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Introduction: Lowering the ratio of ω6:ω3 polyunsaturated fatty acids (PUFA) in maternal diets may promote the health and performance of the sows and their piglets. Yet, the optimal ratio of PUFA in the diet and the mode of transmitting the effects from mother to offspring are largely unknown. The effects of two different ω6:ω3 ratios in the sows' diet on the microRNA profile in the piglets' plasma exosomes were tested.
Methods: Sows were randomly allocated to either the control group (CR, n = 8) receiving a standard diet (ω6:ω3 = 10:1), or the treatment group (LR, n = 8) fed at a lower dietary ω6:ω3 ratio (4:1) from day 28 of gestation until the end of lactation. Plasma samples were collected at weaning (day 26 of life) from 12 piglets (6 per maternal diet group, all from different mothers). Exosomes were isolated using Exoquick™ Precipitation Solution and characterized by Nanoparticle Tracking Analysis. RNAs were purified and small RNA libraries were prepared for miRNA and short non-coding RNAs. The miRNA sequencing was performed using the NextSeq 500 sequencer (Illumina). Data analysis was carried out using the docker4seq package in R studio.
Results: In total, 193 differently expressed miRNA, 145 up-regulated and 48 down-regulated, were identified when comparing the plasma exosomes from piglets born to sows from the CR versus the LR group. These results indicate that exosomes may play a significant role in the transmission of cargo from dietary sources from mother to offspring, which may finally alter gene expression in the infant.
Summary/Conclusion: Different dietary ω6:ω3 ratios in the maternal diet affected the exosomal miRNA composition of their piglets' plasma exosomes. This information may improve the understanding of the mother-to-offspring crosstalk, with impacts on newborn health and future biotechnological perspectives.

PS20.09 | Comparative proteomics of extracellular vesicles subsets isolated from pig seminal plasma

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Introduction: Extracellular vesicles (EVs) of seminal plasma (SP) play a key role in sperm functionality, even fertility. Little is known about the seminal EVs composition. This study evaluated the proteomic profile of EV subsets of pig SP.
Methods: Three pools of SP were analyzed. Each pool contained SP from 3 ejaculates from boars used in artificial insemination. The pools were sequentially centrifuged (3,200g/15 min and 20,000g/30 min at 4°C), and the pellets (washed and solubilized in PBS), and supernatants (0.22μm-filtered and concentrated with Amicon(R)-10K), and were subjected to size-exclusion chromatography for EVs isolation. The fractions 7–9 were selected and mixed. The isolated EVs were analyzed by nanoparticle tracking analysis (NTA) and transmission electron microscopy (TEM). Quantitative proteomics was performed using a SWATH-MS strategy. Proteins were considered quantitatively different with a p ˂ 0.05 and a Log2 fold-change > ±2. Gene Ontology (GO) enrichment analysis was performed using Cytoscape plug-in ClueGO.
Results: The concentration (mean±sd) of EVs was 1.4 × 1012 ± 1.1 × 1012 and 3.7 × 1011 ± 2.2 × 1011 particles/mL in samples from supernatants and pellets, respectively. NTA showed differences in EVs size-distribution between pellet (peaks between 75 and 194 nm) and supernatant samples (more polydisperse, showing peaks between 167 and 354 nm). TEM confirmed the above differences in size. Accordingly, pellets contained larger EVs (enriched in microvesicles, MVs) than supernatant (enriched in exosomes, EXOs). A total of 737 proteins were identified and quantified. Differential quantification analysis revealed that 151 proteins were upregulated and 25 downregulated in MV samples compared to EXO samples. GO enrichment for biological processes revealed that EXO samples were enriched in proteins related to exopeptidase activity, whereas MV samples in those related to cell redox homeostasis and intrinsic apoptosis signaling pathway in response to hydrogen peroxide.
Summary/Conclusion: This study showed clear differences in the proteomic profile between the large and small EV subsets of pig SP.