Time to Reaching Target Cooling Temperature and 2-year Outcomes in Infants with Hypoxic-Ischemic Encephalopathy

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Objective To determine if time to reaching target temperature (TT) is associated with death or neurodevelopmental impairment (NDI) at 2 years of age in infants with hypoxic-ischemic encephalopathy (HIE).

Study design Newborn infants ≥36 weeks of gestation diagnosed with moderate or severe HIE and treated with therapeutic hypothermia were stratified based on time at which TT was reached, defined as early (ie, ≤4 hours of age) or late (>4 hours of age). Primary outcomes were death or NDI. Secondary outcomes included neurodevelopmental assessment with Bayley Scales of Infant and Toddler Development, third edition (BSID-III) at age 2.

Results Among 500 infants, the median time to reaching TT was 4.3 hours (IWR, 3.2-5.7 hours). Infants in early TT group (n = 211 [42%]) compared with the late TT group (n = 289 [58%]) were more likely to be inborn (23% vs 13%; $P < .001$) and have severe HIE (28% vs 19%; $P = .03$). The early and late TT groups did not differ in the primary outcome of death or any NDI (adjusted RR, 1.05; 95% CI, 0.85-0.30; $P = .62$). Among survivors, neurodevelopmental outcomes did not differ significantly in the 2 groups (adjusted mean difference in Bayley Scales of Infant Development-III scores: cognitive, −2.8 [95% CI, −6.1 to 0.5], language −3.3 [95% CI, −7.4 to 0.8], and motor −3.5 [95% CI, −7.3 to 0.3]).

Conclusions In infants with HIE, time to reach TT is not independently associated with risk of death or NDI at age 2 years. Among survivors, developmental outcomes are similar between those who reached TT at <4 and ≥4 hours of age. *(J Pediatr 2024;266:113853)*.

Trial Registration High-dose Erythropoietin for Asphyxia and Encephalopathy (HEAL); NCT02811263; https://beta.clinicaltrials.gov/study/NCT02811263.

Therapeutic hypothermia (TH) initiated within 6 hours of birth and maintained for 72 hours in newborn infants with hypoxic-ischemic encephalopathy (HIE) reduces brain injury and improves the chances of survival without neurodevelopmental impairment (NDI).[1-4] Preclinical experimental studies of TH suggest that the degree of neuroprotection following hypoxia-ischemia depends on the start time, duration and depth of hypothermia, as well as on the severity of injury.[5-11]

Randomized controlled trials suggest that TH initiated within 6 hours of birth offers more benefit than starting this therapy after 6 hours.[12,13] However, it remains unclear whether reaching target temperature (TT) sooner rather than later within this 6-hour window offers an additional advantage. Small retrospective cohort studies suggest that initiating TH sooner and reaching the TT earlier may decrease seizures and improve motor outcomes in survivors.[14,15] However, other studies suggest that reaching TT earlier has no impact on outcomes or may be associated with an increased rate of death, possibly because infants who are more severely affected demonstrate spontaneous cooling and therefore reach TT sooner.[14-17]

In the High Dose Erythropoietin for Asphyxia and encephaLopathy (HEAL) trial, the use of erythropoietin in addition to TH did not alter the rate of death or NDI.[18] We performed a secondary analysis to evaluate the association between the time to reaching TT and outcomes at 2 years of age. We hypothesized that reaching TT at <4 hours of age compared with at ≥4 hours is independently associated with death or NDI after controlling for the severity of HIE.

BSID-III	Bayley Scales of Infant and Toddler Development, third edition
GLM	Generalized linear model
GMFCS	Gross Motor Function Classification System
HIE	Hypoxic-ischemic encephalopathy
NDI	Neurodevelopmental impairment
TH	Therapeutic hypothermia
TT	Target temperature

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Methods

The HEAL trial was a prospective multicenter randomized controlled trial that enrolled 500 newborn infants with moderate or severe HIE who were randomized to receive erythropoietin or placebo.[18] The study protocol and main study findings have been previously reported.[18,19] All infants were born at ≥36 weeks of gestation; had ≥1 signs of perinatal depression (ie, Apgar score of <5 at 10 minutes; resuscitation beyond 10 minutes of age; or pH <7.00 or base deficit ≥15 mmol/L in an umbilical cord or infant arterial or venous gas obtained within 60 minutes of birth); had moderate or severe encephalopathy based on a modified Sarnat examination performed between 1 and 6 hours of age; and received active or passive TH initiated at <6 hours of age. Exclusion criteria included birthweight <1800 g, head circumference <30 cm, genetic or congenital condition affecting neurodevelopment, hematocrit of >65%, parents and medical team considering redirection to palliative care, encephalopathy attributed to a postnatal event, guardian of diminished cognitive capacity, or patient anticipated to be unavailable for evaluation at age 2 years.

Maternal data included self-assigned race, mode of delivery, and delivery complications, including sentinel events (uterine rupture, tight nuchal cord, placental abruption, prolapsed cord, or shoulder dystocia) and site of delivery. Infant characteristics included sex, birth weight, gestational age, resuscitation efforts, severity of encephalopathy, and the time from birth to reaching TT (ie, 33.5 ± 0.5°C).

The primary outcome of this study was death or NDI at 22-36 months of age. NDI was defined as any of the following: (1) cerebral palsy, determined by a validated standardized neurologic examination, (2) Gross Motor Function Classification System (GMFCS) >1 (ie, unable to walk 10 steps independently), or (3) cognitive score of <90 on Bayley Scales of Infant and Toddler Development, third edition (BSID-III). Severity of NDI was defined as previously described.[18,20] Moderate or severe NDI was defined as a GMFCS of 1 and cerebral palsy, and a GMFCS of ≥2 as quadriplegic cerebral palsy, or BSID-III cognitive score of <85. Mild NDI was defined as NDI not meeting criteria for moderate or severe NDI.

Secondary outcomes consisted of BSID-III cognitive, language and motor scores; severity and pattern of injury on brain MRI performed at 4-6 days of age using a harmonized neuroimaging protocol; and a 4-level ordinal outcome consisting of death, moderate or severe NDI, mild NDI, or no NDI.[18,21] Short-term inpatient outcomes included clinical or electrographic seizures, status epilepticus, inotropic support, steroids for blood pressure, pulmonary hypertension, use of inhaled nitric oxide, extracorporeal membrane oxygenation, need for tracheostomy, nasogastric tube or gastrostomy tube at discharge, and discharged alive.

Statistical Analyses

Baseline characteristics were compared between early and late TT groups using mean ± SD or median (IQR) as appropriate. Owing to intermittent missing data across the set of predictors and outcome data, we first conducted a multiple imputation analysis (mice R package; m = 10 imputations) to impute values for missing predictors and outcomes.[22] To compare outcomes statistically between TT groups, we used generalized linear models (GLM) for continuous (Gaussian), binary (logistic, log link), and ordinal (proportional odds logistic) outcome measures respectively. Each GLM model adjusted for prognostic and potential confounding variables (maternal race, maternal ethnicity, maternal age, gestational diabetes, gestational age, APGAR score at 10 minutes, delivery room chest compressions, delivery room epinephrine), as well as recruitment site, severity of HIE, and randomized treatment assignment as fixed factors reflecting the original study design. Baseline variables were identified as prognostic for BSID-III cognitive scores at 22-36 months using a liberal screening strategy (if associated with $P < .15$, GLM) and these were included as covariates to evaluate TT group. Confounding variables were included if they were associated with both TT group membership and BSID-III at 22-36 months ($P < .15$ in both analyses), and their inclusion in the BSID-III model changed the effect of TT group on the outcome by >10%. GLM analyses to identify prognostic and confounding variables adjusted for randomized treatment assignment, HIE severity, and recruitment site. In an exploratory analysis, a statistical interaction term between HIE severity and TT group was included to assess whether the association between TT group and BSID-III scores was different for moderate vs severe HIE. All analyses were conducted using the R statistical package (Version 4.0.2, Foundation for Statistical Computing, Vienna, Austria, https://www.R-project.org).

Results

Among the 500 enrolled infants, 104 (21%) reached TT within 3 hours, 211 (42%) within 4 hours, and 407 (81%) by 6 hours of birth. The median time to reaching TT was 4.3 hours (IQR, 3.2-5.7 hours) (**Figure 1**, online available at www.jpeds.com). Further, when examining TT as a continuous variable and outcome of death or NDI, the area under receiving operating characteristic curve was 0.54 with an optimal cut point near 4 hours. We, therefore, stratified infants into 2 groups depending on whether they reached TT at or before 4 hours (early TT group; n = 211 [42%]) or after 4 hours (late TT group; n = 289 [58%]) of age. The time to reaching TT for the whole cohort ranged from 29 minutes to 15.9 hours and varied across the 17 study sites (**Figure 2**, online available at www.jpeds.com). The median time to TT was 3.05 hours (IQR, 2.25-3.55 hours) in the early TT group and 5.48 hours (IQR, 4.77-6.30 hours) in the late TT group. Twenty infants (4%) did not complete 72 hours of cooling, including 9 infants in the early and 11 in the late TT groups. Reasons for the shorter duration of cooling included redirection of care

(n = 15), severe pulmonary hypertension (n = 1), and extracorporeal membrane oxygenation (n = 4).

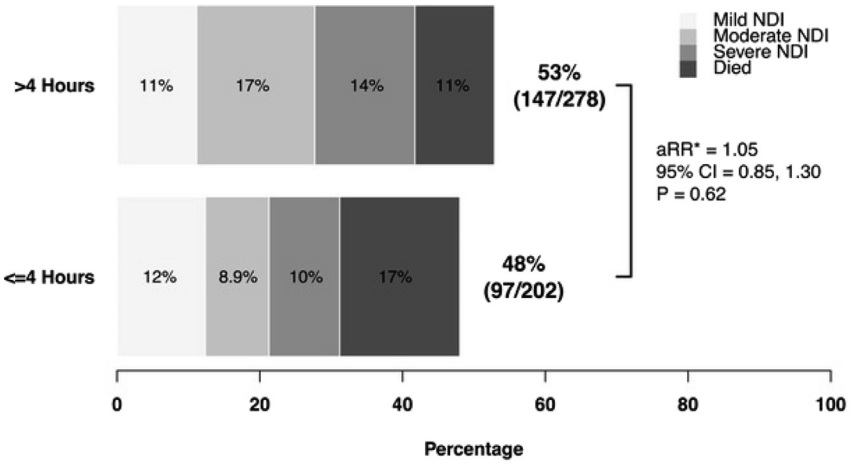
Compared with the late TT group, infants in the early TT group had a higher proportion of White mothers ($P = .04$), were more likely to be inborn (23% vs 13%; $P < .001$) and more likely to have severe HIE (28% vs 19%; $P = .03$). The 2 groups were otherwise similar with respect to demographic and clinical factors (Table I).

The primary outcome of death or NDI was available for 480 of 500 (infants 96%) (Figure 3). Of these 480 infants, outcomes included 65 deaths (13%), 123 (25%) with moderate/severe NDI, 56 (11%) with mild NDI, and 236 (42%) with no NDI. After adjusting for potential confounders (maternal race, maternal ethnicity, maternal age, gestational diabetes, gestational age, encephalopathy severity, APGAR score at 10 minutes, delivery room chest compressions, delivery room epinephrine, recruitment site, and randomized treatment assignment), there were no meaningful differences in the primary outcome of death or NDI among infants who reached TT before compared with after 4 hours of age (adjusted RR, 1.05; 95% CI, 0.85-1.30; $P = .62$). Similarly, there was no difference in the 4-level

Table I. Maternal and neonatal clinical and demographic characteristics in the early compared with late TT groups

Characteristics	Early TT group (≤4 hours)	Late TT group (>4 hours)	*P* value*
No. present with data	211	289	
Maternal characteristics			
Race			**.04**
White	157 (74)	199 (69%)	
Black	28 (13)	38 (13%)	
Asian	11 (5.2)	22 (7.6%)	
Multiple/other	6 (2.8)	10 (3.5%)	
Unknown/not reported	9 (4.3)	20 (6.9%)	
Hispanic ethnicity	50 (24)	72 (25%)	.60
Age (y)	29.8 ± 6.1	29.6 ± 6.5	.72
Nulliparous (parity = 1)	81 (38)	133 (46%)	.98
SSRI use	11 (5.2)	18 (6.2%)	.12
Pregnancy and delivery complications			
Maternal chorioamnionitis or fever	37 (18)	40 (14%)	.24
Maternal preeclampsia or eclampsia	13 (6.2)	32 (11%)	.90
Gestational diabetes	21 (10)	37 (13%)	.06
Maternal obesity (body mass index >30)	39 (19)	50 (17%)	.20
Sentinel event	56 (27)	87 (30%)	.58
Shoulder dystocia	17 (8.1)	15 (5.2%)	.20
Placental abruption	27 (13)	44 (15%)	.47
Prolapsed cord	8 (3.8)	15 (5.2%)	.20
Uterine rupture	8 (3.8)	16 (5.5%)	.72
Delivery mode			.33
Spontaneous vaginal	54 (26)	65 (22%)	
Vacuum or forceps (vaginal assisted)	25 (12)	27 (9.3%)	
Elective cesarean	4 (1.9)	8 (2.8%)	
Emergency or urgent cesarean	128 (61)	189 (65%)	
Place of birth			**<.001**
Inborn	48 (23)	37 (13%)	
Other hospital	143 (68)	220 (76%)	
Other (home, clinic, in transit)	20 (9.5)	32 (11%)	
Infant characteristics			
Female	110 (52)	165 (57%)	.80
Birth weight (g)	3415 ± 552	3341 ± 621	.29
Gestational age (w)	39.2 ± 1.4	39.0 ± 1.5	.99
5-minute Apgar			.23
0-3	123 (59)	135 (48%)	
4-6	72 (35)	120 (43%)	
7-10	13 (6.3)	27 (9.6%)	
10-minute Apgar			.07
0-3	64 (33)	59 (23)	
4-6	96 (49)	121 (47)	
7-10	36 (18)	80 (31)	
Resuscitation > 10 minutes†	193 (91)	267 (92)	.86
Chest compressions	75 (36)	83 (29)	.88
Lowest pH‡	6.91 ± 0.18	6.94 ± 0.17	.15
Worst base deficit‡	18.58 ± 6.30	18.13 ± 6.08	.63
Delivery room epinephrine	55 (26)	38 (13)	.06
Severe HIE	59 (28)	54 (19)	**.03**
Randomized to erythropoietin	109 (52)	148 (51)	.99

Values are number (%) or mean ±SD. *P* values <.05 were considered significant and shown as bolded.
*GLM adjusted for HIE severity, recruitment site, and randomized treatment assignment.
†Ongoing resuscitation with chest compressions, mechanical ventilation, or both at 10 minutes of age.
‡Lowest pH or worst base deficit among cord arterial, cord venous or arterial blood gas samples obtained at <60 minutes of age.



*aRR: adjusted relative risk (see text for details).

Figure 3. Primary outcome of death or NDI at age 2 years, in infants who reached TT at <4 hours of age compared with those who reached TT at ≥4 hours of age.

ordinal outcome of death, moderate/severe NDI, mild NDI, or no NDI (aOR, 1.11; 95% CI, 0.68-1.79) (**Table II**).

Among survivors of HIE, the unadjusted mean differences in the BSID III scores were lower in the late TT group compared with the early TT group as follows: mean difference in cognitive score −5.9 (86.9 vs 92.8, P = .04), motor score −4.2 (89.1 vs 93.3, P = .04), and language score −6.5 (84.8 vs 91.3, P = .08). However, after adjusting for potential confounders (maternal race, maternal ethnicity, maternal age, gestational diabetes, gestational age, HIE severity, APGAR score at 10 minutes, delivery room chest compressions, delivery room epinephrine, recruitment site, and randomized treatment assignment), the magnitude of group differences is attenuated with a difference in mean cognitive score of −2.8 (95% CI, –6.1, 0.5; P = .09), language score of −3.3 (95% CI, –7.4 to 0.8; P = .11), and motor score of −3.5 (95% CI, –7.3 to 0.3; P = .07) (**Figure 4**; **Table III**, online available at www.jpeds.com). Further, when stratified by severity of encephalopathy, the relationship between time to reaching TT and BSID III scores, across all domains were similar (**Table III**, online available at www.jpeds.com).

Brain MRI was performed in 199 of 211 infants (95%) in the early TT group, and in 274 of 289 infants (94%) in late TT group. There were no significant differences in severity or pattern of brain injury between the 2 groups (**Table IV**, online available at www.jpeds.com). The early TT and late TT groups did not differ with regard to frequency of neonatal complications such as clinical or electrographic seizures, status epilepticus, inotropic support, steroids for blood pressure, pulmonary hypertension, use of inhaled nitric oxide, extracorporeal membrane oxygenation, tracheostomy, nasogastric or gastrostomy tube feeds upon hospital discharge, or being discharged alive (**Table IV**, online available at www.jpeds.com).

Table II. Four-level secondary outcome in infants who reached TT early compared with those who reached TT later

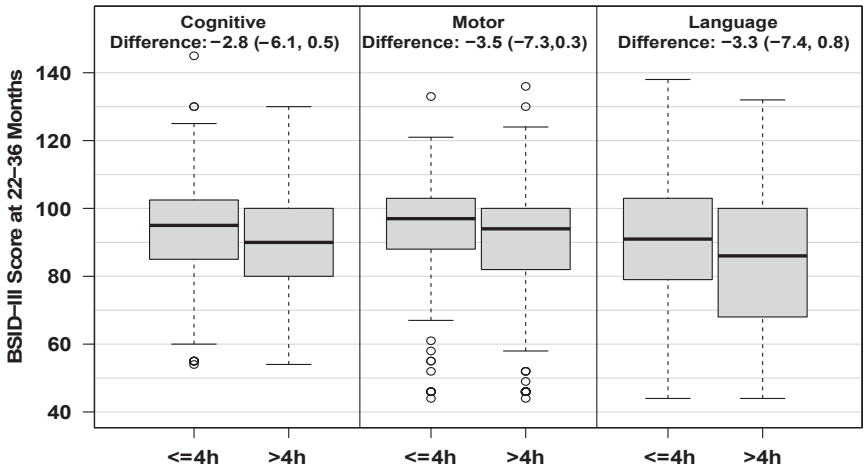
Outcomes	Early TT group (≤4 hours)	Late TT group (>4 hours)	aOR* (95% CI); *P* value
No.	202	278	1.11 (0.68-1.79);
No NDI	105 (52)	131 (47)	*P* = .39
Mild NDI	25 (12)	31 (11)	
Moderate/severe NDI	38 (19)	85 (31)	
Death	34 (17)	31 (11)	

Values are number (%) unless otherwise specified.
*OR, 95% CI, and *P* value evaluated with an adjusted proportional odds logistic regression model adjusted for maternal race, maternal ethnicity, maternal age, gestational diabetes, gestational age, HIE severity, APGAR score at 10 minutes, delivery room chest compressions, delivery room epinephrine, recruitment site, and randomized treatment assignment.

Discussion

In a large prospective cohort of infants with moderate or severe HIE treated with TH, we found that reaching TT by 4 hours of age was not associated with risk of death or NDI at 2 years of age. Similarly, among survivors of HIE, the mean BSD-III cognitive, language, and motor scores were similar in the early and late TT groups.

Infants in previous clinical trials of TH were randomized to hypothermia or standard care between 4 and 5 hours of age and typically reached the TT 60-120 minutes thereafter.[1,3,23] These trials were not designed to evaluate outcomes based on time to reaching TT. Post hoc analyses from the TOBY trial[3] suggested a trend toward lower rates of death or severe NDI in infants who were randomized before 4 hours (OR, 0.77; 95% CI, 0.44-1.04) compared with those randomized after 4 hours of age (OR, 0.95; 95%



Data shown as adjusted* mean difference (95% CI). See text for details.

Figure 4. Two-year BSID-III scores in survivors of HIE who reached TT at <4 hours of age, compared with those who reached TT at ≥4 hours of age.

CI, 0.72-1.25), whereas other trials noted no effects of age at randomization on outcomes.[23,24]

Prior retrospective studies have yielded conflicting results with regards to time to initiate TH and HIE outcomes. Youn et al noted fewer seizures in infants who were started on TH by 1 hour of age.[14] Thoresen et al noted no differences in moderate to severe disability in survivors of HIE who initiated TH at <3 hours of age; however, BSID-II psychomotor scores were higher among surviving infants who initiated TH at <3 hours of age.[15] In contrast, Guillot et al found no differences in 18-month outcomes when comparing infants who initiated TH before and after 3 hours of age, although they included infants with mild HIE and relied on parental reporting on the Ages and Stages Questionnaire instead of in-person developmental assessments.[16] Gilmore et al observed no association between cerebral autoregulation measured by near infrared spectrometry and the time to initiation of TH; however, they did note a lower apparent diffusion coefficient scalar in the right posterior centrum semiovale in infants cooled at >3 hours of age, which can be associated with gait dysfunction.[25]

Although most studies examined HIE outcomes based on age at initiation of TH, our study examined the time to reaching TT. Among prior studies that also evaluated HIE outcomes in relation to the time to reaching TT, one found no association with death or NDI,[17] whereas another reported a higher rate of long-term adverse outcomes in infants who reached TT sooner after birth.[26] Our finding that the time to reaching TT has no effect on MRI brain injury parallels a study by Guillot et al, in which there were similarly no differences in severity or pattern of brain injury on MRI based on age at initiation of TH.[16]

Similar to previous studies, the infants in our cohort who reached TT earlier were more likely to have severe encephalopathy.[15,16,26] This may be because acidotic infants have decreased heat production, and more severely asphyxiated infants have a steeper decline in body temperature after birth.[27-29] Infants in our cohort who were in the early TT group were also more likely to be inborn, which likely resulted in earlier treatment initiation.

In preclinical models, the precise timing and severity of brain injury are well-documented, and cooling is neuroprotective if initiated within 5.5 hours of injury.[6] Severe injury is associated with a shorter latent phase and a worse secondary severe energy failure, likely limiting the window of treatment and efficacy of TH.[8,9] In clinical practice, however, the timing of injury is not typically known, and sentinel events are identified in only about a third of infants, and have not been shown to impact neurodevelopmental outcomes.[18,30] We noted no difference in the frequency of sentinel events in the 2 groups. Finally, although we noted no effect of time to TT on neurodevelopmental outcomes by encephalopathy severity, clinically, severe encephalopathy is associated with a 10- to 20-fold higher odds of death or NDI[17] and benefit to survival or limiting disability in survivors is largely limited to infants with moderate and not severe encephalopathy.[13,31]

We report one of the largest prospective cohorts of infants with HIE treated with TH to undergo detailed 2-year neurodevelopmental assessments. However, our dataset has limitations. We did not record the age at initiation of TH, the baseline temperature before starting cooling, or whether infants received active cooling on transport, which could influence the time to reaching TT.[32] We did not record every temperature measurement during cooling and rewarming and, therefore, cannot comment on the effects of hyperthermia or overcooling that are known to affect outcomes.[26,33-35] Finally, although we adjusted for the effects of recruitment

site, we are not able to account for all differences in clinical management that may have impacted the time to reaching TT.

In conclusion, among infants with moderate or severe HIE, reaching TT at <4 hours of age is not associated with the risk of death or NDI. Similarly, among survivors of HIE, reaching TT before 4 hours of age is not associated with BSID-III cognitive, language, or motor scores at age 2 years. ■

Declaration of Competing Interest

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The authors declare no conflicts of interest.

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Data Statement

Data sharing statement available at www.jpeds.com.

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