
6 Summary

The objective of our investigation was the evaluation of ileal digesta - qualitatively and quantitatively - in piglets before and after weaning, with special reference to protein and amino acid digestion. To implement this aim we conducted two main experimental complexes: a long-term slaughter trial for the qualitative aspects and two balance trials for the quantitative part. During the slaughter trial animals were sacrificed at different stages before and after weaning: -6, -4, -2 days preweaning, weaning day (28 d of age), +1, +2, +5, +8 and +15 days postweaning. Dietary regimes comprised sow milk as sole feed (no creep feed) before and after weaning four different starters, two reference diets based on soybean meal and whey and two home-produced diets based on cereals and legume seeds. Reference diets differed in supplementation of in-feed antibiotic avilamycin (+AB, -AB) and home-produced diets in crude fibre content, being either 3 or 8% (LF, HF).

In the slaughter trial we applied a wide range of analysis, determining pH, DM, LA, VFA, NH₃, biogenic amines and D-alanine and investigating the luminal microflora with classical and molecular techniques at ileal level. Although we observed changes in some parameters during weaning transition – decrease in ammonia and VFA, increase in LA – these scarcely proofed to be statistical significant. Also classical and molecular microbiology revealed a remarkable stability in the universal ileal microflora of piglets receiving sow milk or starter diets. Observed changes were more age-related than influenced by dietary treatment. Only the *Lactobacillus* spp. group displayed a differential response: here we could establish a diet-dependent change in the *Lactobacillus* spp. population. The home-produced diets induced a more diverse population than the reference diets, being more comparable to the *Lactobacillus* spp. population of suckling piglets. Lactic acid producing bacteria are regarded as health beneficial, especially in the GIT of young animals such as weaning piglets. Therefore the promotion of this group by the home-produced diets can be considered as a major benefit for intestinal health. However, more studies are required to receive statistical relevant evidence. Furthermore we detected a negative correlation between the *Lactobacillus* spp. group and yeast (Pearson Correlation Coefficient $r = -0.24$, $p = 0.02$) and a highly positive correlation to amines (putrescine = 0.41, $p < 0.0001$; histamine $r = 0.36$, $p = 0.0005$; cadaverine $r = 0.35$, $r = 0.0006$; spermidine $r = 0.23$, $p = 0.03$; spermine $r = 0.28$, $p = 0.007$). The inverse relationship with yeast was reported from Maribo et al., 2000, but evidence for biogenic amines in the porcine gut is rare in literature. There are some *in vitro* studies reporting production of biogenic amines in this bacterial species, but the main contribution to this field comes from food technology rather than from animal physiology research. These results suggest a crucial role of *Lactobacillus* spp. in the terminal ileum of weaning piglets.

Further investigation is required to elucidate those interesting interrelationships in the GIT of farm animals, especially in swine.

Balance trials were conducted with artificially reared piglets (milk replacer) to assess the preweaning quantitative aspects and postweaning with weaners fed the four starters already applied in the slaughter trial. The ^{15}N tracer technique was used to estimate the endogenous nitrogen loss and the D-alanine method to assess the bacterial nitrogen contribution.

The both balance trials aimed to investigate the nitrogen digestibility, endogenous and bacterial nitrogen at ileal in unweaned and weaned piglets fed different diets. The ^{15}N tracer technique was applied to estimate endogenous nitrogen losses and calculate real ileal digestibility of nitrogen. Bacterial nitrogen was assessed by means of D-alanine as bacterial marker. Our results demonstrate that in unweaned piglets nitrogen digestibility was highest and endogenous and bacterial N flow (g/100g DMI resp. CPI) significantly lower compared to weaned pigs. Looking at total ileal N endogenous nitrogen contributed to the greatest and exogenous (dietary) nitrogen to the least part. This underlines the high nitrogen utilisation of milk replacer at the terminal ileum. Similar results were established for the low-fibre starter LF: real nitrogen digestibility was equal to that of milk replacer ($98.5 \pm 1.3\%$ vs. $98.2 \pm 0.4\%$) and endogenous nitrogen was the main contributor to total ileal N. The remaining starters +AB, -AB and HF showed markedly lower RID_N (90.2 ± 1.3 , 88.3 ± 1.4 , $94.0 \pm 1.4\%$). In the reference diets endogenous N was lower and exogenous N contribution greater than in home-produced starters and milk replacer. Bacterial nitrogen proved to be less affected by weaning itself but influenced by dietary regime. Unweaned animals and those fed +AB and -AB exhibited equal bacterial nitrogen contribution (11.6 ± 1.5 , 11.4 ± 0.7 , $11.8 \pm 0.9\%$ of total N), whereas in piglets receiving LF and HF bacterial contribution was markedly lower (9.1 ± 0.8 , $7.9 \pm 0.8\%$ of total N). This might be attributed to differential microbial population, though these differences were not present in the results of the classical plate counting. One reason could be that the major part of the gastrointestinal microflora belongs to yet uncultured microorganisms, thus not to be proofed by classic microbiological techniques. However, D-alanine as bacterial marker can be assumed to have well covered the yet uncultured bacteria. D-alanine was used as a novel approach in swine. At present it is used successfully in ruminants, but not in monogastric animals. However, research of Schönhusen and Kwella (unpublished data) indicate its suitability as a bacterial marker in monogastric animals as well. Our results support this notion. Results obtained from both the balance trials imply that the magnitude of the main components of total ileal nitrogen – endogenous and bacterial N – are more dependent on the applied dietary regime, whether being milk replacer or starter diets, than on the actual weaning process.

Comprising the experiments of this study – slaughter and balance trials - we could demonstrate that the small intestinal environment is subjected to a short-termed adaptation period during the actual weaning process. During this period the ileum adjusts to a physiological equilibrium, which goes along with minor changes in various parameters such as ammonia, VFA and LA. Postweaning our results showed no advantage of dietary supplementation with the in-feed antibiotic avilamycin compared with the other three starters. In fact we could demonstrate considerable beneficial characteristics of the home-produced low-fibre diet LF: higher nitrogen digestibility (apparent and real ileal N digestibility) as evidenced in the balance trial and a promotion of the *Lactobacillus spp.* population in the terminal ileum as showed in the slaughter trial. Both features might proof advantageous in the practical application of this diet: higher digestibility has the potential to improve ADG in long-term administration (exceeding 15 days postweaning), which is of great importance for commercial swine production in a financial aspect. Furthermore, enhancing the diversity of *Lactobacillus spp.* in the terminal ileum indicates that this cereal-based diet has probably prebiotic properties and therefore could be of good use in commercial swine production, particularly with regards to the EU-wide ban of antimicrobial feed additives in weaning piglets.

One crucial aspect of research, and especially in animal nutritional research, is to gain a wholesome picture of the physiological processes going on in the animal while it is subjected to such disturbing events like the weaning transition. In order to achieve such objective, we collaborated with various institutional partners in the frame-work of the EU-project "Healthypigut". Besides the analysis and results we just demonstrated in this thesis we provided samples from these very piglets used in the outlined experiments to be investigated in different respects. Tissue sections from the small and large intestine of piglets from the slaughter trial were subjected to immunological and immuno-histological analysis (Medical School Hannover / Germany, group Hermann-Josef Rothkoetter). Digesta samples collected during the postweaning balance trial were provided for the analysis of mucine content (INRA Rennes / France, group Jean-Paul Lalles). Results of these investigations will be compiled and statistically analysed, thus providing the chance to integrate information about the same animal from different fields. Such approach is useful to obtain a wholesome image of the physiological state and its alterations during weaning and the linkage of digestive physiology, microbiology and immunology.

Nevertheless more research is required to elucidate underlying mechanisms and develop suitable strategies for a gentle weaning process.