WP 1.6a: Minimal enteral colostrum for preterm infants

1. WP (related WPs, MG contact person): Synergies to WPs 1.4a,2.3. MG contacts: Per Sangild, Gorm Greisen

2. Key involved personnel, their institution and mail address (project leader + main study site underlined):
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3. Main aim and sub-aims:
a) To investigate the safety, tolerability and preliminary effects of bovine colostrum, used as the first enteral diet for preterm infants at 1000-1800 g body weight.
b) To assess the feasibility of study procedures, including recruitment rates, parental consent, adherence, sample collection, clinical routines, observing adverse effects.
c) To facilitate the determination of the primary endpoint and sample size for a future larger randomized, controlled trial with bovine colostrum administration.
d) To perform a randomized, controlled trial investigating bovine colostrum versus donor milk or formula.

4. Background and a central hypothesis:
Minimal enteral nutrition (MEN) is a widely used method where small volumes of milk are fed within the first few days after birth with the purpose to promote gut maturation and to supplement nutrients and energy. It is assumed that MEN feedings allow more rapid advancement to full enteral feeding (EN, e.g 120-160 ml/kg/d) and weaning from parenteral nutrition (PN). It remains unclear what milk diet is best when mother's own milk is not available. Mother´s milk is superior to infant formula in promoting feeding tolerance, body growth, intestinal function, and NEC resistance in preterm infants. Feeding with human donor milk is also believed to be beneficial, relative to formula, although this pasteurized milk obtained from mothers later in lactation may less beneficial, relative to the first mothers milk, the so-called 'colostrum'. Maturational and NEC-protective effects of bovine colostrum have been documented in preterm pigs when bovine colostrum is used as the first diet after birth.
We hypothesize that bovine colostrum, used as MEN for preterm infants, is safe and helps to provide nutrients and improve gut maturation in preterm infants, when enough mother's milk is not available.

5. Key analyses and methods
Phase a: Pilot study to test safety/tolerability of bovine colostrum, and feasibility of study procedures (n=20).
Phase b: Randomized controlled study, fully powered to detect differences in primary endpoints (n≈150)
Phase a will be run both at Copenhagen University Hospital and at Foshan Woman's and Children's Hospital.
Study site(s) for phase b is to be decided. Diet interventions for a maximum of 10 days. Control group: Donor milk or infant formula supplemented to mother´s own milk. Intervention group: Bovine colostrum supplemented to mother´s milk or infant formula as needed.

6. Expected results
We expect to document whether bovine colostrum can be used as a beneficial first enteral diet for preterm infants that have limited or no access to mother´s own milk. Results include clinical neonatal outcomes (time to

full feeding, feeding intolerance, combined incidence of serious infections/NEC, days of hospitalization, anthropometry data, days to regain birth weight, days on PN, and stool characteristics) as well as paraclinical outcomes (plasma citrulline, lactase activity, intestinal permeability, fecal microbiota composition, and fecal bacterial fermentation.

7. Estimated time frame

Task	2013				2014				2015				2016				2017			
Planning of exp., ethical protocols	x	x	x																	
Phase a) execution, Copenhagen				x	x	x														
Phase a) execution, Foshan				x	x	x														
Laboratory analyses, Copenhagen							x	x	x											
Laboratory analyses, Foshan							x	x	x											
Publication from both phase a)										x	x									
Planning of experiments, phase b)						x	x													
Phase b) execution, Copenhagen (?)									x	x	x	x								
Phase b) execution, Foshan (?)									x	x	x	x								
Laboratory analyses, Copenhagen (?)												x	x	x						
Laboratory analyses, Foshan (?)												x	x	x						
Publication from phase b) (?)															x	x				

8. Estimated budget from NEOMUNE:

Phase a) 0.6 mio DKK (PhD, post doc salaries, Yanqi Li, Sandra Meinich)
Phase b) 1.0 mio DKK (PhD, post doc salaries, Yanqi Li, Sandra Meinich)

9. Estimated budget from elsewhere:

Phase a) 0.6 mio DKK (Zhu Yanna, Yanqi Li, PhD students, MSc students, hospital personnel)
Phase b) 1.0 mio DKK (Zhu Yanna, Yanqi Li, PhD students, MSc students, hospital personnel)

10. Additional comments:

- Ethical approval of the studies will be applied for in both Denmark and China. The exact planning of Phase b will depend on the results from Phase a. If results from Phase a are not promising, then Phase b will be cancelled.
- A large effort is involved in securing that the product, powered bovine colostrum, can meet current legislation for use as infant formula. The investigators work with Biofiber to secure this.
- Very close collaboration between Copenhagen University Hospital and Foshan Women's and Children's Hospital, China.

WP 1.6b: Probiotics for preterm infants

1. Related WPs, MG contact person: Synergy with WP 1.3a, 1.4b, 1.5, 2.2, 3.2. MG contact: Gorm Greisen

2. Key involved personnel, their institution, mail address (project leader + main study site underlined):
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3. Main aim and sub-aims:
To plan and possibly initiate a large-scale randomized clinical trial in newborn infants with probiotics in China.
a) To explore the ability of Chinese hospitals and their interest
b) To develop a protocol
c) To organize the trial

4. Background and a central hypothesis:
Probiotics have been shown to reduce the incidence of necrotizing enterocolitis (NEC) and overall mortality in infants with birth <1500 g and/or gestational age <32 weeks. This has been demonstrated in systematic meta-analysis covering more than 15 randomized trials and more than 2,500 infants. Several large trials are at present awaiting conclusions. While probiotic use in clinical routine is far from universal, its use is increasing, and a standard large-scale placebo-controlled trial runs the risk of becoming ethically difficult within such a trial's lifetime.
However, there are several problems and questions that remain unanswered before use of probiotics can be recommended as part of standard clinical care for preterm infants. In previous studies, many different probiotic strains, or combinations of strains, have been used in many different concentrations. In most trials, probiotics have not been given during the first days of life and there may or may not be an advantage in allowing spontaneous bacterial colonization take place prior to introduction of probiotics. Finally, the interactions between probiotic effects and the timing, dose and type of antibiotics given to preterm infants have not been investigated. Clearly, this WP cannot answer all these important questions. The choice of intervention in this WP shall, after careful evaluation of a) already ongoing international trials, b) current practice at NEOMUNE hospitals (WP 4.1b), and c) supporting evidence from NEOMUNE animal model studies (WP 2.2 and WP 3.2), be built on the following hypotheses:
2. The clinical effects of 10^9 and 10^{10} CFU per day do not differ.
3. Initiation of probiotics administration on day one, prior to the spontaneous colonization of the gut improves the clinical outcome.
4. The clinical effects of probiotics are more pronounced following use of antibiotics for preterm infants.

5. Key analyses and methods:
Primary outcome is survival of preterm infants without NEC at 37 weeks of gestational age. Blinded allocation. 2 x 2 factorial design (dose and time). Large scale, pragmatic design. External monitoring.

6. Expected results:
- A trial with probiotics in preterm infants that is ready to start. The results from such a study may be generalizable to other types of probiotics and other types of patients.
- Results concerning the time, dose and interaction with antibiotics are relevant for assessing the risk of side effects (probiotic overgrowth or inappropriate immune stimulation) in particularly vulnerable infants. For such results, this WP may have to rely partly on the NEOMUNE animal studies for translational conclusions,

considering the size of this task as clinical infant studies.

7. Estimated time frame

Task	2013				2014				2015				2016				2017			
1. Exploring data base information				x	x	x	x	x												
2. Evaluation of pig model studies						x	x	x												
3. Evaluation of mouse model study						x	x	x												
4. Protocol formation								x	x	x										
5. Organization of clinical study											x	x								
(6. Clinical trial in preterm infants)												x	x	x	x	-	>			
(7. Analyses of results)																	x	x	x	x
8. Publication											x	x							x	x

8. Estimated budget from NEOMUNE:

3.0 mio DKK (financed by funding from Chr. Hansen A/S)

9. Estimated budget from elsewhere:

3.0 mio DKK (university and hospital partners, internal staff and equipment). Co-funding from other NEOMUNE projects (e.g. animal studies)

10. Additional comments:

- This WP is closely linked with other WPs (mostly WP 4.1b, 2.2, 3.2) that are initiated at the start of the NEOMUNE period to maximize knowledge transfer to this WP before the project plans are fixed. Considering the extensive NEOMUNE network in this area, there will be ideal conditions for formulating a clinical trial that answers the most important question at the time of initiation.
- The total scale and nature of proposed clinical trial is heavily dependent on sponsoring from relevant industry partner(s) and the amount of funds spent during the planning phase for the trial.
- While the costs of the planning phase for this project (see above Gantt diagram) are mainly covered by funds from elsewhere in NEOMUNE, some funds may be allocated also from this project.
- The main outcome of this WP is to make the protocol. The actual clinical trial is not budgeted.
- Differs from original plan with Hans, which included a control group without probiotics.