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Research article

Association of Clinical Events to the Time to a Strict Definition of Sustained Feeding Tolerance in Premature Infants in the ´Connection Trial'

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Abstract

Objective. The association of clinical events with the time to a strict definition of sustained enteral feeding tolerance (SFT) is evaluated in extremely low birth weight (BW) infants. Study Design. The Connection Trial (Id: NCT03978000) is a randomized, double-blind phase 3 study under US IND and EU CTX of the pharmaceutical grade probiotic IBP-9414 (L. reuteri). A primary endpoint is time to SFT defined as the first day of the first period of 10 consecutive days during which the infant receives a total daily enteral feeding of ≥120 ml/kg body weight without use of parenteral macronutrients and with an average growth of ≥10g/kg/day. Association of the time to SFT was analyzed for 28 clinical events with univariable and multivariable stepwise analyses. Results. 519 infants reached SFT after mean 18 days (SD, 11 d). Univariable analysis correlated ($p<0.001$-0.03) the time to SFT with 23 of the explored events with the numerically greatest delay to SFT for gastrointestinal (GI) perforation, hypotension, serious cardiac events, and pneumonia (mean delay, 10.1-21.0 days). The time to SFT strongly influenced the duration of hospital stay. Multivariable analysis correlated ($p<0.001$- 0.04) the time to SFT with GI perforation, serious cardiac events and hypotension (mean delay, 5.7-15.4 days) as well as to e.g., sepsis, antibiotic treatment days, serious respiratory events, and pneumonia. Infants not reaching SFT (n=122 infants) had higher incidences ($p<0.001$-0.028) of serious adverse events including GI perforation, obstruction, and necrotizing enterocolitis as well as hypotensive events and systemic antibiotic treatment duration. Conclusion. Considering the importance of enteral feeding to the development of premature infants, current routines on the progress of enteral feeding may benefit from evaluation.

Introduction

Full enteral feeding is advantageous to reach early in life in the intensive care treatment of premature infants [1]. Adherence to a variety of standardized feeding protocols has been shown to assist in both the initiation and advancement of enteral feeding volumes [2,3]. Many clinical events may limit enteral feeding volumes, and these characteristically are the most prevalent in the very low birth weight (BW) infants [4-6]. These events include recognized complications especially in the gastrointestinal (GI) tract and those relating to risks of inducing or aggravating extra-GI events like hypotension, cardiac and respiratory com-

plications. The extent to which these complications should be expected to limit enteral feeding, either by themselves or in combinations characteristic of the clinical situation, has not been detailed.

A recognized measure of the degree of enteral feedings is the age at which the infant reaches full enteral feeding. This usually is defined as the infant tolerating a total daily volume of 120 to 150 ml/kg or an absent need of parenteral nutrition for up to a few days [7-10]. In a study of infants recruited into the 'Connection Trial', we introduced a strict definition of sustained feeding tolerance (SFT) and displayed the coupling of a one-day shift in SFT to a range of clinical events that occur perinatally in the care of extremely low BW infants [11]. In the current analysis, we explore a larger group of infants of the 'Connection Trial' as to the effect on the time to SFT for a rather extensive number of clinical events. The principal aim of this analysis is to provide numerical data on the time to SFT whenever clinical complications occur in the care of extremely preterm infants.

Material and Methods

The 'Connection Trial'

The 'Connection Trial' is a phase 3, placebo-controlled multicenter study in the US and EU, including Israel, on the safety and efficacy of IBP-9414 (Table 1). Infants of 500-1500g BW are randomized 1:1 to a single daily enteral dose of IBP-9414 containing L. reuteri at 10^9 colony- forming units, or sterile water placebo, from within 48 hrs. of birth through a corrected gestational age (cGA) of 34 weeks + 6 days. Follow-up is conducted at cGA 40 weeks + 7 days. The primary endpoints of the 'Connection Trial' are the incidence of necrotizing enterocolitis (NEC) and the time to SFT.

Sustained feeding tolerance

SFT is defined as the first period of at least 10 consecutive days during which the infant tolerates a total daily enteral feeding volume of at least 120 ml/kg body weight with no use of parenteral nutrition (i.e., amino acids or fat) and with an average body weight increase of at least 10g/kg/day. The 10-day period was recommended by an expert panel as a relevant duration for defining SFT and a single day shift in the time to SFT, defined in this way, has previously been shown to be sensitive to a number of clinically relevant outcomes [11]. The time to SFT is measured from the first dose of study product to the first day of the SFT period.

Patient cohort

Patient recruitment into the 'Connection Trial' was initiated, as a precaution, in infants of 750-1000g BW with extension to 500-1000g BW after independent safety evaluation. A second, independent safety evaluation occurred as per the clinical trial protocol when 731 infants had completed the treatment period. Among these infants, 90 infants had withdrawn early from the study due mainly to reasons of death (n=48), withdrawn consent on study participation and transfer to a non-participating center (Table 2).

The remaining 641 infants constitute the investigated cohort of the present study. Data for the infants were collected to cGA 34 weeks +6 days (mean, 53 days), but for the duration of hospitalization that was collected for up to 40 weeks + 7 days. In total, 519 (81%) of the infants reached SFT and were investigated for an association of 28 clinical events and event categories with the time to SFT (Table 3). They had mean 851g BW (range 510- 1 000g) and GA 27 weeks (range 23-32 w) at birth (Table 4). The 122 infants completing the study without reaching SFT had the same GA at birth and a numerically, but not statistically significantly lower BW.

Statistics

Variables analyzed for an association with the time to SFT was performed without knowledge of the allocation of the infants into the treatment arms of the 'Connection Trial'. They consisted of data reported by investigators managing the infants. The diagnosis of NEC, however, relied on independent adjudication of abdominal X-rays demonstrating intestinal pneumatosis and/or portal venous gas, and NEC recognized at exploratory laparotomy or autopsy. Analysis of the 28 clinical events related with the time to SFT was conducted in two steps. In the first step all events were tested using linear regression with the event as the independent variable and the time to SFT as the dependent variable. Thereafter, all events with a univariable p-value

Table 1. The 'Connection trial' at a brief (ClinicalTrials.gov Id: NCT03978000)

Study design: Randomized, placebo-controlled, parallel group, multicenter phase 3 study under US IND and EU CTX.
Eligible patients: Infants with a birth weight of 500- 1500g and GA 23-32w meeting the inclusion and exclusion criteria of the study.
Primary Endpoints: Incidence of NEC and time to SFT. Secondary endpoints: Medical NEC, Surgical/autopsy NEC, all-cause mortality, weight gain, duration of hospitalization, feeding intolerance.
Study drug: IBP-9414 containing pharmaceutical grade L. Reuteri given orally or by enteral tube at a single daily dose of 10^9 CFU or sterile water placebo from within 48 hrs. of birth to 34 weeks +6 days GA.
Contribution centers in Bulgaria, France, Hungary, Israel, Poland, Romania, Serbia, Spain, UK, and US.

CFU, Colony- forming units; CTX, Clinical trail exemption; EU, European Union; g, gram; GA, gestational age; IND, investigative new drug; NEC, necrotizing enterocolitis; SFT, sustained feeding tolerance; US, United States

Table 2. Data at birth for infants discontinuing the study early and those investigated for SFT.

	Total n=731	Infants withdrawn early from the study n=90*	Investigated infant cohort n=641
Weight (gram)			
Mean (sd)	844 (115)	827 (129)	847 (113)
Median	860	840	860
GA (week)			
Mean (sd)	27 (2)	27(2)	27 (2)
Median	27	26	27

*Reasons for study withdrawal include death (48 infants), withdrawn consent (10 infants), adverse event (6 infants), lost to follow-up (11 infants) and miscellaneous (mainly transfer to non-participating centers, 15 infants).

Table 3. Clinical outcomes analyzed for association to the time to SFT.

Birth weight.
GA at birth.
Apgar score at 5 min.
Number of days with clinical signs of feeding intolerance as defined by investigators (e.g., vomiting, abdominal distention, bradycardia) per infant.
Number of infants with 'abdominal signs' defined as abdominal distension, abdominal discoloration, vomiting, reflux, disturbed motility, impaired gastric emptying, hematochezia, and hematemesis of any severity.
Number of infants with a gastrointestinal adverse event like abdominal distension, diarrhea, gastroenteritis, gastrointestinal hemorrhage, gastrointestinal necrosis, hematochezia, ileus, intestinal perforation, necrotizing colitis, necrotizing enterocolitis, peritonitis, pneumoperitoneum, vomiting of any severity.
Number of infants with any serious gastrointestinal event*.
Number of infants with gastrointestinal perforation like spontaneous intestinal perforation, esophageal perforation, gastric perforation.
Number of infants with gastrointestinal obstruction like ileus, bowel obstruction, volvulus.
Number of infants with NEC confirmed by independent evaluation of abdominal radiographs, explorative laparotomy or autopsy.
Number of infants with any serious adverse event*.
Number of infants with clinically suspected sepsis ≥72 hrs. of age.
Number of infants with sepsis (blood culture positive >72 hrs. after birth).
Number of days with antibiotic use (class intended for systemic use) per infant.
Number of infants with pneumonia (incl. bronchiolitis).
Number of infants with a serious respiratory event* like bronchopulmonary dysplasia, respiratory failure, apnea, pulmonary hemorrhage, pulmonary hypertension, pulmonary emphysema, pneumothorax, tracheomalacia.
Number of infants with bronchopulmonary dysplasia, chronic respiratory insufficiency.
Number of infants with intracranial, intraventricular and periventricular hemorrhage.
Number of infants with persistent ductus arteriosus of any severity.
Number of infants with a serious cardiac event* like bradycardia, cardiac or cardiopulmonary failure, cardiac congestion.
Number of infants with bradycardia of any severity.
Number of infants with hypotension of any severity.
Number of infants with a renal event like renal impairment, oliguria, anuria, hydronephrosis, nephrocalcinosis and hematuria of any severity.
Number of infants with a serious metabolic event* like hyperkalemia, metabolic acidosis, hyponatremia, hyper- or hypoglycemia.
Number of infants with hyperbilirubinemia of any severity.
Number of infants with retinopathy of prematurity of any severity.
Number of days in hospital per infant.
Weight gain during weeks 2-3 of life.

*Defined as e.g., life-threatening, prolonging hospitalization or causing significant incapacity.

Table 4. Descriptive data for the infants subdivided into those reaching and not reaching SFT

	Reached SFT	Not reached SFT	Total	P
	n=519	n=122	n=641	
Time to SFT (days)				NA
Mean (sd)	18 (11)	NA	18 (11)	
Median	15		15	
Birth weight (g)				ns
Mean (sd)	851 (111)	828 (120)	847 (113)	
Median	860	849	860	
GA at birth (week)				ns
Mean (sd)	27 (2)	27 (2)	27 (2)	
Median	27	27	27	
Apgar score at 5 min				ns
Mean (sd)	7 (2)	7 (2)	7 (2)	
Median	7	7	7	
Feeding intolerance (days)				ns
Mean (sd)	6 (10)	5 (9)	6 (10)	
Median	2	2	2	
Abdominal sign, n (%)	125 (24.1)	25 (20.5)	150 (23.4)	ns
GI event, n (%)	271 (52.2)	70 (57.4)	341 (53.2)	ns
Serious GI event*, n (%)	20 (3.9)	22 (18.0)	42 (6.6)	<0.001
GI perforation, n (%)	10 (1.9)	7 (5.7)	17 (2.7)	0.028
GI obstruction, n (%)	5 (1.0)	8 (6.6)	13 (2.0)	0.001
Necrotizing enterocolitis, n (%)	41 (7.9)	20 (16.4)	61 (9.5)	0.009
Any serious event*, n (%)	97 (18.7)	36 (29.5)	133 (20.7)	0.013
Clinical sepsis, n (%)	90 (17.3)	27 (22.1)	117 (18.3)	ns
Culture positive sepsis, n (%)	56 (10.8)	16 (13.1)	72 (11.2)	ns
Days with antibiotics (n)				0.001
Mean (sd)	10 (24)	20 (29)	12 (25)	
Median	5	11	5	
Pneumonia, n (%)	47 (9.1)	16 (13.1)	63 (9.8)	ns
Serious respiratory event*, n (%)	37 (7.1)	11 (9.0)	48 (7.5)	ns
Bronchopulmonary dysplasia, n (%)	169 (32.6)	35 (28.7)	204 (31.8)	ns
Intracranial hemorrhage, n (%)	126 (24.3)	28 (23.0)	154 (24.0)	ns
Persistent ductus arteriosus, n (%)	194 (37.4)	44 (36.1)	238 (37.1)	ns
Serious cardiac event*, n (%)	5 (1.0)	1 (0.8)	6 (0.9)	ns
Bradycardia, n (%)	67 (12.9)	8 (6.6)	75 (11.7)	ns
Hypotension, n (%)	22 (4.2)	12 (9.8)	34 (5.3)	0.022
Renal event, n (%)	30 (5.8)	10 (8.2)	40 (6.2)	ns
Serious metabolic event*, n (%)	3 (0.6)	1 (0.8)	4 (0.6)	ns
Hyperbilirubinemia, n (%)	43 (8.3)	19 (15.6)	62 (9.7)	0.025
Retinopathy of prematurity, n (%)	168 (32.4)	20 (16.4)	188 (29.3)	<0.001
Days in hospital				0.007
Mean (sd)	79 (20)	84 (18)	80 (20)	
Median	81	84	81	
Weight gain weeks 2-3 (g)				ns
Mean (sd)	115 (52)	113 (57)	115 (53)	
Median	115	106	113	

*Defined as e.g., life-threatening, prolonging hospitalization or causing significant incapacity.
GA, gestational age; GI, gastrointestinal; NA, not applicable; n, number; ns, not significant; P, probability with <0.05 considered significant; sd, standard deviation.

<0.05 were entered into a multivariable model, where the optimal model was determined in a stepwise fashion using the Akaike Information Criterion as the measure of model fit [12]. In the univariable model, the coefficient of determination (R^2) is presented as a measure of the strength of the relationship. R^2-values in the multivariable, stepwise analysis represent a measure on how much of the variability in time to SFT the model explains. In the interpretation of the final model, a p-value <0.05 was considered significant, but due to the exploratory nature of these analyses, all p-values should be interpreted as descriptive.

Results

Frequency of investigated events

GI events occurred in 341 infants (53%) with 42 (6.6%) of them deemed serious (Table 4). Other frequent events included persistent ductus arteriosus (PDA, 238 infants), bronchopulmonary dysplasia (BPD, 204 infants), retinopathy of prematurity (ROP, 188 infants) and intracranial hemorrhage (154 infants). Late onset sepsis was recognized clinically in 18.3% of the infants with positive blood culture in almost two thirds of them. On average, the infants were treated with antibiotics for systemic use for 12 days (i.e., 22.6% of days prior to cGA 34w+6d). In total, 75 infants had reports of bradycardia and 34 infants a hypotensive event. Recorded days with signs of feeding intolerance were few as were the incidences of serious cardiac, renal or metabolic events.

In comparison to the infants reaching SFT, those not reaching SFT during the study period were hospitalized for a longer time (p=0.007) and had significantly higher overall incidences of serious adverse events (p=0.013) (Table 4). This included serious GI events as perforation, obstruction and NEC (p< 0.001-0.028). They received antibiotic treatment for mean 20 days (vs. 10 days, p=0.001) and a higher number of them suffered hypotensive events, ROP and hyperbilirubinemia (p<0.001- 0.025).

Table 5. Output from univariable regressions for infants reaching SFT (n=519)

	Coefficient*	95% CI	P	R-square***
Birth weight (100 g intervals)	-3.04	(-3.89--2.19)	<0.001	8.7
GA (week)	-2.74	(-3.22--2.26)	<0.001	19.4
Apgar score 5 min	-1.03	(-1.61--0.45)	0.001	2.3
Feeding intolerance	-0.05	(-0.15-0.04)	ns	0.2
Abdominal sign	3.37	(1.09-5.65)	0.004	1.6
Gastrointestinal event	1.87	(-0.09-3.83)	ns	0.7
Serious GI event**	8.46	(3.41-13.51)	0.001	2
GI perforation	21.04	(14.12-27.95)	<0.001	6.4
GI obstruction	8.11	(-1.92-18.14)	ns	0.5
Necrotizing enterocolitis	6.75	(3.15-10.34)	<0.001	2.6
Any serious event**	6.75	(4.30-9.20)	<0.001	5.3
Clinical sepsis	5.55	(3.00-8.10)	<0.001	3.4
Culture positive sepsis	5.19	(2.06-8.32)	0.001	2
Days with antibiotics	0.11	(0.07-0.15)	<0.001	4.9
Pneumonia	10.07	(6.76-13.38)	<0.001	6.4
Serious respiratory event**	7.98	(4.23-11.74)	<0.001	3.3
Bronchopulmonary dysplasia	2.43	(0.34-4.51)	0.023	1
Intracranial bleeding	3.77	(1.50-6.03)	0.001	2
Persistent ductus arteriosus	6.17	(4.21-8.13)	<0.001	6.9
Serious cardiac event**	13.56	(3.57-23.55)	0.008	1.4
Bradycardia	1.17	(-1.76-4.10)	ns	0.1
Hypotension	14.38	(9.66-19.09)	<0.001	6.5
Renal event	5.87	(1.69-10.05)	0.006	1.4
Serious metabolic event**	-8.08	(-21.02-4.85)	ns	0.3
Hyperbilirubinemia	3.93	(0.39-7.48)	0.03	0.9
Retinopathy of prematurity	4.87	(2.81-6.92)	<0.001	4
Days in hospital	0.25	(0.21-0.30)	<0.001	20.4
Weight gain (100 g intervals)	-3.79	(-5.64--1.95)	<0.001	3

*Coefficient refers to the number of days difference in time to SFT related to a 1-unit change in the specific variable. **Defined as e.g., life-threatening, prolonging hospitalization or causing significant incapacity. ***R-square depicts the coefficient of determination for the time to SFT.

CI, confidence interval; g, gram; GA, gestational age; GI, gastrointestinal; ns, not significant; P, probability with <0.05 considered statistically

Table 6. Output from stepwise selected multivariable model*

	Coefficient**	95% CI	P
Birth weight (100g intervals)	-0.87	(-1.78-0.04)	0.063
cGA	-0.74	(-1.43--0.04)	0.038
Abdominal sign	2.54	(0.47-4.60)	0.016
Gastrointestinal perforation	15.36	(9.14-21.58)	<0.001
Clinical sepsis	3.46	(1.19-5.72)	0.003
Days with antibiotics	0.04	(0.00-0.08)	0.031
Pneumonia	4.16	(1.04-7.27)	0.009
Serious respiratory event	4.51	(1.10-7.92)	0.01
Persistent ductus arteriosus	1.51	(-0.42-3.44)	0.126
Serious cardiac event	9.01	(0.46-17.56)	0.04
Hypotension	5.73	(1.43-10.04)	0.009
Renal event	2.92	(-0.68-6.52)	0.113
Days in hospital	0.1	(0.04-0.17)	0.001

Model overall R^2, 34.8%

*The full multivariable model is subject to a stepwise procedure where the optimal model is based on the Akaike information criterion (Akaike 1992). **Coefficient refers to the number of days difference in the time to SFT related to a 1-unit change in the specific variable, taken all other variable in the model into account.

CI, confidence interval; g, gram.GA; gestational age; p, probability.

Univariable analysis of the time to SFT

Univariable analysis of infants reaching SFT showed significant association with the time to SFT for many of the explored events (p<0.001- 0.03, Table 5). The numerically greatest effect for events of statistical significance included intestinal perforation (delaying SFT by mean 21.0 days), a hypotensive event, a serious cardiac event and pneumonia (mean delays, 10.1 -14.4 days). Furthermore, a one-day longer treatment with antibiotics for systemic use associated with mean 0.11 days longer time to SFT, a diagnosis of BPD with mean 2.4 days and a week lower GA at birth with mean 2.7 days. The duration of in-hospital stay was strongly dependent on the time SFT (R^2, 20.4), and the coefficient of determination for the time to SFT was noteworthy also for GA, BW, GI perforation, PDA, hypotension, and pneumonia (R^2,6.4-19.4).

Multivariable analysis of the time to SFT

Multivariable analysis after stepwise selection showed maintained association of statistical significance (p<0.001- 0.04) with the time to SFT for GA, the set of abdominal signs, GI perforation, clinical sepsis and days with antibiotics treatment, serious respiratory and cardiac events, pneumonia, and hypotension (Table 6). Numerically a GI perforation, a serious cardiac event and hypotension were associated with the longest increases in the time to SFT (mean delays of 5.7 - 15.4 days). The corresponding values for e.g., pneumonia, clinical sepsis, abdominal signs were 2.5 - 4.2 days and for a day with systemic antibiotics 0.04 days. A one day increase in the time to SFT associated with a 0.1 day longer in-hospital stay. Overall, the multivariable analysis explained 34.8% of the variation in the time to SFT.

Discussion

The 'Connection Trial' analyzing the safety and efficacy of IBP-9414 is the first clinical study of a probiotic under US IND and EU CTX. IBP-9414 is of pharmaceutical grade with quality standards equivalent to drug products. Manufacturing requires full compliance with 'Good Manufacturing Processes' from cell banking to final drug product, which include rigorous controls of raw materials, ingredients and excipients. The requirements also demand verified absence of a range of potential contaminants, batch control, shelf-life determination and validated dosing procedures. This contrasts to all currently available probiotic products that are food additives and quite widely used in the fragile population of premature infants despite their limited verification as regards content, efficacy and safety [13-15].

The currently investigated infants constitute those included in a second safety analysis as per the clinical study protocol agreed with the Health Authorities. This safety analysis involved unblinded data on treatment group allocation restricted to the independent safety committee in order to maintain the integrity of the ongoing study. In total, 90 infants participating in this safety analysis withdrew early from the study. Although these infants may have contributed to the evaluation of the time to SFT, their brief duration in the study compared to those included into the analysis (mean, 25 days vs. 80 days, not shown) may have skewed data especially with respect to conclusions on reasons preventing the attainment of SFT.

The prospective gathering of daily infant data was ended at cGA 34 w + 6 d as per the clinical study protocol, whereby additional events, including reaching SFT may have occurred after the study end. Randomization into the study requires GA of 23 to 32 weeks, and an inverse relationship between time in the study and GA and weight at birth should be expected occur. It also is noteworthy that the recruitment of the infants was extended from BW 750-1000 g down to a lower limit of 500 g only after a first safety analysis had concluded. The BW distribution of the presently investigated infants consequently exceed that expected to occur in an unselected population of surviving infants within the analyzed weight interval. In addition, the clinical study protocol stipulates that infants in extremis and e.g., those with positive blood cultures or recognized GI conditions cannot be randomized into the study. This restriction may have contributed to a

selection towards healthier infants as compared to an unselected preterm infant population. All data examined herein are blinded as to the allocation of infants into the respective treatment arms of the 'Connection Trial', and any conclusion as to the potential effect of L. reuteri awaits unblinded analysis at the study end.

All variables explored for potential influence on the time to SFT were based on reports by the neonatology teams managing the infants. Variation in feeding protocols and diagnosis criteria including the characterization of an event as serious, or not, may thus have occurred between the 80 involved neonatal intensive care units across 10 countries. The variables selected for regression analysis comprised an extension of those previously explored in a sensitivity analysis of a one-day shift in the time to SFT [11]. This extension was selected from almost 500 reported event categories and some of them were grouped together for ease of the output interpretation.

The infants not reaching SFT during the time in the clinical trial had an over-representation of serious adverse events, remained in-hospital and received antibiotics for systemic use for longer periods of time. Serious GI events occurred in almost one fifth of them and these included ileus, intestinal perforation and NEC that were 2 to 6 times more common in these infants than in those reaching SFT. They also had more prevalent periods of hypotension and ROP. In contrast, BW and GA at birth did not appear to be negative predictors of the chance to attain SFT within the investigated period of time.

The 519 infants reaching SFT did so at mean 18 days. This is longer than the time to a full enteral feeding in previous studies of similar, prematurely born cohorts [4, 9,16,17]. This difference likely is a consequence of the currently applied, strict definition of SFT that involves a minimum of 10 consecutive days during which the combined requirement of enteral feedings ≥120 ml/kg/day, no use of parenteral nutrition and an average weight gain of at least 10 g/kg/day must be fulfilled. As an endpoint in clinical trials under IND and CTX, the relevant Health Authorities suggested such a set of variables to increase reliability and reproducibility. A previous analysis substantiated sensitivity of a one-day shift in SFT defined in this way to several clinical events occurring in the very low BW population, which included GI events like NEC, respiratory events, ROP, sepsis and antibiotic treatment days [11].

Univariable analysis revealed several factors associated with the time to SFT. On average, a 100 g lower BW, a week lower GA and a one-point lower 5-min Apgar score at birth extended the time to SFT by one to three days. The effect of a serious GI event, an intestinal perforation and NEC was 7 to 21 days. Intestinal obstruction was rare in the sample reaching SFT and this potentially limited an association of statistical significance. Sepsis, regardless of culture positive or not, pneumonia and serious respiratory events associated with SFT-delays of 5 to 10 days. Other factors of numerically greater impact included serious cardiac events, hypotension and PDA. The duration of in-hospital stay was strongly associated with the time to SFT, and the more important coefficients of determination of the time to SFT included GA, intestinal perforation, PDA, pneumonia and hypotension.

The univariable analyses show the time to SFT without adjustment for the co-occurrence of clinical events. Assuming an infant acquires an intestinal perforation, this would potentially coexist with e.g., sepsis and hypotension, and the expectation would be an average delay to SFT by 3 weeks as well as a considerably longer in-hospital stay. The multivariable analysis is designed to limit this caveat, which explains why e.g., hypotension associated with mean delays to SFT of 14 days and 6 days in the univariable analysis and the multivariable analysis, respectively. Utilizing the multivariable model with a stepwise approach, GI perforation stood out as an event causing the longest delay to SFT (mean, 15 days), followed by a serious cardiac event, hypotension, respiratory events including pneumonia and clinical sepsis. In addition, abdominal signs like distension, discoloration or vomiting, GA at birth, and the duration of antibiotics treatment appeared as independent prolongators of the time to SFT.

The importance of full enteral feeding to the short and longer-term development of the premature infant has been substantiated [1-3]. This analysis of a rather extensive cohort of extremely low BW infants with clinical events gathered under 'Good Clinical Practices' and ICH guidelines shows that not just GI symptoms and complications, but also sepsis, respiratory, cardiac and circulatory events have major impacts on the time to SFT. Several factors have been forwarded as reasons for withholding, slowing down or stopping enteral feeds with the intent to prevent further complications and avoid aggravation of those that exist in preterm infants [4-6]. Some of these factors are beyond the scope of conventional adverse event reporting, which may have contributed to the present multivariable analysis explaining 34.8% of the variation in the time to SFT. The present data details the association of clinical complications to the time to SFT and from the results we suggest that feeding routines commonly applied to the premature infant may benefit from evaluation.

Conflict of Interest Declaration

Scott O Guthrie, MD, is a principal investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Josef Neu, MD, is the global coordinating investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Benedict Doctor, MD, is a principal investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Mobolaji E Famuyide, MD, is a principal investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Ray Hayes, DO, is a principal investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Peter Porcelli, MD, is a principal investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Anthony C Rudine, MD, is a principal investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Carol Wagner, MD, is a principal investigator of the Connection Trial sponsored by Infant Bacterial Therapeutics. Marcus Thuresson is a consultant statistician employed by Infant Bacterial Therapeutics. Anders Kronström, Staffan Strömberg, and Jonas Rastad are all employees of Infant Bacterial Therapeutics sponsoring the Connection Trail.

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Author Contributions

SOG is involved in recruiting infants into the study, in its design and evaluation, and in the authoring of this manuscript; JN is involved in designing, evaluating and coordinating the study and in the authoring of this manuscript; BD is involved in recruiting infants into the study, in its design and evaluation, and in the review of this manuscript; MF is involved in recruiting infants into the study, in its design and evaluation, and in the review of this manuscript; RH is involved in recruiting infants into the study, in its design and evaluation, and in the review of this manuscript; PP is involved in recruiting infants into the study, in its design and evaluation, and in the review of this manuscript; ACR is involved in recruiting infants into the study, in its design and evaluation, and in the review of this manuscript; CW is involved in recruiting infants into the study, in its design and evaluation, and in the review of this manuscript; MT, AK, SS and JR are leading the study design, the data evaluation and in the authoring of this manuscript.

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References

1. Thoene M, Andersson-Berry A. Early enteral nutrition in preterm infants: A narrative review of the nutritional, metabolic and developmental benefits. Nutrients. 2021; 13: 2289-304.
2. Wittwer A, Hascoët J-M. Impact of introducing a standardized nutrition protocol on very premature infants' growth and morbidity. PLOS ONE. 2020; 15: e0232659.doi: 10.1371.
3. Bakker L, Jackson B, Miles A. Oral-feeding guidelines for preterm neonates in the NICU: A scoping review. J Perinatol. 2021; 41: 140-9.
4. Bell EF, Hintz SR, Hansen NI, et al. Mortality, in-hospital morbidity, care practices, and 2-year outcomes for extremely preterm infants in the US, 2013-2018. JAMA. 2022; 327: 1-16.
5. Hay WW. Optimizing nutrition of the preterm infant. Chin J Contemp Pediatr. 2017; 19: 1-21.
6. Young L, Oddie SJ, McGuire W. Delayed introduction of progressive enteral feeds to prevent necrotising enterocolitis in very low birth weight infants. Cochrane Database Syst Rev. 2022; 1: CD001970.
7. Aceti A, Gori D, Barone G, et al. Probiotics and time to achieve full

enteral feeding in human milk-fed and formula-fed preterm infants: Systematic review and meta-analysis. Nutrients. 2016; 8: 471. doi 10.3390/nu8080471.

8. Kreissl A, Sauerzapf E, Repa A, et al. Starting enteral nutrition with preterm single donor milk instead of formula affects time to full enteral feeding in very low birthweight infants. Acta Paediatr. 2017; 106: 1460-7.
9. Baldassarre ME, Di Mauro A, Fanelli M, et al. Shorter time to full preterm feeding using intact protein formula: A randomized controlled trial. Int J Environ Res Public Health. 2019; 16: 2911; doi:10.3390/ijerph16162911.
10. Dorling J, Hewer O, Hurd M, et al. Two speeds of increasing milk feeds for very preterm or very low-birthweight infants: the SIFT RCT. Health Technol Assess. 2020; 24: 1-94.
11. Neu J, Del Moral T, Ferry J, et al. Clinical outcomes correlating to a one-day shift in sustained feeding tolerance in very low birth weight infants in the 'Connection Trial'. Br J Gastroenterol. 2022;4, 255-60.
12. Akaike H. Information theory and an extension of the maximum likelihood principle. Breakthroughs in Statistics. 1992; Vol 1: 610-24.
13. Lewis ZT, Shani G, Masarwe CF, et al. Validating bifidobacterial species and subspecies identity in commercial probiotic products. Pediatr Res. 2016; 79: 445-52.
14. Juber BA, Boly TJ, Pitcher GJ, et al. Routine administration of a multispecies probiotic containing Bifidobacterium and Lactobacillus to very low birth weight infants had no significant impact on the incidence of necrotizing enterocolitis. Frontiers Pediatr. 2021; 9: 1-10.
15. Pointdexter B. Use of probiotics in preterm infants. Pediatrics. 2021; 147: e202151485
16. Salas AA, Li P, Parks K, et al. Early progressive feeding in extremely preterm infants: A randomized trial. Am J Clin Nutr. 2018; 107: 365-70.
17. Wang N, Zang J, Wang B, et al. Transition from parenteral to enteral nutrition and postnatal growth in very preterm infants during their first 28 days of life. Frontiers in Pediatr. 2022; 10:775667. doi: 10.3389/fped.2022.775667.

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