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The Limited versus Extended Trophic Feeding (LET-FEED) Trial: Study Protocol of a Multi-Center Individually Randomized Controlled Clinical Trial

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Study protocol

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Abstract

Background: Nearly 30% of all very preterm (VP; defined as those born before 32 weeks' gestation) newborns develop late onset sepsis (LOS), a life-threatening infection that occurs after 72 hours from birth. Early human milk feeding is associated with reduced risk of LOS. Systematic reviews have reported that the strategy of starting enteral feeds shortly after birth, with advancement in volume before four days from birth, decreases the risk of LOS. However, limited evidence exists to guide advancement before four days. Thus, clinicians are uncertain whether advancing enteral feeds at one versus three days after birth affects outcomes.

Methods: This is a multi-center, open-label, parallel-group, individual randomized controlled trial comparing two different trophic feeding regimens in preterm infants born between 25 0/7 and 31 6/7 weeks of gestation and with birthweights <1500 grams. Neonates will be randomly assigned (1:1 allocation) to either limited trophic feeding (20 to 25 mL/kg birthweight/day for one day) or extended trophic feeding (20 to 25 mL/kg birthweight/day for three days) prior to advancing enteral feeds until full feeding volume (≥140 mL/kg day). The primary outcome is LOS, defined as positive blood, urine, and/or cerebrospinal fluid cultures in the presence of compatible clinical signs of sepsis occurring after 72 hours from birth and before hospital discharge, and treated with antibiotics for ≥ 5 days. Secondary outcomes will include length of hospital stay, all-cause in-hospital mortality, duration of intravenous fluids and central line utilization, necrotizing enterocolitis, severe intraventricular hemorrhage, bronchopulmonary dysplasia, or retinopathy of prematurity requiring intervention. Growth metrics will be evaluated using change in weight, length, and head circumference z-scores from birth to 36 weeks' postmenstrual age.

Discussion: The LET-FEED trial will investigate whether advancement of enteral feeds after 1 or 3 days of trophic feeds leads to improved outcomes without evidence of harm among VP newborns. As the trial is a multi-center clinical trial conducted across geographically diverse regions of the US, the findings will have external validity and generalizability to many settings, increasing the potential for directly actionable results.

Trial Registration: The present study is registered at ClinicalTrials.gov with identifier No: NCT06893939.

Administrative information

Note: The numbers in curly brackets in this protocol refer to SPIRIT checklist item numbers. The order of the items has been modified to group similar items.

Structured Summary {1b}

Title [1]	**The Limited versus Extended Trophic Feeding (LET-FEED) Trial**
Primary Registry and Trial Identifying Number [2] {4}	ClinicalTrials.gov: NCT06893939; https://clinicaltrials.gov/study/NCT06893939; date of registration: 03-11-2025
Secondary Identifying Numbers	Not applicable
Source of Monetary or Material Support	Gerber Foundation
Primary Sponsor and contact information {3b}	Gerber Foundation 4747 West 48th Street, Suite 153, Fremont, MI 49412 231.924.3175
Role of sponsor and funder {3c}	The sponsor and funder does/did not have any role in the design, conduct, analysis, or reporting of this clinical trial.
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Public Title	The Limited versus Extended Trophic Feeding (LET-FEED) Trial
Scientific Title	The Limited versus Extended Trophic Feeding (LET-FEED) Trial
Countries of Recruitment	United States of America

Health Condition(s) or Problem(s) studied	Late onset sepsis; early feeding of very preterm infants
Intervention(s)	One vs three days of trophic enteral feedings
Key inclusion and exclusion criteria	Newborns born between 25 0/7 weeks to 31 6/7 weeks' gestation, have a birthweight of <1500 grams, with parent's consent to feed donor human milk when parent's own milk is not available or of insufficient quantity for clinical needs. Exclusion criteria include: (a) birthweight <5% on the Fenton 2013 growth curve, (b) parent or guardian unable to provide consent within 36 hours of birth, (c) congenital anomaly affecting decisions on enteral feedings, (d) known genetic condition affecting growth, feeding, or mortality, (e) terminal illness requiring limited or withheld support, and (f) any vasopressor use in first 24 hours of birth, excluding hydrocortisone.
Study type	Multi-center, open-label, superiority, parallel-group, individual randomized controlled trial with 1:1 allocation ratio
Date of first enrollment	July 3, 2025
Sample size	N=350
Primary outcome(s)	Late onset sepsis (LOS)
Key secondary outcome(s)	All-cause in-hospital mortality; Length of hospitalization; Necrotizing enterocolitis (NEC; defined as modified Bell's stage IIa or higher); Severe intraventricular hemorrhage (defined as grade III or IV present unilaterally or bilaterally); Bronchopulmonary dysplasia (defined as an oxygen requirement (grade 1), non-invasive distending pressure or positive pressure (grade 2) or invasive positive pressure ventilation at 36 weeks PMA (grade 3); Retinopathy of prematurity requiring intervention; Patent ductus arteriosus requiring intervention (medical or surgical treatment, not including pharmacologic prophylaxis); Number of culture-proven LOS episodes in each participant; Type of LOS (hematogenous, urinary, meningitis); Bacterial species in LOS; Change in z-scores of anthropometric measurements (weight, length, and head circumference) from birth to 36 weeks PMA and/or hospital discharge.
Ethics review {30}	The University of Washington has reviewed this protocol and approved it.
Individual trial participant data sharing statement	Deidentified individual clinical trial participant-level data will be shared to investigators who reach out to Dr. Valentine with a study question, and IRB-approved study, after approval by the LET-FEED trial oversight committee.

Protocol version {2}

March 26, 2025: Version 1

Introduction

Background and rationale {9a}

Up to 30% of all very preterm (VP; defined as those born before 32 weeks' gestation) newborns born in the United States develop late onset sepsis (LOS), a life-threatening infection that occurs after 72 hours from birth.[3,4] LOS is a leading cause of morbidity in all VP newborns. Specifically, VP newborns who develop LOS have significantly lower survival rates (adjusted risk ratio [aRR] 0.89, 95% confidence interval [CI] 0.87-0.90), increased risk of being discharged on oxygen (aRR 1.32, 95% CI 1.26-1.38), increased risk of needing a tracheostomy (aRR 2.88, 95% CI 2.47-3.37), and increased risk of requiring a gastrostomy tube (aRR 2.09, 95% CI 1.93-2.57).[5] Thus, clinical practices that can reduce LOS are critically important to ensure VP newborns not only survive, but thrive, living lives free of disability or long-term impairment.

One of the most protective factors in preventing LOS is providing human milk-based feeds (prioritizing the use of parent's own milk [POM] supplemented with pasteurized donor human milk).[6,7] Common clinical practice for VP newborns includes initiation of human milk-based enteral feeds at approximately 20 mL/kg birthweight/day, which clinicians routinely categorize as "trophic feeds", with subsequent progressive advancement of enteral feeds daily until reaching a sufficient quantity, termed "full feeds", that achieves adequate hydration and meets nutritional needs to support optimal growth.[6,7] While enteral feeds are being advanced from trophic feeds to full feeds, VP newborns receive additional fluids and nutrition via intravenous (IV) infusions. Yet, any indwelling catheter to provide IV nutrition increases the risk of LOS because the catheter breaches the newborn's protective skin barrier, thereby increasing risk of invasive infection from skin microbes.[8,9] It is therefore not surprising that the most common bacterial causes of LOS are skin flora, predominantly *Staphylococcus aureus* and coagulase-negative Staphylococcus.[5]

Early initiation of enteral feeds may reduce the duration of IV fluid administration. Historically, clinicians have been conservative in deciding when to advance from trophic feeds. There is a paucity of data on optimal advancement strategies for enteral feeds in VP newborns, leading to uncertainty about how to provide optimal nutrition during the critical fetal-to-neonatal transition over the first week after birth. However, while non-definitive, the few published studies on this topic have supported the notion that earlier advancement of enteral feeding from trophic feeds may limit the duration of peripheral and central catheter use and concurrently reduce the risk of LOS.[8-11] Furthermore, the intestinal tract's defense function is enhanced by early exclusive human milk feeding, leading to a favorable intestinal microbiome and enhanced barrier function against bacterial invasion and a decreased risk of LOS.[6,7,12-14] By preventing the growth of harmful, pathogenic, sepsis-causing bacteria within the gut of VP newborns, these newborns may have lower risk of developing LOS than similar newborns fed with slower enteral feeding advancement strategies.[6,7]

However, there also exist concerns that too much enteral feeding or too fast progression of enteral feeding may increase the risk of adverse intestinal morbidities such as necrotizing enterocolitis (NEC),[15]

a devastating condition associated with 30-40% mortality.[16] However, significant concerns about enteral feeding and risk of NEC were particularly heightened during the 1990s to early 2000s in which a lack of exclusive human milk-based diet (including the use of formula) was common practice in feeding preterm newborns.[17-19] With the increase in pasteurized donor human milk and the promotion of exclusive human milk-based feeding practices since then, more recent literature has demonstrated that delayed initiation of enteral feeds or slower progression of enteral feeds are unlikely to be protective against NEC, as once believed.[7,17,20,21] Yet, clinical concerns for increased risk of NEC persist due to limited clinical trials on enteral feeding advancement in the vulnerable, very preterm population.

Explanation for the choice of comparator {9b}

Previously we conducted a pilot randomized trial that compared early versus delayed progressive feeding in 60 extremely preterm infants (<28 weeks) born at the University of Alabama at Birmingham Hospital in which we observed that early progressive feeding (also known as limited trophic feeding) reduced the duration and/or need central venous access (p=0.0001) and parenteral nutrition by four days (p=0.0005).[3] Culture-proven sepsis (10% vs. 27%; p= 0.18) was lower in the early progressive feeding group but did not reach statistical significance, likely due to the limited power of the small sample size in this preliminary study.[3]

The natural question is whether these findings are generalizable. Specifically, does early progression of enteral feeds in a multi-center trial improve clinical outcomes, namely prevention of LOS, without an increased risk of other adverse morbidities, such as necrotizing enterocolitis or mortality? The lack of multi-center clinical trials evaluating earlier advancement of enteral feeds to full volume feeds has left clinicians with uncertainty about the risks and benefits of the early advancement of feeding approach that undoubtedly leads to inconsistent clinical practices for feeding premature newborns. The uncertainty in feeding strategies has led to significant differences in how VP newborns are fed in the US and globally, with some centers initiating and advancing feeds 24 hours after initiation of enteral feeds and others continuing small volume trophic feeds without any advancement until day four after birth. Thus, clinicians caring for VP newborns need evidence in this vulnerable population to determine optimal feeding advancement strategies to reduce the risk of both short- and long-term adverse outcomes.

Objectives {10}

The objective of this study is to determine the ideal feeding strategy for enteral feeding advancement in very premature neonates between 25 0/7 weeks to 31 6/7 weeks gestation. We hypothesize that in infants in this range of gestations, advancing enteral feeds after 24 hours from birth (limited trophic feeds) has a lower risk of all cause LOS than when feeds are advanced after 72 hours (extended trophic feeds) with no difference in the risk of adverse outcomes. In the limited versus extended trophic feeding (LET-FEED) trial, we purposefully did not include infants who are periviable, but we anticipate a future similar trial pending the LET-FEED trial's results.

For the purposes of this trial, the primary outcome of LOS will be defined as a positive blood, urine and/or cerebrospinal fluid (CSF) culture, obtained in the presence of clinical signs compatible with sepsis, occurring after 72 hours from birth and before hospital discharge, and treated with antibiotics for 5 days or more. While previous publications have considered LOS to include blood or CSF culture positivity after 72 hours from birth and if ≥5 days of antibiotics,[5] studies have also demonstrated a significant association between duration of central lines and parenteral nutrition with urinary tract infections.[22,23] Thus, we elected to include urine culture positivity for a more comprehensive evaluation of LOS. Secondary outcomes will include length of hospitalization, all-cause in-hospital mortality, necrotizing enterocolitis (Bell's stage IIA or higher)[24,25] severe intraventricular hemorrhage (grade III or IV either unilaterally or bilaterally)[26], bronchopulmonary dysplasia (oxygen requirement, non-invasive distending pressure, or invasive positive pressure ventilation at 36 weeks [PMA] age)[27], or retinopathy of prematurity requiring intervention.

Additionally, we will assess the following secondary objectives:

1. The impact of limited versus extended trophic feedings on duration of IV fluid administration which is usually for parenteral nutrition and catheter usage between groups by measuring: (a) duration of IV fluids within the first two weeks after birth, (b) duration of IV fluids throughout hospitalization, (c) time to reach enteral feeds commonly accepted as sufficient to discontinue parenteral nutrition or IV fluids (defined as ≥120 mL/kg/day), and (d) days of central line utilization.

2. The impact of limited versus extended trophic feeds on growth evaluating growth metrics from birth to discharge by determining changes in Z-scores from birth to 36 weeks' post menstrual age in weight, length, and head circumference.

Methods: Patient and public involvement, and trial design

Patient and public involvement {11}

Patient and public involvement was not involved in the design, conduct, or reporting of this trial.

Trial design {12}

This is a multi-center, open-label, superiority, parallel-group, individual randomized controlled trial comparing two different trophic feeding regimens in preterm neonates born between 25 0/7 and 31 6/7 weeks of gestation and with a birthweight <1500 grams. These infants will be randomly assigned to either one of two groups: (1) limited trophic feeding (20 to 25 mL/kg birthweight/day for one day) or (2) extended trophic feeding (20 to 25 mL/kg birthweight/day for three days) prior to advancing enteral feeds until full feeding volume (of at least 140 mL/kg /day) is achieved.

Randomization

Participants will be randomly assigned to one of the study groups following computer generated random-block sequences at the individual level. The study's biostatistician at the University of Washington will develop the sequence and implement stratified randomization by gestational age (25 0/7 weeks – 27 6/7 weeks' gestation and 28 0/7 weeks -31 6/7 weeks' gestation) and site using a computerized central telephone system. Multiple gestation newborns will be randomized individually.

Methods: Participants, interventions and outcomes

Trial setting {13}

We seek to enroll N=350 VP infants born at 25 0/7 to 31 6/7 weeks' gestation who are also <1500-gram birthweight admitted to the five participating neonatal intensive care units. The participating centers include: (1) St. Joseph's Medical Center (Tacoma, WA), (2) Texas Children's Hospital (Houston, TX), (3) University of Alabama (Birmingham, AL), (4) University of South Florida (Tampa, FL), and (5) University of Oklahoma (Oklahoma City, OK). Of note, the study may expand to other sites if needed to enhance recruitment during the conduct of the trial.

Characteristics of the people are needed for the trial

See PRO EDI (Equity, Diversity and Inclusion) Table[28] for further information on information pertaining to characteristics of included participants (**Table 1**).

Table 1: Pro EDI Table	
Item	**Explanation**
Age	**All included participants will be preterm neonates at 25w0d-31w6d and enrolled and randomized within 36 hours of delivery at the latest.** We will report age at enrolment.
Sex	**All sexes are included. If a neonate is born with ambiguous genitalia, they can be enrolled as long as they do not meet any exclusion criteria.** We will report sex at birth.
Gender	**As all participants are neonates, we will utilize sex at birth but not gender to classify participants.** We will report sex at birth.
Sexual identity	**As all participants are neonates, this is not applicable.** Not applicable
Race, ethnicity and ancestry	**We will include participants regardless of race, ethnicity, and ancestry.** We will evaluate race, ethnicity, and ancestry through the following categories: Black, White, Asian, Native American, Pacific Islander, Other, Unknown (Participant families, preferably the mother, can select all that apply) We will have an additional question asking if they are of Hispanic or Latino ethnicity
Socio-economic status (SES)	**We will include participants regardless of socioeconomic status.** We will evaluate socioeconomic status through these specific categories: Type of insurance (Private, Medicaid, lack of insurance)
Level of education	**As all participants are neonates, this is not applicable.** Not applicable
Disability	**As all participants are neonates, this is not applicable.** Not applicable
Location (country/ countries of data collection and site coordination)	**We will recruit at the following sites, with the potential for expansion to other sites if recruitment efforts are needed to expand:** University of Washington (Seattle, Washington) St. Joseph Medical Center (Tacoma, Washington)

	Baylor College of Medicine (Houston, Texas) University of South Florida (Tampa, Florida) University of Alabama at Birmingham (Birmingham, Alabama) University of Oklahoma (Oklahoma City, Oklahoma)

Eligibility criteria for participants {14a}

To be eligible for the LET-FEED trial, newborns must be born between the gestational age of 25 0/7 weeks to 31 6/7 weeks' gestation, have a birthweight of <1500 grams, with parent's consent to feed donor human milk when parent's own milk is not available or of insufficient quantity for clinical needs. All newborns will be eligible for this study without regards to gender, ethnicity, race, socioeconomic status, or language spoken. Exclusion criteria include: (a) birthweight <5% on the Fenton 2013 growth curve, (b) parent or guardian unable to provide consent within 36 hours of birth, (c) congenital anomaly affecting decisions on enteral feedings, (d) known genetic condition affecting growth, feeding, or mortality, (e) terminal illness requiring limited or withheld support, and (f) any vasopressor use in first 24 hours of birth, excluding hydrocortisone.

Eligibility criteria for sites and those delivering interventions {14b}

Eligible sites must be able to care for very preterm infants meeting the eligibility requirements for this trial and monitor them through discharge. They must have access to donor human milk and be willing to provide both feeding regimens as outlined in this trial.

Who will take informed consent? {32a}

To ensure timely enrollment, written informed consent will be obtained no later than 36 hours following birth, with a goal of enrollment prenatally or within 24 hours of birth. Consent may occur prenatally among pregnant patients at high risk of preterm delivery within the gestational age window of eligibility for the LET-FEED trial. Identification of potential participants will prompt a visit from a study team member who has completed human subjects training, who will meet with the parent(s) in their room or the baby's room to explain the study. During this consent process, the risks and benefits associated with participation will be thoroughly discussed, and ample time will be allocated for the parents to ask any questions they may have. The consent will include that their baby will be fed parent's own milk as the prioritized enteral feed with pasteurized donor milk as the alternative if parent's own milk (POM) is of insufficient quantity.

Importantly, participants will be assured that their infant will receive all necessary treatments regardless of their involvement in the study. Upon obtaining consent, randomization will determine the assignment of each participant to their study group.

Additional consent provisions for collection and use of participant data and biological specimens {32b}

Consenting to participation in the LET-FEED trial also includes an optional consent to have infant stool samples collected from the participating infant. These biospecimens will be biobanked for future analyses exploring potential mechanisms or pathways related to early feeding practices and the developing gut microbiome.

Interventions

Intervention and comparator description {15a}

All enrolled newborns will be randomly assigned to receive either limited trophic feeds (24 hours) or extended trophic feeds (72 hours) prior to the advancement of enteral feeds (see **Table 1**). Trophic feeds are recommended to start within 6-12 hours of birth. Because IV fluids and/or parenteral nutrition need to be ordered up to 12 hours prior to the implemented change, we have included a 12-hour period of advancement (24 to 36 hours versus 72 to 84 hours after birth) to account for these operational logistics and ensure the generalizability of this pragmatic trial's results. Feeding volumes will be advanced by 25 to 30 mL/kg/day until full feeding volumes (≥140 mL/kg/day) are reached. Infants will be fed POM or donor human milk through 34 weeks PMA. After 34 weeks, the supplementation of donor milk will cease and preterm formula will be introduced as an alternative, following the practices established at each study site.

The mL/kg birthweight/day for determining enteral feeding volume will continue until day 7 after birth at which time it may become mL/kg/day using a higher weight than birthweight if it occurs on day 8 after birth. Additionally, if there is a concern about the initial birthweight being incorrect, the team will discuss with the site PI to determine if a different value for the birthweight should be used instead. If this occurs, the correction will be documented on the case report forms accordingly.

All participating centers will provide POM as the prioritized feed, with donor human milk provided in cases where POM is not available in sufficient quantities. Preterm formula will not be used until at least 34 weeks PMA. The guidelines for this trial are that fortification occurs once enteral feeding volumes of 40 to 60 mL/kg birthweight/day or higher are achieved and that IV catheters will be removed once the newborn is above 120 mL/kg birthweight/day of enteral feeds. Feeds will be advanced on days of fortification.

Table 2: Feeding Plan by Group*		
	Limited Trophic Feeds: **(≤24-36 hours of Trophic Feeds)**	**Extended Trophic Feeds:** **(72 hours of Trophic Feeds)**
Day 0 **(Day of birth)**	20-25 mL/kg birthweight/day	20-25 mL/kg birthweight/day
Day 1	50 mL/kg birthweight/day (at ~24-36 hours)	20-25 mL/kg birthweight/day
Day 2	80 mL/kg birthweight/day	20-25 mL/kg birthweight/day
Day 3	110 mL/kg birthweight/day	20-25 mL/kg birthweight/day (at ~72-84 hours)
Day 4	140 mL/kg birthweight/day	50 mL/kg birthweight/day
Day 5	140-160 mL/kg birthweight/day	80 mL/kg birthweight/day
Day 6	140-160 mL/kg birthweight/day	110 mL/kg birthweight/day
Day 7	140-160 mL/kg birthweight/day	140 mL/kg birthweight/day
Day 8	140-160 mL/kg birthweight/day	140-160 mL/kg birthweight/day
*Advancement will be by approximately 30 mL/kg birthweight/day for every day after the trophic feeding phase. Enteral feeds are recommended to begin in all babies within 6-12 hours from birth.		

Criteria for discontinuing or modifying allocated interventions {15b}

Deviations from the feeding protocol will be allowed if signs of feeding intolerance are present before establishment of full enteral feeding. Feeding intolerance will be defined as an interruption or cessation of enteral feeds for a period *greater than 12 hours for abnormal abdominal examination within the feeding advancement period (through day 8 as per Table 2)*. Information will also be collected on the percentage of enteral feeds that had a >50% reduction in planned volume over each 24-hour period to serve as an additional metric of feeding intolerance.

Strategies to improve adherence to interventions {15c}

The study team will create an enteral feeding document for each participant that will be at the bedside to help guide the nursing staff and clinical team on the enteral feeding volume prescribed daily based on the feeding group assignment. Additionally, the research coordinators and site PIs will check in daily with the clinical team to ensure that the feeding plan is clear to the clinical team and is being followed.

Relevant concomitant care permitted or prohibited during the trial {15d}

During the LET-FEED trial, aside from enteral feeding practices, all other care will be conducted as indicated clinically. Thus, concomitant care will be permitted unless it interferes with the early feeding practices as delineated in Table 2.

Ancillary and post-trial care {34}

There are no specific provisions for post-discharge care other than current routine standard practice and follow-up as per the current site-specific standards and guidelines. However, future studies may evaluate the neurodevelopmental and long-term outcomes of the children born in this study.

Outcomes {16}

The primary outcome will be culture-proven LOS, defined as occurring after 72 hours from birth and prior to discharge with a positive blood, urine, and/or cerebrospinal fluid culture in the presence of compatible clinical signs of sepsis, in which treatment with antibiotics is done for at least 5 days.

Secondary outcomes include the following:

- All-cause in-hospital mortality
- Duration of parenteral nutrition
- Duration of central venous access
- Duration of IV access
- Length of hospitalization
- Time to reach full enteral feeding volumes (≥120 mL/kg birthweight/day)
- Necrotizing enterocolitis (NEC; defined as modified Bell's stage IIa or higher[24,25])
- Spontaneous intestinal perforation (SIP)
- Severe intraventricular hemorrhage (defined as grade III or IV present unilaterally or bilaterally)
- Bronchopulmonary dysplasia (defined as an oxygen requirement (grade 1), non-invasive distending pressure or positive pressure (grade 2) or invasive positive pressure ventilation at 36 weeks PMA (grade 3). The grades of BPD are defined as per a recent publication from Jensen et al. and the neonatal research network.[27]
- Retinopathy of prematurity requiring intervention
- Patent ductus arteriosus requiring intervention (medical or surgical treatment, not including pharmacologic prophylaxis)
- Number of culture-proven LOS episodes in each participant
- Type of LOS (hematogenous, urinary, meningitis)
- Bacterial species in LOS

- Change in z-scores of anthropometric measurements (weight, length, and head circumference) from birth to 36 weeks PMA and/or hospital discharge. Anthropometric z-scores will be calculated using Fenton 2013 growth curves.
- Length and head circumference measurements will occur weekly throughout study duration following birth.

Potential Harms {17}

Definition of Adverse Events (AE)

In the LET-FEED trial, an adverse event will be defined as any untoward medical occurrence associated with the use of an intervention in humans, whether or not considered related directly to the study intervention (21 CFR 312.32 (a)).

Definition of Serious Adverse Events (SAE)

An AE or suspected adverse reaction will be considered "serious" if, in the view of either the investigator or sponsor, it results in any of the following outcomes: death, a life-threatening adverse event, prolongation of existing hospitalization, a persistent or significant incapacity, or substantial disruption of the ability to conduct normal life functions. Important medical events that may not result in death, be life-threatening, or require prolonged hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the participant and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

Classification of an Adverse Event

Severity of Event

For AEs not included in the protocol-defined grading system, the following guidelines will be used to describe severity. Mild event will be classified as any event requiring minimal or no treatment and do not interfere with the participant’s daily activities. A moderate AE will be defined as any event resulting in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning. A severe AE will be defined as any event interrupting a participant’s usual daily activity and may require otherwise unanticipated systemic drug therapy or other treatment. Of note, the term “severe” does not necessarily equate to “serious”.

Relationship to Study Intervention

All AEs must have their relationship to the study intervention assessed by the site PIs and Co-PIs, who will examine and evaluate the participant based on the temporal relationship and his/her clinical judgment. The degree of certainty about causality will be graded using the categories below. In a clinical trial, the study intervention must always be suspect. An AE will be considered related to the study intervention if the AE is known to occur with the study intervention, there is a reasonable possibility that

the study intervention caused the AE, or there is a temporal relationship between the study intervention and event. Reasonable possibility means that there is evidence to suggest a causal relationship between the study intervention and the AE.

An AE will be considered not related to the intervention (feeding advancement of either limited or extended trophic feedings) if there is not a reasonable possibility that the administration of the study intervention caused the event, there is no temporal relationship between the study intervention and event onset, or an alternate etiology has been established.

Expectedness

The Co-PIs, in conjunction with the site PIs and DSMB, will be responsible for determining whether an AE is expected or unexpected. An AE will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the study intervention.

Time Period and Frequency for Event Assessment and Follow-Up

The occurrence of an AE or SAE may come to the attention of study personnel during the study visit(s) or upon review by a study monitor. All AEs including local and systemic reactions not meeting the criteria for SAEs will be captured on the appropriate case report form (CRF). Any medical condition that is present at the time that the participant is screened will be considered as baseline and not reported as an AE. The Research Coordinator will record all reportable events with start dates occurring any time after informed consent is obtained until the last day of study participation. Throughout the LET-FEED trial, a member of the research staff will inquire about the occurrence of AE/SAEs each hospitalized day. Events will be followed for outcome information until resolution or stabilization.

Participant timeline {18}

The participants will be consented in the prenatal period to no later than 36 hours after birth, with ideal timing being prior to delivery. They will be randomized to receive either limited (24-36 hours) or extended (72-84 hours) trophic enteral feeds, where trophic feeds are defined as 20-25 mL/kg birthweight/day POM or donor human milk. Advancement of enteral feeds will be approximately 30 mL/kg birthweight/day for each subsequent day until reaching a goal of at least 140 mL/kg birthweight/day. They will continue in the LET-FEED trial until discharge from the neonatal intensive care unit (NICU).

Sample size {19}

In our preliminary pilot trial, culture-proven LOS was reported in 10% of the early progressive feeding group and 27% of the delayed progressive feeding group, p=0.18.[3] Because the incidence of LOS may be different between centers, we have elected for 175 per group to provide 80% power to detect an absolute 11% difference in culture-positive LOS between groups. With these numbers, we will have 99% power to detect the 17% difference in the incidence of culture-proven LOS reported in the pilot trial.[3]

Recruitment {20}

All pregnant individuals who are at risk for preterm delivery between 25 0/7 weeks to 31 6/7 weeks' gestation will be approached if they meet the eligibility requirements to be in the LET-FEED trial. We will also enroll within 36 hours of birth for any newborn born during this gestational age window in which mothers were not approached about the study prior to delivery.

Assignment of interventions: randomization

Sequence generation: who will generate the sequence {21a}

The study statistician will generate the randomization sequence.

Sequence generation: type of randomization {21b}

Randomization to limited or extended trophic feeding groups will be via a central randomization sequence generated by the Research Electronic Data Capture (REDCap) database.[29]

Allocation concealment mechanism {22}

This will be an open-label trial as the feeding intervention cannot be masked.

Implementation {23}

This trial will compare the limited versus extended trophic feeding under usual clinical circumstances with no effort to strictly control interventions other than feeding volume targets. Therefore, all clinical care will be conducted at the clinician's discretion other than the feeding advancement rate as delineated in this protocol.

Transfer for Convalescent Care: While uncommon, if a study participant is transferred to an outside hospital for convalescent care prior to 36 weeks PMA, our study team will contact the family weekly until the newborn is discharged to collect the necessary study outcome measures.

Lost to Follow Up (LTFU): This study is not prone to LTFU as we are specifically focusing on VP neonates during their initial hospitalization.

Competing Studies: Neonates can be enrolled in this trial and other trials as this trial will be individually randomized, thus not biasing the results. However, the one exception is if a separate clinical trial or study is evaluating different feeding or fluid management decisions within the first two weeks after birth, and if so, the neonate cannot participate in both studies.

Assignment of interventions: Blinding

Who will be blinded {24a}

Caregivers and primary outcome evaluators will not be masked given that the intervention is a change in feeding quantities that cannot be blinded.

How will blinding be achieved {24b}

Not applicable.

Procedure for unblinding if needed {24c}

Not applicable

Data collection and management

Plans for assessment and collection of outcomes {25a}

All enrolled newborns will be assessed daily by the clinical team in the NICU and provided the feeding advancement plan as randomized. For the primary outcome of LOS, clinical evaluation for sepsis will occur per discretion of the clinical neonatologist. Every instance of an evaluation for LOS will require an additional form be filled out for the LET-FEED trial purposes to monitor any occurrence of LOS. Each instance will include the date the sepsis evaluation was initiated, total number of days of antibiotic administration, and whether urine, blood and/or CSF were obtained and if any were positive. If any cultures return positive, what pathogen was identified will be recorded.

For the secondary outcomes, a daily log will be completed by the research staff that will include all secondary outcome data. Specifically, each day of hospitalization for the first 28 days will require study staff to document daily nutrition information, any peripheral IVs inserted and their associated complications, any central catheters inserted and their associated complications, timing and type of fortification, daily weight, whether a blood culture was obtained, any antibiotics prescribed and if so, type of antibiotic, supplemental oxygen and/or other forms of respiratory support, volume of donor human milk, volume of parent's own milk, volume of parenteral nutrition with amino acid and lipids, volume of additional IV fluids, volume of blood products, volume of IV medications. If any protocol violations occur, these will be included in the protocol violation case report form. Such violations may include if a participant's enteral feeds are held due to concerns for necrotizing enterocolitis or ileus. If such event occurs, feeds will be restarted per discretion of clinical team after discussions with the site PI. We will additionally obtain duration of IV fluids administered in the first 14 days and for the entire hospitalization, which will be further stratified by type of IV infusion (e.g. PIV, peripherally inserted central catheter (PICC), peripheral arterial line (PAL), umbilical venous catheter (UVC), umbilical arterial catheter (UAC). We will also collect information on any IV infiltration or extravasation injuries during hospitalization, and, if so, the total number of infiltration or extravasation injuries. Anthropometric values including height (cm and Z-score), weight (cm and Z-score), and head circumference (cm and Z-score) will be obtained at days 14, 21, 28, 35, 42, 49, 32 weeks PMA, 36 weeks PMA, and discharge using Fenton 2013 growth curve. The diet at 36 weeks PMA and discharge will be entered as either exclusive human milk, mixed feeds (human milk and formula), or formula only. Finally, all comorbidities will be evaluated and, if

present, date of diagnosis. See **Figure 1** for the Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) figure of overall LET-FEED trial-related activities and time points of assessment.

Plans to promote participant retention and complete follow-up {25b}

It is highly unlikely we will not meet enrollment numbers given that we are recruiting 350 VP newborns across five institutions throughout the United States. All participating centers were evaluated for the number of patients that would meet the criteria for enrollment historically and the data indicated that enrollment above 30 percent of eligible patients would meet the enrollment goal. Of note, if one institution has limited enrollment, we will seek to increase enrollment numbers at the other institutions to account for this difference. We will continually monitor enrollment numbers and have monthly study meetings to discuss any issues with the study and problem solve solutions to overcome unforeseen issues. The data and safety monitoring board (DSMB) will provide oversight and direction for the conduct, results, and interim findings of this study.

Data management {26}

Source Documentation

All source documents with participant study-related data (i.e., “clinical metadata”) will be stored as paper records and considered the study source documents. All source documents will be maintained as hard copies, converted to electronic copies, and secured in a manner accordant with the institutional review board (IRB) protocol. Data will be handled in accordance with Good Clinical Practices, U.S. federal regulations, local regulations, and instructions from the Gerber Foundation.

Case Report Forms (CRFs)

Data will be entered manually onto paper CRFs and then into an online, encrypted REDCap database developed and maintained by study investigators at the University of Washington and each participating site. Data entry should be current to within 72 hours of the last visit for each study participant unless otherwise specified.

Data Review and Quality

The site Principal Investigator (PI) and/or staff will review data for quality and provide quality assurance reports to ensure that study data entered into the REDCap database match the physical charts and are all study data collections completed in a timely manner. Quality assurance reports will include, but are not limited to, the following: missing forms, missing and out of range values, automated data queries, and targeted manual reviews of study data. Data queries must be addressed within 1 week unless otherwise specified.

Data verification, auditing, and adjudication:

Verification: All data inputted into the online database will be verified with the paper charts. The study team members, including the local research coordinators, working with the site PIs and local research team will perform these verifications.

Auditing and Adjudication: The LET-FEED trial site PIs and research coordinators will evaluate 20% of the assessments in physical charts to compare the information to that entered into the online database. These audits and adjudication process will ensure all data recorded in the online, encrypted database are reliable and accurate. If discrepancies in the data are identified, the data in the LET-FEED trial database will be corrected based upon this adjudication process.

Protocol violations: Any protocol violations during the feeding advancement will be documented in the CRFs with a description of what occurred and subsequent remedies to the violation, if any.

Archiving of Data

Records and documents pertaining to the conduct of this study, including CRFs, source documents, consent forms, and laboratory test results, will be retained by the investigator for a minimum of seven years after study completion. Electronic data will include clinical information and cluster allocation. No study records will be destroyed without prior authorization from the study's lead co-PIs and the Gerber Foundation.

Confidentiality {33}

Confidentiality of data will be protected through careful training and close supervision of project staff, the requirement that all staff sign oaths of confidentiality, the maintenance of locked files for data, and the use of restricted passwords for computer access to the study data. Study IDs will be generated for this study which will not include identifying information other than the date of birth. Health history and other identifying information will be kept in locked file cabinets at each clinical site. Electronic files are stored on a secure, encrypted server and paper files are stored in a locked room. The data will only be made available to individuals conducting the proposed research when it is necessary for the safe and accurate completion of the investigation. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the trial's Co-PIs.

Plans for collection, laboratory evaluation and storage of biological specimens for genetic or molecular analysis in this trial/future use {33}

Stool samples to assess the intestinal microbiome of the newborns will be collected and stored in a -80°C freezer on site at the participating centers. The collection of these specimens is part of an ad hoc study and consent will be obtained in addition to primary study. It is permissible to obtain consent for the LET-FEED clinical study and for the consenters to withhold consent for the stool microbiome study. If consent is obtained for microbiome assessment, four stool specimens will be collected and include first stool before 7 days, at 7 and 14 days after birth and at 36 weeks postmenstrual age or immediately before discharge whichever comes first. These biospecimens will be transferred to the University of

Washington for storage in -80°C freezers using commercial services such as provided by Biocare (or similar company) to preserve the cold chain and prevent freeze-thaw cycles that could compromise the integrity of the samples. These biospecimens will biobanked as a source for important future research, primarily studies evaluating the effect of early feeding strategies on intestinal microbiome and the potentially related outcomes assessed in this trial.

Statistical methods

Statistical methods for primary and secondary outcomes {27a}

The effect size for the primary efficacy endpoint (all-cause LOS) will be expressed as a relative risk with 95% confidence intervals. This approach is similar to the one used in the pilot trial. Adjusted mean differences with 95% confidence intervals will also be reported. The adjusted analysis for evaluating the risk of culture-positive LOS events will be conducted using generalized estimating equations (GEE) relative risk regression with robust standard errors.[30,31]

Secondary outcomes (all-cause mortality, length of hospitalization, ROP requiring treatment, NEC modified Bell's stage IIa or higher, PDA requiring treatment, BPD, any IVH, severe IVH, among the others as aforementioned) will be analyzed in a similar manner to the primary outcome.

Using an intent-to-treat approach, duration of IV fluids within the first two weeks after birth, duration of IV fluids throughout hospitalization (in days), time to reach full enteral feeds (in days), and days of IV catheter utilization will be analyzed as continuous outcomes using linear GEE regression by treatment group. Sensitivity analyses will include adjustment for other factors and interactions that may affect these outcomes including institution of enrollment, birthweight z-score, gestational age at birth, exposure to antenatal corticosteroids, multiples/singleton status, need for vasopressors after 24 hours, type of respiratory support (invasive compared to non-invasive), and days of antibiotic administration after 48 hours from birth.

Changes in Z-score will be explored using linear GEE models. We will evaluate each outcome in relation to the others, for instance how changes in Z-score for length, head circumference, or weight as a result of feeding regimen group are related to other assessed metrics such as occurrence of sepsis, duration of IV fluids, number of IV attempts throughout hospitalization, among others. These analyses will include additional regression models and graphical network analyses. Non-linear associations may be explored through polynomial and/or spline basis expansion of the variables of interest, or machine-learning based approaches such as regression tree models.

All efficacy and safety endpoints of the trial will be analyzed with the intention-to-treat principle using SAS 9.4 and/or R.

Safety Analyses

Interim safety outcomes include incidence of mortality up through 36 weeks PMA, NEC, and SIP. The DSMB may examine other outcomes at its discretion. To protect patient safety, less evidence will be required to stop the trial for harm than benefit. Thus, formal statistical testing for an imbalance in mortality or NEC/SIP in either treatment arm at 36 weeks PMA will be performed at the first interim safety data analysis to guard against false positives while allowing for stopping at reasonable levels of evidence. Thus, at each interim safety data analysis, an increased incidence of death/NEC/SIP in either treatment group with $p < 0.01$ will be considered as statistically significant evidence of harm that the DSMB may use to recommend suspension of the trial for safety reasons. In addition to SAEs, the DSMB will examine other safety outcomes, including all reported serious adverse events by treatment group in considering a recommendation to suspend the trial for safety reasons.

Who will be included in each analysis {27b}

We will include all participants within the analyses unless otherwise prespecified or if a participant has withdrawn from the study and requested to not provide their data for analyses.

Methods in analysis to handle protocol non-adherence and any statistical methods to handle missing data {27c}

Because of the design of the study to be completed within the initial NICU hospitalization, missing data will be minimal. Missing data patterns will be assessed and evaluated for missingness at random and missing at random and multiple imputation techniques may be employed if appropriate.

Methods for additional analyses (e.g. subgroup analyses) {27d}

Additional a priori determined additional analyses include the following:

- Evaluation of outcomes (both primary and secondary) per stratified analyses in gestational age categories: (a) 25 0/7 weeks – 27 6/7 weeks' gestation and (b) 28 0/7 weeks – 31 6/7 weeks' gestation. These will be performed using similar methodology as described in the primary and secondary outcome statistical methods above.
- Evaluation of outcomes (both primary and secondary) per stratified analyses in birthweight categories: (a) <1000g and (b) 1000-1499g. These will be performed using similar methodology as described in the primary and secondary outcome statistical methods above.

Interim analyses {28b}

The DSMB will review an interim efficacy analysis of the trial once 50% of the participants (n=175) are enrolled and completed the trial. The DSMB will have overall responsibility for interim data monitoring. Treatment groups will be compared statistically using the analysis approach planned for the final analysis.

Plans to give access to the full protocol, participant level-data and statistical code {31c}

Outside investigators seeking to access the full LET-FEED trial protocol and statistical code can contact the LET-FEED trial Co-PIs (Valentine and Salas) for any additional information not already freely available either in this manuscript or forthcoming publications, including the findings. Additionally, if an investigator has a scientifically sound and reasonable proposal that is IRB approved, that individual can contact the LET-FEED trial Co-PIs with the proposed study design and data analysis plan to request access to the participant-level de-identified data after the trial results have been published.

Protocol and statistical analysis plan {5}

This study protocol will be published for public access along with the statistical analysis herein included. If investigators have additional information related to the protocol or statistical analysis plan, they can contact Dr. Gregory Valentine (gcvalent@uw.edu).

Oversight and monitoring

Composition of the coordinating center and trial steering committee {3d}

The Co-PIs will be responsible for all aspects of conducting the trial, including clinical operations; oversight of all committees and working groups; development of the protocol, consent, manual of operations and all amendments; maintenance of regulatory documents, certification of personnel, recruitment, education, and retention, monitoring of study progress; presentations, and publications.

An **Executive Committee** will meet monthly to assure smooth operation of all aspects of the study. If not all members can attend a meeting, the co-PIs will meet individually with the majority of individuals at least quarterly to update them on the status of the LET-FEED trial.

The **Clinical Steering Committee** will include members of the Executive Committee plus additional representatives, including the grant administrators and site research coordinators working on the LET-FEED trial, and will develop policies and procedures for the implementation of the trial in each clinical stie, collection and verification of data, general issues of patient management and ensuring that all aspects of the proposed study are accomplished and monitor accuracy of data entry. Completed study forms are reviewed prior to entry into the database, clarifying issues as necessary with site coordinators. All study forms will be evaluated for meeting all entry criteria, adherence to protocol, primary and secondary outcomes, and adverse events. Each site's research coordinator in conjunction with the site PIs will monitor the safety of the trial and immediately evaluate any Serious Adverse Events associated with any of the testing procedures involved in this trial, writing summary reports to the site PI, LET-FEED trial co-PIs, the IRB and Gerber Foundation as appropriate.

Clinical Monitoring

Clinical site monitoring will be conducted to ensure that the rights and well-being of trial participants are protected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of the study complies with the currently approved protocol and amendment(s). Monitoring for this study will be

performed by the site PIs, LET-FEED trial Co-PIs, and the LET-FEED trial's statistician. On-site monitoring will occur prior to the start of the study to determine readiness of all participant services and then at 6-month intervals and be conducted by the site PIs. Independent audits may be conducted by each respective site's internal oversight entities (e.g. institutional review board) to ensure monitoring practices are performed consistently.

Composition of the data monitoring committee, its role and reporting structure {28a}

The DSMB will consist of three neonatologists that have expertise in medical directorship, neonatal nutrition, and/or clinical research trials. These individuals will meet at least biannually with the Co-PIs of the LET-FEED trial and additional study team members as needed. Additional meetings will be held as requested by either members of the DSMB or the study team. The DSMB members do not have any direct relationship with any of the LET-FEED trial's co-PIs. The DSMB can request that the LET-FEED trial be halted, paused, or stopped at any point based on concerns raised in their overarching monitoring and evaluation of the safety and efficacy evaluations of the LET-FEED trial.

Frequency and plans for auditing trial conduct {29}

See *data verification, auditing, and adjudication*section above.

Protocol amendments {31}

No changes to the study protocol will be allowed unless they have been discussed in detail and have received the concurrence of the site and Co-PIs. Any amendment/modification to the protocol will be adhered to by the participating center and will apply to all research subjects. Written IRB approval of protocol amendments will be required prior to implementation.

Dissemination policy {8}

Investigators have a responsibility to the public to make study results available as soon as possible. The LET-FEED trial results will be published after all participants have completed the trial, safety data analyzed, and the database has been locked. Additional publications will be submitted as appropriate.

All manuscripts containing the LET-FEED data will be subject to review by the executive committee before they are submitted for publication. All publications will be peer reviewed and published in peer reviewed journals

The LET-FEED trial is registered at ClinicalTrials.gov (NCT06893939), and results information from this trial will be posted to this site.

Discussion

The LET-FEED trial seeks to overcome the critical evidence gap on whether advancing enteral feeds among very preterm newborns after 24 hours or after 72 hours of trophic feed initiation has different

effects on adverse health outcomes. As the previous Cochrane systematic review and meta-analysis on this topic highlighted a paucity of evidence of advancing enteral feeds within the first 72 hours, the authors could only conclude that advancing enteral feeds after four days is associated with an increased risk of invasive infections.[32] The lack of evidence in optimal enteral feeding among very preterm newborns in the first three days after birth has led NICUs across the world to adopt different feeding practices. Thus, the findings of the LET-FEED trial have the potential to lead to immediate changes in feeding practices for very preterm newborns and improve outcomes.

Trial status

This protocol is version 1.0 (March 25, 2025). Recruitment for the LET-FEED trial officially started on July 3, 2025. The approximate date for when recruitment will be completed is December 31, 2028.

Abbreviations

AE: Adverse Event

BCM: Baylor College of Medicine

CI: Confidence Interval

Co-I: Co-investigator

CFR: Code of Federal Regulations

CRF: Case Report Form

CSC: Clinical Steering Committee

DSMB: Data and Safety Monitoring Board

IRB: Institutional Review Board

LET-FEED: Limited versus Extended Trophic Feeding Trial

LOS: Late Onset Sepsis

NIH: National Institutes for Health

PI: Principal Investigator

PMA: Postmenstrual Age

POM: Parent's own milk

SAE: Serious Adverse Event

SPIRIT: Standard Protocol Items: Recommendations for Interventional Trials

UP: Unanticipated Problems

UW: University of Washington

Declarations

Acknowledgements

First and foremost, we would like to extend our acknowledgement and gratitude to the study participants and their families, without which, we would not be able to conduct the LET-FEED trial. We would also like to thank and acknowledge the clinical teams at each of the participating sites: University of Washington (data coordinating center), St. Joseph's Medical Center (Tacoma, WA), Baylor College of Medicine (Houston, TX), University of Oklahoma (Oklahoma City, OK), University of South Florida (Tampa, FL), University of Alabama at Birmingham (Birmingham, AL). We are appreciative to the Gerber Foundation for their support of the LET-FEED trial. We are grateful for the oversight and guidance by our DSMB members including Dr. Kara Calkins (chair), Dr. Jae Kim, and Dr. Jose Perez.

The trial design has been approved by each participating site's institutional review boards. The LET-FEED trial is registered with the Clinical Trial Registry at www.clinicaltrials.gov (NCT06893939).

Authors' contributions {3a}

Dr. Gregory Valentine composed the initial draft of this LET-FEED protocol manuscript. Dr. Stephen Welty, Dr. Robin LaGrandeur, Dr. Katie Strobel, Dr. Yo Nishihara, Ms. Kim Deynaka, Dr. Erynn Bergner, Dr. Steven Ford, Dr. Muralidhar Premkumar, Dr. Amy Hair, and Dr. Ariel Salas provided revisions and updates to the initial draft. All authors agreed to the final version for submission and eventual publication.

Sources of funding and other support {7a}

The LET-FEED trial is funded by the Gerber Foundation.

Availability of data and materials {6}

Outside investigators seeking access to the full LET-FEED trial protocol, statistical code, and de-identified data and materials can contact the LET-FEED trial co-PIs (Valentine and Salas) for any additional information not already freely available either in this manuscript or other publications. Additionally, if an investigator has a scientifically sound and reasonable proposal that is IRB approved, they can contact the LET-FEED trial co-PIs (Valentine and Salas) with the proposed study design and data analysis plan to request access to the participant-level de-identified data after the trial results have been published.

Ethics approval and consent to participate {30}

The LET-FEED trial was conducted after IRB and ethics approvals were obtained by each of the participating sites as there was no single IRB for this trial. All participants must provide consent to participate in the LET-FEED trial.

Consent for publication

This study will be conducted in accordance with the following publication and data sharing policies and regulations. All participants consent to having their data published, and the data will be de-identified.

Competing interests {7b}

The independence of this study from any actual or perceived influence is critical. Therefore, any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the design and conduct of this trial. The study leadership in conjunction with the Gerber Foundation and each participating site have established policies and procedures for all study group members to disclose all conflicts of interest and will establish a mechanism for the management of all reported dualities of interest. Financial conflict of interest attestations will be completed prior to the trial initiation.

Authors' information (optional)

Not applicable

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Figures

	Enrollment	Allocation		Post Allocation									Hospital Discharge
Time Point	-t	t_b	t_0	t_1	t_{14}	t_{21}	t_{28}	t_{35}	t_{42}	t_{49}	t_{32w}	t_{36w}	
Enrollment:													
Eligibility Screen	x	x											
Informed Consent	x		x										
Allocation			x	x									
Interventions:													
Limited Trophic			x										
Extended Trophic			x										
Assessments:													
Baseline		x											
Process Parameters			x				x						
Late Onset Sepsis				x									x
Growth		x											x

Figure 1

Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) diagram for LET-FEED trial

The figure indicates the conduct of the trial as it pertains to screening, interventions and endpoints. Screening for eligibility ranges from prenatal for pregnancies at risk for delivery of a baby or babies between 25 and 0/7 weeks and 31 and 6/7 weeks gestation to newborns delivering without prenatal consent up to 36 hours of life. In research participants meeting inclusion criteria with consent the primary intervention is advancing from trophic feeds at 24-36 hour of life (limited trophic) or at 72-84 hours of life (extended trophic). The primary endpoint is late onset sepsis monitored from 72 hours to discharge. The secondary endpoints are growth parameters, monitored at as indicated. The process measures are attained daily from 24 hours to 28 days and represent the quantification of fluids, enteral and parenteral and the means to deliver parental fluid.

Time points:

-t; is prenatal prior to delivery with a high likelihood of delivering a potential research participant.

t_b; time of birth

t_0; informed consent verified as valid if prenatal consent obtained, or within the first 36 h of life post allocation assignment to limited trophic feed

t_1; post allocation assignment to extended trophic feed between 72 and 84 h of life

t_{14}; 14 days of life

t_{21}; 21 days of life

t_{28}; 28 days of life

t_{35}; 35 days of life

t_{42}; 42 days of life

t_{49}: 49 days of life

If any days of life happen at or after 32 weeks postmenstrual age assessments will be recorded for the post menstrual age (32 and 36 weeks) will be entered subsequent days of life assessments will not be recorded.

t_{32w}; postmenstrual age of 32 and 0/7 weeks

t_{36w}; - postmenstrual age of 36 and 0/7 weeks