund state. Written informed consent was obtained from the parents prior to initiation of study procedures. The study protocol and procedures were reviewed and approved by the institutional review board.

Sample characteristics

Comprehensive sample characteristics were collected for all included infants. Antenatal factors included mode of delivery, arterial cord blood gas pH, and the Apgar scores at one and five minutes. Demographic factors included estimated gestational age (EGA) in

completed weeks, birth weight, and sex. Clinical factors included inotrope use in the first 72 hours of life (inclusive of dopamine, dobutamine, epinephrine, and norepinephrine), hydrocortisone administration (for augmentation of inotropic support), culture-positive sepsis, presence or absence of seizures, and mortality.

Data collection

All data were collected with a sampling rate of 0.5 Hz using a proprietary data acquisition program (CAS Medical Systems, Inc., Branford, CT) which integrates the NIRS and blood pressure signals in a single, time synchronized file on a laptop computer.

NIRS—Cerebral tissue oxygen saturation (SctO₂) was obtained using 5-wavelength near-infrared spectroscopy (FORE-SIGHT Elite, CAS Medical Systems, Inc., Branford, CT) with a transducer containing an LED-emitter and an optical detector located 25 mm from the light source. The non-adhesive optode was placed in a standardized location on the right frontoparietal scalp (chosen due to the limited amount of hair and ease of access to monitor for skin irritation), and recording was conducted over the first 96h following birth.

Blood pressure—Per clinical practice, invasive umbilical blood pressure measurements are made using a pressure transducer (TruWave, Edwards Lifesciences, Irvine, CA), which interfaces the umbilical arterial catheter (3.5 Fr Argyle single lumen umbilical vessel catheter, Medtronic, Minneapolis, MN) with the patient monitor (IntelliVue MP70 or MX800, Philips Medical, Andover, MA).

For blood pressure support, our institutional practice is to consider inotrope initiation when either (a) the mean arterial blood pressure (MABP) <40 mmHg, (b) there is poor urine output (< 1 mL/kg/h) and/or (c) there are clinical signs of poor perfusion (prolonged central capillary refill time) or tachycardia). Hydrocortisone is added as a secondary-line of treatment when there is insufficient response to inotropes.

Data analysis

Pre-processing and error-correction—Prior to analysis, all recordings underwent an automated multistep error correction process. Each recording was first partitioned into 10-second (5 sample) blocks. Blocks were then examined to identify samples with values flagged by the NIRS device or patient monitor as out of range (i.e. probe not in contact with skin) or motion artifact (defined as a sudden, non-physiologic change in MABP or SctO₂ > 5 units over a two-second period). All blocks containing errors were excluded from further analysis.

Autoregulatory window calculation—Using a derivation of the cerebral oximetry index (COx) model developed by Lee and Brady et al. (8–10) the autoregulatory window, the range of blood pressures over which cerebral autoregulation was intact and functioning, was derived. Using the remaining error-corrected data, a moving Pearson correlation coefficient between was calculated for each epoch of 30 consecutive, 10-sec averaged values of MABP and SctO₂ data (with 80% overlap), resulting in values between −1 and +1. In this model, negative values of COx represent active autoregulation, while positive values

represent passive or impaired autoregulation. This correlation value is then paired with the mean MABP during the 300-second epoch resulting in the COx index. COx indices are then sorted by MABP “bin” and the mean COx at each MABP can be calculated.

In this way, the cerebrovascular autoregulatory window can be identified, with a broad range of negative correlations across the range where autoregulation is active with a shift to positive correlations at the upper and lower limits of autoregulation. An example of this output for one infant is shown in Figure 1. In this example, the range of MABP where autoregulation is active (negative bars), can be clearly distinguished from areas where it is impaired or less active (positive bars) and the transition point for the upper and lower limits can be identified.

Hyperperfusion and hypoperfusion burden calculation—For each infant in the study, the bounds of the autoregulatory window were identified for the during the 0–24h, 24–48h, and 48–72h epochs following initiation of TH. The hyperperfusion burden was defined as the proportion of MABP measurements which exceeded the calculated upper limit of autoregulation.

Neuroimaging

Per institutional practice, two non-contrast, non-sedated brain MRIs were obtained after completion of rewarming; once on day 4–5, and again on day 7–10. Utilizing a validated MRI scoring system (11), injury was scored on T1-weighted, T2-weighted and DWI sequences, resulting in an injury category of none, mild, moderate, or severe. The worst score of the two scans was used in the analysis.

Statistical Approach

Univariate comparisons of the sample characteristics were made between uninjured and injured groups using the Mann–Whitney U-test for continuous variables or Fisher’s Exact Test (two-sided) for categorical variables. The Kruskal–Wallis rank sum test was used for comparison of continuous variables when there were more than two groups. Given the novel nature of the analytic method, *a priori* sample size calculation was not performed, rather this was a convenience sample recruited over the 18-month study period. Statistical comparisons were made using R version 3.3.2 (The R Foundation for Statistical Computing, Vienna, Austria).

RESULTS

Sample characteristics

Sixteen infants with HIE were included in the study. Thirteen of the included infants had moderate encephalopathy and three had severe encephalopathy; median date of MRI used in analysis was 5 days of life (range 4–10); brain injury consistent with hypoxic-ischemic injury was noted in 7/16 infants (none=9; mild=4; moderate=2; severe=1). All infants successfully completed the study interventions (NIRS monitoring and MRI scan), although two infants died after the MRI scans, owing to elective withdrawal of support secondary to severity of brain injury. The injured and uninjured groups of infants were comparable except

for the median 1-min Apgar score (1 vs. 2, $p=0.04$), median 5-min Apgar score (2 vs. 5, $p=0.04$) and the incidence of seizures treated with anti-epileptics (57% vs. 0%, $p=0.02$). Inotrope use was limited exclusively to the injured group (43% vs. 0%, $p=0.06$) as was hydrocortisone administration (57% vs. 0%, $p=0.02$). Inotropes were started at a mean age of 15.7 ± 9.5 hours and stopped at a mean age of 120.0 ± 27.7 hours. A complete listing and comparison of sample characteristics can be found in Table 1.

Data quality

The mean recording length was 75 ± 22 hours with a mean data rejection rate of 13%. Of the identified errors, 57% were the result of NIRS probe not in contact with the skin, 33% were the result of out of range blood pressure values (i.e. during blood sampling, infusion of blood products or fluids, or re-calibration of transducer), and 10% were due to motion artifact in either the MABP or NIRS data stream. The mean daily recording length after error correction was 9.5, 16.0, and 16.3 hours on days 1, 2, and 3 respectively. A significant portion of the infants (9/16) had the umbilical arterial catheter removed during day 4, thus there was insufficient data for analysis during that period. There was no significant difference in daily error-corrected recording length between injured and uninjured infants on day 1 ($p=0.19$), day 2 ($p=0.32$), or day 3 ($p=0.28$).

Mean MABP and limits of autoregulation

The mean MABP during the 72-hour TH window was 53.6 ± 2.2 and 50.9 ± 1.5 in the injured and uninjured groups, respectively. The mean lower limit of autoregulation was 46.7 ± 3.9 mmHg, while the mean upper limit of autoregulation was 55.6 ± 5.0 mmHg; the upper and lower limits were not statistically different between the injured and uninjured infants ($p=0.25$ upper limit, and $p=0.67$ for lower limit, respectively). A smoothed plot of the mean MABP for the two groups is shown in Figure 2, with the injured infants depicted with a solid line and uninjured infants shown with a dotted line. Note that the mean MABP in the injured group exceeds the upper limit of autoregulation (upper dashed line) for much of the third day.

Hyperperfusion burden

There was no significant difference in average time spent with MABP above the autoregulatory threshold (i.e. hyperperfusion burden or failed autoregulation) between injured and uninjured infants during day 1, (7 vs. 10%, $p=0.88$) or day 2 (4% vs. 2%, $p=0.88$). In contrast, there was marked increase in time spent with MABP above the autoregulatory threshold on day 3 (54% vs 14%, $p=0.02$) for those with injury compared to those without (Figure 3). Although caution is urged with interpretation in a small sample size, a graded hyperemia response by injury status was also noted (none 14%, mild 47%, moderate 76%, severe 37%, $p=0.09$) for the third day. A difference was also noted between infants who survived and those who died (38% vs. 64%, $p=0.16$).

Hypoperfusion burden

There was no significant difference in average time spent with MABP below the autoregulatory threshold (i.e. hypoperfusion burden) between injured and uninjured infants

during day 1, (32 vs. 27%, $p=0.45$) or day 2 (39% vs. 46%, $p=0.57$). In contrast, there was marked decrease in time spent with MABP below the autoregulatory threshold on day 3 (6% vs 24%, $p=0.05$) for those with injury compared to those without (Figure 4).

DISCUSSION

These data support the previous observation that autoregulatory failure and pathologic reperfusion may play an important role in the development of brain injury following a hypoxic-ischemic insult. Furthermore, we have demonstrated that this effect persists, despite treatment with therapeutic hypothermia, although the onset is shifted later by approximately 24–48 hours when compared to older studies in the pre-TH era. These results suggest that, in infants with brain injury, this reperfusion exceeds the attenuating effect of TH and that there may be a wider therapeutic window (up to 48 hours) to add other therapeutic modalities such as erythropoietin (12) or melatonin (13).

The optimal blood pressure in the neonatal period remains a topic of significant controversy (14). While a target MABP GA in weeks is often used, the underlying data for this assumption is based on infrequent, non-invasive measurements (15,16). As noted, there is a steadily increasing body of literature which suggests that ischemia significantly impairs the function of the autoregulatory system (17,18), exacerbating the impact of blood pressure values above or below the regulatory window. In this study, inotropic agents (both dopamine and hydrocortisone) were used exclusively in the injured group, raising the possibility that the resulting increase in blood pressure in the setting of impaired autoregulation may have exacerbated reperfusion-related brain injury. The finding is in line with adult stroke research which has identified increased risk of poor outcomes for patients with persistently elevated MABP (19). However, the late hyperperfusion observed in this study does not correspond with the timing of inotropic medication initiation, suggesting that inotrope use is perhaps another indicator of cardiovascular instability, rather than the proximal cause of the hyperemia.

The graded hyperemia response noted on day three suggests a linear relationship between the degree of hyperemia and degree of injury. However, it is interesting to note that infants with severe brain injury on MRI had less hyperemia than those with mild or moderate injury. For those infants, it is possible that the severity of the initial injury has drastically reduced overall brain metabolism to such a degree that they are unable to mount a robust hyperemia. This speculation is supported by the TH literature (20) which suggests that infants with severe injury gain very little from TH treatment, as secondary brain injury is a smaller component of the overall injury. However, given the small sample size of the study, this conclusion is purely conjecture and needs additional study in larger cohorts.

Intriguingly, infants in this study spent minimal time with MABP below the regulatory window, suggesting that *hypoperfusion* likely does not play a role in HIE-related brain injury, at least not after the initial ischemic event. This finding is particularly striking given the apparent deleterious relationship between inotrope use and brain injury, and suggesting a possible role for permissive hypotension, a concept gaining some favor in trauma resuscitation (21). Although similar trials have not been conducted in term infants, a recent

clinical trial of active, moderate, and permissive blood pressure management strategies in premature infants demonstrated no difference in clinical, EEG, or hemodynamic outcomes (22). As the upper and lower boundaries of MABP for optimal outcome in the HIE infants are not known, and may even be different between individuals, at a minimum, permissive hypotension should undergo further evaluation.

The underlying factors which generate this late surge of hyperperfusion in the injured infants are not identified in this study but do highlight the 48-hour period prior to autoregulatory failure as a window of opportunity to provide additional neuroprotective intervention to prevent this secondary injury. While the results of this study highlight the mechanism by which secondary injury may occur, additional research is needed to identify early markers of cerebrovascular autoregulatory failure. This population can then be targeted for further neuroprotective strategies to reduce the burden of injury in this group.

There are several limitations to this study which should be considered. First, this study has a relatively small sample size, one which should be expanded prior to broad application of this concept to the general HIE population. Given the relatively small number of infants treated for HIE at each center (and the even smaller number with umbilical arterial lines needed for the study methodology) future studies may require multi-center collaborative efforts. Second, the nature of NIRS is such that it measures cerebral oxygenation over an indiscriminate volume of cerebral tissue and is not localized to specific regions of the brain. Future high-resolution optical imaging devices may be developed which permit a more granular estimation of regional blood flow in longitudinal study (23). Third, as previously mentioned, this study demonstrates an association between injury and late hyperperfusion. As imaging is not performed until after the cooling process is complete, it is not possible to determine whether the injury is driving the sharp increase in blood flow outside of the autoregulatory window or some other factor is precipitating the shift to blood pressures outside the window, causing brain injury in the process. Fourth, this study is focused on a short-term primary outcome, namely injury on brain MRI after completion of therapeutic hypothermia. Future studies should examine the relationship between hyperperfusion and long-term neurodevelopmental outcomes.

These results suggest that careful attention should be paid to blood pressure management during the later stages of cooling, incorporating a management approach that considers the current state of cerebral autoregulation, rather than a fixed, arbitrary number. Evaluating the use of inotropic medications, painful stimuli (such as heel sticks for blood sampling) or interventions which increase the basal metabolic rate (aggressive weaning of the ventilator or sedation) may help to prevent unnecessary elevation in blood pressure, above the autoregulatory threshold, which might trigger further brain injury in some infants. As mentioned, this study also highlights the need to develop a robust, dynamic monitor of the cerebrovascular autoregulatory system, informing blood pressure management and potentially triggering the use of additional neuroprotective strategies. Furthermore, this study underscores the need to add routine, longitudinal brain monitoring to the standard package of vital signs monitored in the NICU to better understand the underlying pathophysiology influencing injury.

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Figure 1 –.
Mean COx by MABP bin is shown. Negative values represent areas of active autoregulation, positive values represent areas of passive or inactive autoregulation.

Figure 2 –. Mean MABP during the first 72 hours of life. Injured infants are shown with a solid line; uninjured with a dotted line. The mean upper and lower limits of autoregulation are shown as horizontal dashed lines.

Figure 3 –.

Box and whisker plot of hyperperfusion burden by day, divided by injury group. Note no significant difference on days 1 and 2, with marked increase in the injured group on day 3.

Figure 4 –.

Box and whisker plot of hypoperfusion burden by day, divided by injury group. Note no significant difference on days 1 and 2, with decreased in the injured group on day 3.

Table 1.

Patient Characteristics

	No injury (n=9)	Injury (n=7)	P value
Gestational age, mean (SD), weeks	37.4 (1.7)	37.4 (0.9)	1.00
Birth weight, mean (SD), grams	3206.11 (727.5)	3158.6 (739.5)	0.79
Male sex, n (%)	4 (44%)	3 (43%)	1.00
Died, n (%)	0 (0)	2 (29%)	0.18
Stage of encephalopathy at TH initiation, n (%)			
Mild	0 (0)	0 (0)	0.06
Moderate	9 (100)	4 (57)	
Severe	0 (0)	3 (43)	
1-minute Apgar, median (range)	2 (0–4)	1 (0–1)	0.04 [*]
5-minute Apgar, median (range)	5 (0–8)	2 (0–6)	0.04 [*]
Arterial cord blood pH, mean (SD)	7.01 (0.14)	6.85 (0.26)	0.22
Seizures, n (%)	0 (0)	4 (57)	0.02 [*]
Culture positive sepsis, n (%)	0 (0)	0 (0)	1.00
Inotrope use in first 72h, n (%)	0 (0)	3 (43)	0.06
Postnatal age at inotrope start, mean (SD), hours		15.7 (9.5)	n/a
Postnatal age at inotrope stop, mean (SD), hours		120.0 (27.7)	n/a
Hydrocortisone use in first 72h, n (%)	0 (0)	4 (57)	0.02 [*]

Footnote:

^{*} denotes significance at $p < 0.05$.