Abstract Submission

Nutrition

Nutrition and Metabolism, Mechanisms

ESPGHAN2014-1859

MINIMAL ENTERAL FEEDING WITH AMNIOTIC FLUID IN PRETERM PIGS

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Presentation Preference: No Preference

Objectives & Study: The optimal diet and feeding strategy after preterm birth is unknown, especially if breast-feeding is scarce or absent. Amniotic fluid (AF), colostrum, and milk exert a continuum of beneficial effects on the developing intestine in the perinatal period via growth promoting and immunomodulatory factors. We hypothesized that small enteral feeds with AF, as an adjunct to parenteral nutrition during the first days after preterm birth, improve gut functions.

Methods: Preterm pigs (105-106 d of gestation, term 115-117 d) were delivered by cesarean section and fed parenteral nutrition with *nil per os* (NPO, n = 14) or AF (AF, n = 13) given as small enteral feeds advancing from 24 to 72 ml/kg/d over the first five days after delivery. An enteral galactose challenge test was performed on d4. After an enteral challenge with infant formula and a lactulose-mannitol bolus on d5, blood samples were collected to evaluate gut hormones and citrulline in plasma, and intestinal permeability, respectively. The pigs were then euthanized for tissue collection.

Results: The incidence of NEC (AF 8%, NPO 14%) and diarrhea (AF 23%, NPO 7%) were low and did not differ between groups. Body weight gain was higher in AF vs. NPO pigs (64 ± 3 vs. 23 ± 3 g, $P < 0.001$). The relative weights of the stomach (mean across groups 6.8±0.2 g/kg), small intestine (24.2±0.8 g/kg), and colon (9.4±0.6 g/kg) did not differ between groups, and there was no effect of AF on intestinal mucosa percentage, villus height, and crypt depth. Likewise, galactose uptake capacity (670±79 µM), plasma cortisol (55.0±7.6 ng/ml), citrulline (64.5±4.5 µM), and glucagon-like peptide 2 levels (55.0±3.7 pM) did not differ between groups, whereas gastric inhibitory polypeptide was higher in NPO vs. AF pigs (112 ± 20 vs. 58 ± 11 pM, $P < 0.05$).

Conclusion: Small enteral feeds with AF during five days of parenteral nutrition were well tolerated in preterm pigs. AF stimulated body weight gain, but had limited effects on gastrointestinal growth, structure, and function. Further studies are required to evaluate how minimal enteral AF feeding improves weight gain and any effects on later intestinal adaptation following transition to full enteral milk feeding.

Disclosure of Interest: None Declared

Abstract Submission

Nutrition

Nutrition and Metabolism, Mechanisms

ESPGHAN2014-1649

PREMATURE DELIVERY INDUCES MARKED GROWTH RESTRICTION IN THE POSTNATAL PERIOD OF PIGS

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Presentation Preference: No Preference

Objectives & Study: Optimal nutrition and care is crucial to stimulate growth and development of preterm infants. Relative to term neonates, growth deficits may be caused by low nutrient intake in combination with possible organ dysfunctions, infections and metabolic disturbances. We hypothesized that preterm neonates show diminished growth and organ development during the first 4 weeks after birth, despite being delivered the same way and provided identical diet and rearing conditions as piglets born at full term.

Methods: Pigs were delivered by caesarean section at preterm (106 d, n=12) or term gestation (118 d, n=22). For the first 4 postnatal days pigs from both groups were reared in incubators and fed isoenergetic diets (74-110 kcal/kg/d) of parenteral nutrition (PN), with or without supplemental minimal enteral nutrition (MEN, 0-64 ml/kg/d bovine colostrum). This was followed by individual rearing in open cages and feeding with bovine milk-based formula until day 26 (64-200 ml/kg/d). In-feed antibiotics were provided temporarily to all these immune-compromised pigs to prevent sepsis and infections. Growth was recorded daily and organ weights were recorded on day 26.

Results: Preterm pigs showed severe signs of prematurity, including hypothermia, respiratory distress and delayed motor skill development for the first 5 days of life. The time to first eye lid opening and first stand and walk was markedly increased in preterm vs. term pigs (all $P<0.001$). Likewise, the average number of days with diarrhea was higher in preterm vs. term pigs (+100%, $P<0.001$) indicating a dysfunctional digestive system. Preterm pigs had a slower growth rate (2 and 6 g/kg/d) on d 0-5, compared to that of the term pigs that grew 32 and 19 g/kg/d in the PN and MEN groups, respectively ($P <0.05$).Growth deficits in the preterms remained until d 26 (20-22 vs. 31-34 g/kg/d). Relative to their body weight, preterm pigs had lower weight of the colon, liver and spleen on d 26, compared with term pigs (-32, -10 and -74% respectively, all $P<0.05$), while lungs, kidneys and brain were increased by 11, 14 and 23%, respectively (all $P<0.001$).

Conclusion: Preterm neonatal piglets are severely growth-restricted in the postnatal period, even when provided the same diet and rearing environment as term neonates. Specific reductions in internal organ growth and function in preterm neonates may inhibit digestive and metabolic capacity in the postnatal period. Minimal enteral nutrition relative to total parenteral nutrition during the first days after birth does not affect later growth rates. The immature, immune-compromised preterm pig may be a highly sensitive model to test effects of diet and rearing conditions on organ development in weak newborn infants.

Disclosure of Interest: None Declared

Abstract Submission

Nutrition

Nutrition and Metabolism, Mechanisms

ESPGHAN2014-1644

FORMULA WITH HIGH LEVEL OF BIOACTIVE PROTEINS AND LACTOSE STIMULATES GUT FUNCTION IN PRETERM PIGLETS

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Presentation Preference: No Preference

Objectives & Study: Relative to intact milk diets such as colostrum and milk, formula feeding is associated with compromised intestinal maturation and a higher incidence of necrotizing enterocolitis (NEC) in preterm neonates. This may be explained by the use of whey protein concentrates (WPC) with reduced levels of bioactive proteins or use of maltodextrin rather than lactose as the carbohydrates source. We investigated the effects of two WPCs with high (WPC H) or low level (WPC L) of bioactive proteins on intestinal health in preterm pigs. WPC H had higher levels of native lactoferrin (5 times), IgG (3 times) and IGF-I (3 times) than values in WPC L. Formula predominantly based on lactose (relative to maltodextrin) protects against NEC in preterm pigs. Therefore, each WPC was included into formulas based on either lactose or maltodextrin to test the interaction between the nature of the protein and carbohydrate fractions.

Methods: Sixty-one caesarean-delivered preterm pigs were distributed into four groups and given parenteral nutrition and one of four formulas: lactose-dominant formulas (75% lactose+25% maltodextrin) containing WPC H (L/H, n=15) or WPC L (L/L, n=15), or maltodextrin-dominant formulas (75% maltodextrin+25% lactose) containing WPC H (M/H, n=15) or WPC L (M/L, n=16). The four formulas were isoenergetic and had similar macronutrient concentrations. A series of gut structural and functional indices, including mucosal lesions, were evaluated on day 5.

Results: Birth weight, daily weight gain, the weights of lung, heart, liver, kidney, spleen, stomach, colon and intestine, relative to body weight, and NEC incidence (51% NEC across the groups) were all similar among the four groups. Pigs fed WPC H containing formulas (L/H and M/H) had an increased proportion of mucosa ($P < 0.05$) and increased villus height ($P = 0.09$) in the proximal intestine, relative to the ones fed formula containing WPC L (L/L and M/L), respectively. Only in the lactose-dominant formula, did WPC H (L/H) stimulate galactose and lactose absorptive capacity and lactase activity, relative to WPC L (L/L, $P < 0.05$).

Conclusion: WPCs with different levels of bioactive proteins differ in their effects on gut structure and function in formula-fed preterm pigs. The effects interact with the composition of the carbohydrate fraction, with the beneficial effect of WPC H formula disappearing when the main carbohydrate source in formula is maltodextrin and not lactose. While formulas based almost 100% on lactose are known to be protective against NEC in preterm pigs, we now show that 25 and 75% lactose formulas fail to protect against NEC. Optimization of the levels of bioactive proteins in WPC, together with the composition of the carbohydrate source in formulas, is important to enhance both the nutritional contents as well as the gut protective effects in highly sensitive preterm neonates.

Disclosure of Interest: Y. Li Conflict with: This author was employed at Arla Foods Ingredients Group P/S as an industrial PhD when this study was performed.T. Thymann: None DeclaredD. Chatterton: None DeclaredA. Kvistgaard Employee of: Arla Foods Ingredients Group P/SP. Sangild: None Declared

Abstract Submission

Nutrition

Nutrition and Metabolism, Mechanisms

ESPGHAN2014-1563

HUMAN DONOR MILK AND BOVINE COLOSTRUM IMPROVES BODY GROWTH AND GUT HEALTH RELATIVE TO INFANT FORMULA IN PRETERM PIGS

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Presentation Preference: No Preference

Objectives & Study: Mother's milk is the optimal diet for preterm infants but supply is often limited following preterm delivery. Infant formula (IF) and human donor milk (DM) are therefore used as alternatives, although both diets are not efficient in preventing gut dysfunction and necrotizing enterocolitis (NEC), relative to mother´s own milk. Both DM and bovine colostrum (BC) exert NEC-preventive effects within the first week after birth in preterm pigs (Jensen *et al*. 2013). In this study, we hypothesized that DM and BC would also improve growth and NEC resistance beyond the immediate postnatal period.

Methods: For ten days after birth, caesarean-delivered preterm pigs were fed slowly increasing doses (3-15 mL/kg/3 h) of three iso-energetic diets; preterm IF (n = 14), BC (n = 18), or DM (n = 15). Clinical condition, NEC lesions, mucosa proportion, and body and organ weights were recorded on day 10.

Results: NEC incidence was lowest in the DM pigs (40%), relative to BC (67%) and IF pigs (79%, $p < 0.05$ compared with DM). Mucosal weight in the proximal and middle intestine was elevated in BC pigs, relative to DM and IF pigs (both $p < 0.05$). Throughout the study, diarrhoea severity score was lower in BC pigs, relative to IF pigs ($p < 0.01$), and after day 6, BC diarrhoea score was also lower than in DM pigs ($p < 0.001$). Correspondingly, weight gain was higher in BC pigs (30 g/day), relative to both DM and IF pigs (-3 g/day, $p < 0.01$) and IF pigs (-12 g/day, $p < 0.01$), and food passage time on day 8 was longer (19 h vs. 7 and 3 h, respectively, both $p < 0.05$).

Conclusion: Slowly advancing volumes of DM and BC prevented NEC, diarrhoea, and poor clinical condition within the first week after birth. During the second week after birth, only BC had beneficial effects on weight gain, diarrhoea outcome, and mucosal weight, relative to IF. All three diets were associated with significant NEC lesions on day 10. Both BC and DM may be alternatives to IF during the critical neonatal period of preterm infants, but piglet model studies suggest that the length and volume of feedings must be carefully optimized to avoid negative effects.

References: Jensen ML, Sangild PT, Lykke M, Schmidt M, Boye M, Jensen BB, Thymann T (2013). Am J Physiol Regul Integr Comp Physiol. 305(1):R4-R12.

Disclosure of Interest: None Declared

Abstract Submission

Nutrition

Nutrition and Metabolism, Mechanisms

ESPGHAN2014-1656

MINIMAL ENTERAL NUTRITION INDUCES DIET-DEPENDENT EFFECTS ON GUT MATURATION AND NEC SENSITIVITY IN PRETERM PIGS

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Presentation Preference: No Preference

Objectives & Study: The optimal way to feed newborn preterm infants, when breast-feeding is impossible, is not clear. We hypothesized that feeding small volumes of enteral nutrition just after preterm birth stimulates gut maturation and that the response to infant formula differs from that of a highly bioactive milk diet, such as bovine colostrum.

Methods: Pigs fed total parenteral nutrition (TPN, n=14) for five days after preterm birth were compared with pigs fed slowly advancing volumes (16-64 ml/kg/d) of preterm formula (IF, n=15) or bovine colostrum (BC, n=13), as supplement to PN. The three groups received the same amount of calories (74-110 kcal/kg/d) and fluids (96-144 mL/kg/d). After a meal test on day 5, blood samples were collected to measure gut hormones, urine was sampled to measure intestinal permeability (lactulose-mannitol test), and internal organs were collected and weighed.

Results: On day 5, diarrhea and NEC incidences were markedly increased in the IF group (93 and 60%, respectively), compared with the BC (15 and 0%) and TPN (7 and 15%) groups. At necropsy, the IF pigs also had higher gastric residual volume than the other two groups (26±3 vs. 17±1 ml/kg, pooled values for BC and TPN, p<0.05). The TPN group showed higher weight gain (31±6 g/kg/d) than the other groups (14±2, p<0.05). Intestinal permeability was markedly reduced in the BC group (0.02±0.00) relative to the other two groups (0.11±0.03, p<0.05). Intestinal mass was reduced in the TPN group, compared with the other groups (22±1 vs. 27±1 g/kg, p<0.05). Plasma citrulline (a marker of functional enterocyte mass) was reduced in the IF group (38±3 μM) relative to the other groups (65±5 μM). Plasma levels of glucagon-like peptide 2 (GLP-2) following the meal test, were reduced in the TPN pigs (60±6 vs. 101±7 pmol/L, p<0.05) while levels of gastric inhibitory polypeptide (GIP) were highest in the BC group (139±13 vs. 87±8 pmol/L in the other groups, p<0.05).

Conclusion: The effect of minimal enteral nutrition on the immature gut is highly diet-dependent. Minimal enteral feeding with infant formula increases the risk of NEC compared with TPN, bovine colostrum more effectively promotes gut growth and function and protects against NEC.

Disclosure of Interest: None Declared