WP 2.3: Enteral and parenteral feeding for preterm pigs

1. Related WPs, MG contact person: Synergies with WPs 1.4,2.1-4,3.1. MG contact: Thomas Thymann

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:

The aim is to identify optimal feeding regimens (enteral and/or parenteral) within the first weeks after birth using the preterm pig model, i.e., when to feed, how much to feed, and which diet? Focus is on the transition to natural, unmodified enteral milk diets, such as bovine colostrum, human donor milk and porcine amniotic fluid. To identify the optimal timing, nature and amount of parenteral nutrition is an associated goal.

4. Background and a central hypothesis:

The first milk after birth promotes intestinal growth, bacterial colonization and immune maturation in weak newborns. Enteral feeding may however predispose to harmful inflammatory lesions, especially using large feeding volumes and suboptimal diets. Thus parenteral nutrition is crucial to promote adequate growth and development before full transition to enteral nutrition can be implemented. For both enteral and parenteral nutrition, there is much debate about the optimal diet composition, time and amount of feeding for preterm infants. In this project we investigate some *intact natural perinatal diets* and how these should be fed in association with parenteral nutrition to promote optimal immunity, gut and brain maturation. Newborn, caesarean-delivered preterm pigs on parenteral nutrition are fed graded doses (minimal enteral nutrition, MEN) during the first one or two weeks after birth. Each of the experimental diets will be compared with a corresponding diet of preterm infant formula, or no feeding at all (total parenteral nutrition, TPN). Separate studies on the nature of parenteral nutrition may be added (particularly related to lipid fractions).

We hypothesize that feeding small amounts of colostrum, human milk or amniotic fluid, as enteral adjuncts to parenteral nutrition within the first week, improve immunity, gut and brain functions in preterm neonates.

5. Key analyses and methods:

Study 1: Preterm pigs are fed total parenteral nutrition or bovine colostrum as minimal enteral nutrition for 5 days and then transitioned to a bovine milk formula for up to 26 days
Study 2: Preterm pigs fed 10-60 mL/kg/d bovine colostrum for 5 days are compared with pigs fed formula.
Study 3: Preterm pigs fed porcine amniotic fluid are compared with pigs not fed any enteral diet for 5 days.

Study 4: Preterm pigs fed total parenteral nutrition with improved fatty acid composition.
Human milk is obtained from a local donor bank, amniotic fluid from caesarean section on sows, bovine colostrum from the company Biofiber Damino, also delivering product for corresponding infant studies.
Manipulations of the composition of the parenteral nutrition will be done in collaboration with Fresenius Kabi.
Immunity endpoints: Immune genes, immune proteins (bovine IgG in plasma/feces), inflammation, NEC.
Gut: Histology, digestive enzymes, tight junction proteins, permeability, FISH microbial analyses of tissue and high throughput sequencing fecal analysis, food passage.
Brain: Wet weight, BdNF, , total activity levels, , open field test, activity test, EEG.

6. Expected results:

The results will document whether bovine colostrum is better that preterm infant formula in improving growth and development of preterm pigs as models for infants. It will also provide information about apparent safe and efficacious volume of feeding. Finally, the results will help to improve the regimen for parenteral nutrition as a life-saving therapy for preterm infants necessary during the gradual transition from parenteral to enteral nutrition after birth. This information will be important for interpretation of the corresponding infant studies (WP 1.4a, 1.6a). The results will also show preliminary evidence for possible use of amniotic fluid in preterm infants. If beneficial for growth and development, this may lead to pilot studies on feeding human amniotic fluid to preterm infants as part of NEOMUNE.

Predicted publications:

1) Minimal enteral nutrition with human donor milk or bovine colostrum in preterm pigs
2) Minimal enteral nutrition with amniotic fluid improves growth and development in preterm pigs
3) Parenteral fatty acids influences immunity and brain outcomes in preterm pigs

7. Estimated time frame

Task	2013				2014				2015				2016				2017			
Planning of experiments, protocols	x																			
Study 1 execution	x	x	x	x		X														
Study 2 execution	x	x	x																	
Study 3 + 4 execution	x	x	x						x	x										
Study 1 immunity analyses				x	x	X	X													
Study 1 gut analyses				x	x	x	X													
Study 1 brain analyses			x	x	x	x	X													
Study 2+3 immunity analyses				x	x															
Study 2+3 gut analyses			x	x																
Study 2+3 brain analyses			x	x	x	x														
Study 4 analyses											x	x	x							
Publication				x	x	x							x	x	x					

8. Estimated budget from NEOMUNE:

2.0 mio DKK (costs for pig experiments, analyses, immunity, allocated from Thomas Thymann funds)

9. Estimated budget from elsewhere:

1.8 mio DKK. PhD stipends for Rene Shen and Stine Petersen, post doc salary for Mette Ø
2.0 mio DKK predicted from industry partners (products, direct funds, BioFiber, Fresenius Kabi).

10. Additional comments:

- The results serve as proof-of-concept for pilot infant studies with bovine colostrum, and to support possible pilot intervention studies with human amniotic fluid for human infants (WP 1.6a). The experiments will also support scientific evidence for use of human donor milk (WP 1.4a and WP1.4b).
- The work related to the composition of parenteral nutrition will be run with the industrial partner Fresenius Kabi and may run partly in collaboration with Children's Nutrition research Center in Houston (Prof. Burrin), Aarhus University (Ass. Prof. Charlotte Lauridsen), Univ. Hong Kong (Ass. Prof. Jetty Lee) which all have activities on the dietary fatty acid composition on immunity, gut and brain endpoints.