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## Variability of feed efficiency in growing pigs: towards predictive biomarkers

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#### 4.4. WP2: The effect of genetic type and feed restriction on the urine metabolome of growing rabbits

Mette Skou Hedemann (AU)

It is well known that feed restriction has good effects on the health and productive performance of growing rabbits: It reduces the risk of digestive diseases and improves feed efficiency. The purpose of the present experiment was to study the biological basis of feed efficiency on ad libitum or restricted feeding in two rabbit lines selected for different purpose: the Prat Line that is selected for litter size at weaning and the Caldes Line that is selected for post-weaning growth rate. The metabolome is influenced by genetic as well as environmental factors (e.g. diet, physical activity and medication) and consequently metabolomics is a valuable tool to study genetics and dietary interventions and the interaction between them. A total of 28 rabbits (14 Prat and 14 Caldes) were included in the experiment at weaning (32 d). The animals were fed a standard diet (16.4% crude protein, 15.2% fibre, 4% fat) and water was always available. When rabbits were 53 d old, they were moved to metabolic cages and fed ad libitum until they were 63 d old. Then, they were subjected to a 3-days period of feed restriction at 90-80 and 80-70 % of maintenance requirements in Caldes and Prat lines, respectively. This period was followed by a 4-days recovery period with ad libitum feeding and then, a new 3-days period of feed restriction at 60-50 and 50-40 % of maintenance requirements in Caldes and Prat lines, respectively, until they reached 73 days of age. Urine samples were collected on d 63 and 73 of life. The samples were prepared for non-targeted LC-MS metabolomics and data were acquired on a UHPLC (Dionex, Sunnyvale, CA, USA) coupled to an Impact HD mass spectrometer (Bruker Daltonics, Bremen, Germany). The data were preprocessed using the XCMS R package and Principal Components Analysis (PCA) was performed using LatentX 2.12. The PCA scores plots of the data in both positive and negative mode showed a clear separation between initial samples, with rabbits fed ad libitum, and samples taken at the end of the trial, just after the animals had been subjected to a strong feed restriction (40-50% of maintenance requirements) whereas there was no separation between the breeding lines. Several of the metabolites responsible for the separation between ad libitum and restricted feeding are a result of the microflora metabolism (e.g. p-cresol sulfate, hippuric acid, kynurenic acid, and xanthurenic acid) furthermore the excretion of vitamins, betaines, and feed associated metabolites were changed as a result of the feeding regimen. In conclusion, there was an effect of feed restriction on the urine metabolome of growing rabbits whereas no effect of genetic type was observed.

#### 4.5. WP2: Variability of feed efficiency in growing pigs: towards predictive biomarkers

Florence Gondret (INRA)

Improving food efficiency (FE) is a major challenge for the pork industry, for both economic and environmental reasons. It is therefore essential to identify, on the one hand, the biological mechanisms involved in the inter-individual variability of FE, and on the other hand, to propose biomarkers of this trait. This study aims to identify genes whose expression levels in different tissues contributed significantly to differences in FE. Different transcriptomic datasets in loin muscle or in the blood were obtained in pigs from a divergent selection on residual feed intake. The pigs were from the 6th to 8th generation of selection, weighed between 15 and 115 kg, and were either feed-restricted or fed ad libitum with standard or fiber-rich diets (Vincent et al., 2015 Merlot et al., 2016; Gondret et al., 2017; Reis Furtado Campos et al, 2014). For each animal, the gain to feed (G:F) and gain to feed energy (G:Fe) were calculated, and their genetic values for residual feed intake (RFI) were obtained. In muscle, the expression of 22 400 annotated molecular probes was measured on 71 pigs. In the blood, 6 000 annotated molecular probes were commonly found across the different datasets (n = 84 pigs). To identify the most important predictors of FE,



Random Forests (Breiman, 2001) were implemented with the Salford Predictive Modeler (V8.0) software. In addition, sub-networks of co-expression of genes were elicited using WGCNA package (Langfelder and Horvath, 2007) implemented under R (V3.4.2) software, and related to G:F, G:Fe or RFI. In the muscle, a subset of about fifty genes whose levels of expression explained respectively, 67% of the variability in F:G and 65% of the variability in RFI, was proposed. These genes participated in various biological processes such as the regulation of apoptosis (PSEN1, CSRN3 and HBXIP), inflammatory response (TLR1, SERPINA1 and CD40), regulation of glycogenogenesis (IGF2, PPARGC1A and ADIPOQ), intracellular transport (FRAS1, XPO1, YWHAZ, STX7) or growth regulation (IGF2, PTK2B, STAT5A, AVPR1A, ADAM10, ADAM17, RPS6KB1, PROX1 and NTN1). In the blood, the expression levels of about fifty other genes whose expression levels were assessed during the post-weaning or early growth phases made it possible to predict ( $R^2 = 0.86$ ) the G:F during the subsequent growth period. In conclusion, these results confirmed the integrative nature of FE, because the proposed biomarkers were involved both in productive and non-productive functions.

#### **4.6. WP3: Representation of the transit and digestion of nutrients : transposition from a pig to a broiler model**

Sonia Roger (INRA)

Regarding the context of animal production, monogastric livestock systems have to be more sustainable. For this, an increase in the efficiency of feed utilization is necessary. This requires a better understanding and prediction of the nutritional value of the feeds. Many experimental data provide the digestibility of nutrients in various conditions. However, very few studies describe the digestive mechanisms along the digestive tract and for all the nutrients contained in the diet. However, it seems very important to be able to identify the reasons why a diet is digested more efficiently than another and what the causes of differences in digestibility are in the digestive system. Consequently, a modeling approach seems a good way to integrate information available, be able to represent the digestive processes and predict the digestibility.

The work performed aimed the development of a model representing the transit, hydrolysis, fermentation and absorption/excretion of the main nutrients along the digestive tract. A similar structure was adopted for pig and poultry and only selected model parameters were adapted for each species. Parameters for pigs come from existing models and were then modified for the broiler based on biological knowledge and literature data. The transposition of a digestive model for pigs to a broiler has been done step by step to preserve the genericity and make only those modifications that are really needed. Modifications were driven either for biological reasons or to improve the predictive capacity of the model. For now, a generic model has been developed for growing pigs and broilers.

#### **4.7. WP3: An interactive tool to simulate feed use mechanisms and animal response to environmental perturbation**

György Kövér (KU)

Models are useful tools to predict animal response in a certain scenario, however, they need specific platform to run. In our project the aim was to develop software that is available for the users without knowledge on specific statistical or modelling software. The Decision Support Software (DSS) is going to integrate 4 different mathematical models simulating the digestive mechanisms, the post absorptive nutrient partitioning, the animal's response to an unknown environmental perturbation, and the individual variance of a herd or a flock. The poster presents the concept and the potential functions of DSS developed in WP3. Based upon implementation of