Research Article
Volume 5:5, 2021

Journal of Animal Health and Behavioural Science

Open Access

Effect of Dietary Supplementation with Linseed Oil and Fish Oil on the Reproductive Traits, Colostrum Composition and Microbial Flora Structure of Sow

Tingting Chu, Guofang Wu, Ye Cheng, Liang Zhong, Zitong Xiao, Zhao Xin, Shiduo Sun, Gongshe Yan and Taiyong Yu*

Department of Animal Science and Technology, Northwest A and F University, Yangling, China

Abstract

The purpose of this study was to supplement linseed oil and fish oil in the sows' diets to explore the effects of different types of oil sources for production, which expected to further investigate microbial mechanistic insight into the different type of oil sources. In this research, Landrace sows were selected and randomly divided into 3 groups: Control group (Ctrl), Linseed Group (LO), Fish Oil (FO). Results showed that there were no obvious changes in the sow's reproductive traits and the ingredients of colostrum. However, the content of Short-Chain Fatty Acids (SCFAs) were improved by LO and FO. Besides, in the absence of changes in the dominant bacteria, the relative abundances of microbial flora presented no significant differences at phylum level and genus level, and our study indicated that the discrepancy in the metabolic pathways of microorganisms in each treatment group further revealed the different adaptive mechanism of microorganisms between different oils sources. In summary, LO and FO had no effect on the sow's reproductive traits and the ingredients of colostrum, but increased the content of SCFAs, and differences in metabolic pathways suggest the adaptive mechanism of microorganisms due to the changes of diets.

Keywords

Linseed oil • Fish oil • PUFA • Microorganism • Sow

Introduction

The growth and health of offspring piglets are affected by sows, due to the fact that the energy of the sow is in a catabolic state during the late pregnancy-lactation [1,2]. At this stage, the sows are susceptible to the adverse effects of drastic changes in physiology and nutritional metabolism, which will affect the health of the sow and piglets [3]. And colostrum provides energy supply and nutrient levels for piglets, In addition, newborn piglets cannot acquire innate maternal antibodies due to the specific structure of the placenta and can only acquire immunity from colostrum through passive immunity [4,5]. The colostrum quality of sows plays a crucial role in the subsequent growth and development of piglets, Intestinal flora affects animal growth and body health and other physiological processes, mainly including energy intake from diet, intestinal barrier function and growth performance, moreover, it also plays a crucial role in animal nutrition metabolism and body immunity. The interaction between the microbial flora in the gastrointestinal tract and the immune system has a certain impact on the health of the host [6,7]. Optimal nutrition of sows during lactation stage of gestation has an important influence on maternal health and fetal growth needs [3]. Therefore, it is of great significance to maintain the sustained and excellent production performance of sows through appropriate nutritional control methods [8]. Polyunsaturated fatty acids (PUFAs) play an important role in the growth development, and health of animals, in reducing early embryonic death, promoting the development of fetal organs and immune system, and improving reproductive performance parameters [9-12]. However, due to the lack of Δ-15-desaturase in higher mammals, n-3 PUFA cannot be synthesized in the body, which consists of the precursor fatty acids linoleic acid and α linoleic acid, and their long chain derivative, and the n-3 PUFA are essential fatty acids for pigs. Plant seeds and animal fats are important sources of long-chain unsaturated fatty acids in sow diets. Linseed oil and fish oil, as the representative sources of animal and plant fats, are rich in PUFA, especially n-3PUFA, and are involved in regulating the metabolism and immune response, and improving meat quality, but few scientific studies focus on the regulation of dietary LO and FO addition in metabolism and microbial flora structure of sow [13-16,17]. The purpose of this investigation was to compare the effect on reproductive performance, colostrum quality and microorganism of sow.

Materials and Methods

The study was approved by the Institutional Animal Care and Use Committee of Northwest A and F University (Yangling, Shaanxi, China), and all operations were carried out according to the university's guidelines for animal research.

Experimental design and diet

A total of 27 French purebred Large White sows (Guangxi, China) of same parity (second parity) were assigned into three groups in a completely randomized design, which consisted of nine replicates (pens). The trial began at 85 days of gestation, and the control group was fed a basal diet with corn starch as the main energy source, while the diet of the treatment group was respectively supplemented with 2% different oil structure (LO: Linseed Oil, FO: Fish Oil) as an energy source in the basal diet. The experimental diet was formulated to meet NRC (2012) requirements, consisting of two stages (gestation diet for 85d-107d gestation, lactation diet for gestation 107d-lactation 21d) and the composition and nutrition levels of diets at different stages were shown in Tables S1 and S2. 2.5 kg/d diet was provided from day 85-107 of gestation, at the day of 90-110 of pregnancy, 3.5 kg was provided, after that, from the day 110 of gestation until delivery, feeding amount was decreased daily

***Address for Correspondence:** Taiyong Yu, Department of Animal Science and Technology, Northwest A and F University, Yangling, China; Tel: +86-029-8709-1017; E-mail: yutaiyong310@nwsuaf.edu.cn*

Received date: November 15, 2021; **Accepted date:** November 29, 2021; **Published date:** December 06, 2021

by 0.5 kg. All the experimental sows were transferred to farrowing pens at one week before the due date. No supplementation diet was provided to the sows at the day of parturition. Feed 2.5 kg on the first day after farrowing and increase by 0.5 kg every day until to 6.5 kg of the sows maximize feed intake at the 10th day after delivery, moreover, water was provided ad libitum throughout the trial.

Animals and housing

The sows, with consistent feeding and management immunization procedures during the whole tentative (trial), which was housed in a 1.8 m × 0.6 m × 0.8 m crate with daily feeding at 7:00 am and 14:00 pm, and the temperature of gestation barn kept at 23-25˚C during gestation period. One week before delivery, all the sows were transferred to the farrowing houses, and the sows and piglets were in the same feeding unit which were consistent with two parts, one of that was the cement concrete slatted floor where the sow lived, while the piglets were placed on the other part with cotton heat pads above, that was made of plastic board, and above of which, the heating lamps were placed; the watering troughs and feeders of the sows and the piglets were provided individually. Postpartum care for sows after delivery, meanwhile, stillbirths, mummified piglets, deformities; scrawny piglets (weight below 0.8 kg) were culled. In addition, newborn piglets were cross-fostered based on litter size, litter carrying capacity, lactation performance. The piglets were supplemented with creep feed at the day of 7, along with the daily pens cleaning and the body condition checking of the pigs.

Phenotypic data and sample collection

Record the reproductive performance-related indicators of the sow after delivery, the birth weight and litter weight of the piglets were weighed within 12 hours after birth. After 21 days of lactation, number of weaned piglets was recorded; furthermore, weaned piglet and litter weight were weighed and recorded. When the sows started to give birth and had not been lactating, wiped the nipples with a clean disinfectant cloth, collected the colostrum secreted from the front, middle and back nipples of each sow, and collected a total of 50 mL for each sow. On the 14th day of lactation of the sow, 0.5-1 g of fresh feces sample was taken into the freezer tube, then was placed in liquid nitrogen for quick freezing, and after that, all the samples were placed in the -80° refrigerator for bacteria microbial detection.

Analysis of colostrum content

Place the low-temperature stored colostrum sample in a 40°C water bath to preheat until it melts. Colostrum composition was determined by the milk composition analyzer (FOSS MilkoScanTM FT-120, FOSS Group, Denmark).

The content of SCFAs in feces

0.5 g of fecal samples were mixed with 4.5 ml sterilized PBS, after shaking evenly, centrifuged for 5 min under 4000 rpm at 4˚C, then 2 ml of supernatant (fecal homogenate) was mixed with 400 μl 0.2 mol/L HCl (5:1, v/v), after that the 2.4 ml mixed liquor was transferred into a new 5 ml micro tube with 480 μl 25% metaphosphoric acid solution (6.4 g/L crotonic acid included), which was then stored 4°C overnight. After centrifugation at 4000 rpm for 5 min, filtered the above mixture with Sterile Syringe Filter (0.45 micron pore size) into chromatographic bottles, sealed it with a sealing film, and stored it at 4°C, and 1 μl of the volatile acid were detected by gas chromatograph according to the protocol provided (Agilent, 7890A, USA).

Analysis of fecal bacteria

Total genome DNA from samples was extracted using CTAB method. DNA concentration and purity was monitored on 1% agarose gels. According to the concentration, DNA was diluted to 1 ng/μL using sterile water. 16S rRNA gene sequencing was performed by the Novogene Bioinformatics Technology (Beijing, China) to perform amplicon pyrosequencing on the Illumina HiSeq PE250 platforms. 341F: CCTAYGGGRBGCASCAG

and 806R: GGACTACNNGGGTATCTAAT primers were used to amplify the V3-V4 hypervariable region of the 16sRNA gene, Mix same volume of 1X loading buffer (contained SYB green) with PCR products and operate electrophoresis on 2% agarose gel for detection, Then, the mixture PCR products was purified with Qiagen Gel Extraction Kit (Qiagen, Germany), after that, Sequencing libraries were generated using TruSeq® DNA PCR-Free Sample Preparation Kit (Illumina, USA) following manufacturer's recommendations, The library quality was assessed on the Qubit@2.0 Fluorometer (ThermoScientific) and Agilent Bioanalyzer 2100 system. At last, the library was sequenced on an Illumina NovaSeq platform and 250 bp paired-end reads were generated. The raw data was spliced and filtered to obtain clean data, and then based on the valid data, OTUs (Operational Taxonomic Units) clustering and species classification analysis et al. were performed.

Statistical analysis

All data in this experiment were processed by SPSS 21.0, an analysis of variance (one-way ANOVA) for the effect of treatment groups was conducted The Least-Significant Difference (LSD) was used for multiple comparisons, and the results were presented as Mean ± SD after data processing. Uparse software (Uparse v7.0.1001), clustered all Effective Tags of all samples, clustered sequences into OTUs with 97% identity, annotated species of OTUs sequence, Mothur method and SSU rRNA database of SILVA138 were used to perform species annotation analysis (set threshold 0.8 ~ 1), and obtained the taxonomic information which was at each classification level; MUSCLE (Version 3.8.31) software was used to perform multiple sequence alignment to obtain the phylogeny relationship of all OTUs representative sequences, Qiime software (Version 1.9.1) was used to calculate Alpha diversity, and the Tax 4 Fun function in the R package was to predict and obtain function annotation information [18-21].

Results

The reproductive performance of sows

Table 1 shows the effects of LO and FO on the reproductive performance of sows. There were no significant differences among experiment groups regarding the number of stillbirths, deformed and weak piglets. It can be seen that the reproductive performance of sows wouldn't be adversely affected by diets with LO and FO addition.

Table 1. Effect of dietary supplementation with linseed oil and fish oil on the reproductive performance of sow.

Item	Ctrl	LO	FO
Total number of litters	13.00 ± 4.18	13.38 ± 4.07	13.33 ± 4.58
Number of live births	11.63 ± 5.29	12.50 ± 4.31	12.00 ± 4.24
Piglet born healthy	11.13 ± 4.88	12.13 ± 4.26	11.78 ± 4.12
No. of weak piglets	0.38 ± 0.52	0.25 ± 0.46	1.00 ± 1.22
deformity	0.13± 0.35	0.38 ± 0.52	0.11 ± 0.33

Still piglets	0.38 ± 0.52	0.50 ± 1.07	0.33 ± 0.71
mummified piglet	1.00 ± 2.83	0.13 ± 0.35	0.11 ± 0.33
sow	3.38 ± 2.26	4.88 ± 1.96	6.78 ± 3.46
boar	7.38 ±4.27	7.25 ± 2.96	5.00 ± 2.34
Litter weight at birth （Kg）	15.53 ± 6.34	16.96 ± 5.81	15.63 ± 5.60
No. of piglets alter fostering （Kg）	13.25 ± 1.98	13.50 ± 1.85	13.33 ± 2.65
Litter weight at birth after fostering （Kg）	17.51 ± 2.93	18.43 ± 3.19	17.00 ± 4.60
No. of weaning piglets	12.00 ± 1.07	11.63 ± 0.92	11.44 ± 1.33
Weaning survival rate （%）	90.45 ± 11.39	87.61 ± 15.05	88.50 ± 17.36
Weaning weight of litter （Kg）	70.86± 2.20	76.44 ± 3.63	71.67 ± 6.36
Litter weight gain （Kg）	49.79 ± 7.42	57.89 ± 6.44	54.67 ± 5.63
Average birth weight of piglets （Kg）	1.34 ± 0.12	1.43 ± 0.20	1.35 ± 0.22
Piglet average weight at weaning （Kg）	5.88 ± 0.34	6.56 ± 0.57	6.31 ± 0.69

Note: Results presented by the Mean ± SD, and different superscripts in the same row indicated differences between treatment groups (P< 0.05)

The colostrum composition of sow

In terms of nutrition of colostrum, supplementing different type's oil during the late pregnancy and lactation had no effect on colostrum composition indexes (Table 2).

Table 2. Effect of dietary supplementation with linseed oil and fish oil on colostrum of sow.

Item (%)	Ctrl	LO	FO
Milk fat	2.24 ± 1.23	2.88 ± 1.19	2.56 ± 0.53
Lactose	11.39 ± 2.75	10.73± 1.12	10.06 ± 0.73
Protein	8.61 ± 1.85	8.16 ± 0.75	7.72 ± 0.49
Solid of non-fat	21.78 ± 5.00	20.59 ± 2.04	19.38 ± 1.34
Salts	1.75 ± 0.40	1.66 ± 0.16	1.56 ± 0.10
Total solids	24.02 ± 3.85	23.48 ± 2.23	21.94 ± 1.73

Note: Results presented by the Mean ± SD, and different superscripts in the same row indicated differences between treatment groups (P<0.05)

The components of SCFAs in sow feces

Table 3 shows that the proportions of SCFAs indexes were different among the experimental groups, the proportions of isobutyric acid, isovaleric acid in the FO were apparently higher than those in the control group (P<0.05), meanwhile, LO group had a richer SCFAs compared with those in control group (P>0.05).

Table 3. Effect of dietary supplementation with linseed oil and fish oil on micro flora alpha diversity in sow.

Item	Ctrl	LO	FO
Observed species	711.33 ± 99.29	764.67 ± 36.75	748.00 ± 29.46
Shannon	6.48 ± 0.50	6.28 ± 0.30	6.52 ± 0.48
Simpson	0.97 ± 0.02	0.96 ± 0.01	0.97 ± 0.01
Chao1	759.00 ± 102.53	848.54 ± 71.04	806.06 ± 37.02
ACE	773.23 ± 105.23	854.10 ± 55.48	821.12 ± 34.22
PD_ whole tree	56.07 ± 7.86	56.30 ± 4.17	56.91 ± 3.06

Note: Results presented by the Mean ± SD, and different superscripts in the same row indicated differences between treatment groups (P<0.05).

Alpha diversity of sow microflora

As shown in Table 4, there were no significant differences among flora diversity indicators (P>0.05).

Table 4. Effect of dietary supplementation with linseed oil and fish oil on micro flora Alpha diversity in sow.

Item (%)	Ctrl	LO	FO
Acetate	53.36 ± 6.44	49.66 ± 4.01	47.44 ± 3.08
Propionate	25.35 ± 3.28	25.59 ± 1.65	25.53 ± 0.18
Isobutyrate	2.11 ± 0.51a	2.63 ± 0.23ab	3.34 ± 0.47b
Butyrate	13.00 ± 3.91	14.85 ± 2.09	14.68 ± 2.45
Isovalerate	3.79 ± 0.90a	4.63 ± 0.47ab	5.72 ± 0.55b
Valerate	2.38 ± 0.56	2.64 ± 0.34	3.28 ± 0.41

Note: Results presented by the Mean ± SD, and different superscripts in the same row indicated differences between treatment groups (P<0.05).

Microbial flora structure

The Figure 1a shows the top 10 species in the experimental group at the level of phylum abundance. Unclassified means the species with no taxonomic annotations. Among them, Firmicutes, Bacteroidota, and Euryarchaeota are the dominant phyla in the Ctrl, LO and FO groups. And the abundance of Firmicutes in the LO group was much more numerous than the nother three groups, while the abundance of Bacteroidota and Proteobacteria was lower than the other groups. While at the genera level, microbiota community was dominated by Terrisporobacter, Treponema, Lactobacillus and Clostridium_sensu_stricto_1, and the LO and FO group showed a lower relative abundance of the genera Lachnospiraceae-NK4A136-group (1.46% in LO group and 1.19% in FO group), and results reveal that there are certain differences in the main dominant bacterial groups among the test groups of different classification levels. Additionally, LEfSe analysis was carried out with the aim of further identifying the differences in OUTs, and 16 apparently different microfloras were observed among experimental groups. The proportion of c-Bacilli, o-RF39, g-unidentified-Lachnospiraceae, s-Clostridium-sp-Culture_27 and s-Olsenella-sp-GAM18 presented higher in LO group, meanwhile, FO group showed a richer relative abundance of the f-F082 and g-Lachnospiraceae-AC2044-group, while the control group remarkably increased nine iconic flora (Figures 1a-1f).

The circle represents the species. The size of the circle is proportional to the relative abundance. The closer the circle is to the vertex, the higher the content of this species in this group. Each small circle at a different classification level represents a classification at that level, and the diameter of the small circle is proportional to the relative abundance.

Clustering analysis of relative abundance of function

Tax 4 Fun was used to compare the predicted functional differences between the groups [22]. The results of comparison between microbes and function database after functional gene annotation analysis are as follows. The results show that: the functional gene annotation of microorganisms is mainly annotated to Metabolism, Genetic-information-procession, Environmental-information-Procession, and Cellar-Processes, among which the abundance of Metabolism is higher, followed by Genetic-information-procession; In order to further investigate the distribution of gene number among different groups, analysis the common and unique information of genes among different groups, the Venn Graph in apparently shows that the number of genes shared among the three test groups is 6297. Besides, the gene abundance of cellular processes and organismal systems in LO group were lower than control group, while the organismal systems presented differences between LO group and FO group (Figures 2a-2d).

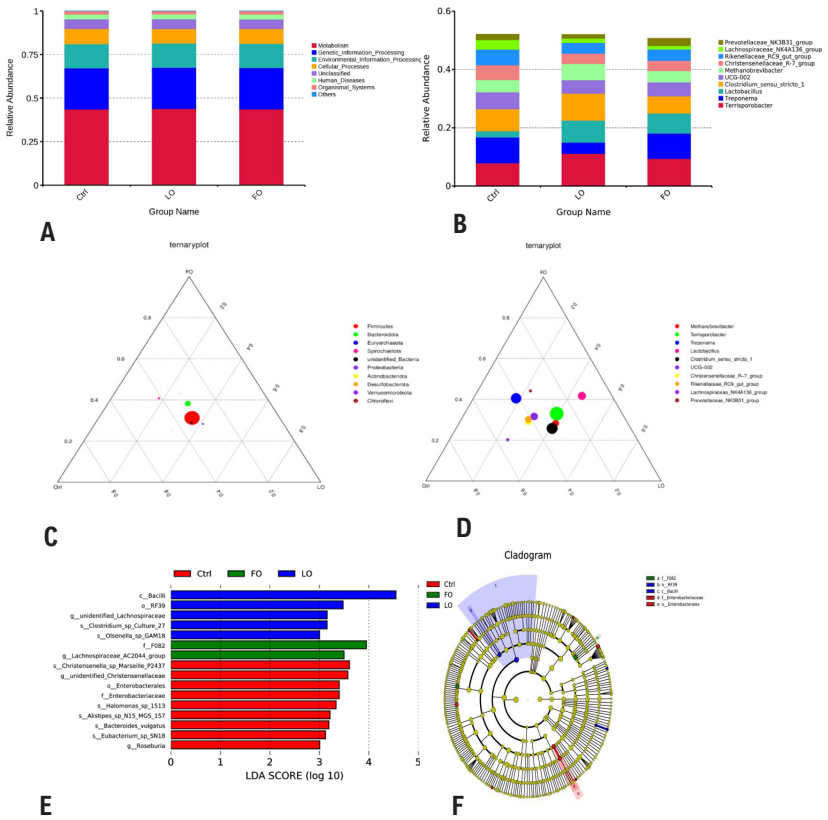

Figure 1 . Effect of dietary supplementation with linseed oil and fish oil on microbial flora structure. (a) Represents the relative abundances of bacteria in fecal of sow at phylum level; (b) Represents the relative abundances of bacteria in fecal of sow at family level; (c) Respectively represent the differences in dominant species in phylum and family level, (d) the three vertices in the figure represent the three sample groups; (e) Represents the LDA value distribution histogram (threshold: >2) and the length of the histogram represents the impact of different species; (f) Represents evolutionary branch diagram and the circles radiating from the inside to the outside represent the classification level from the phylum to the genus in the clade map.

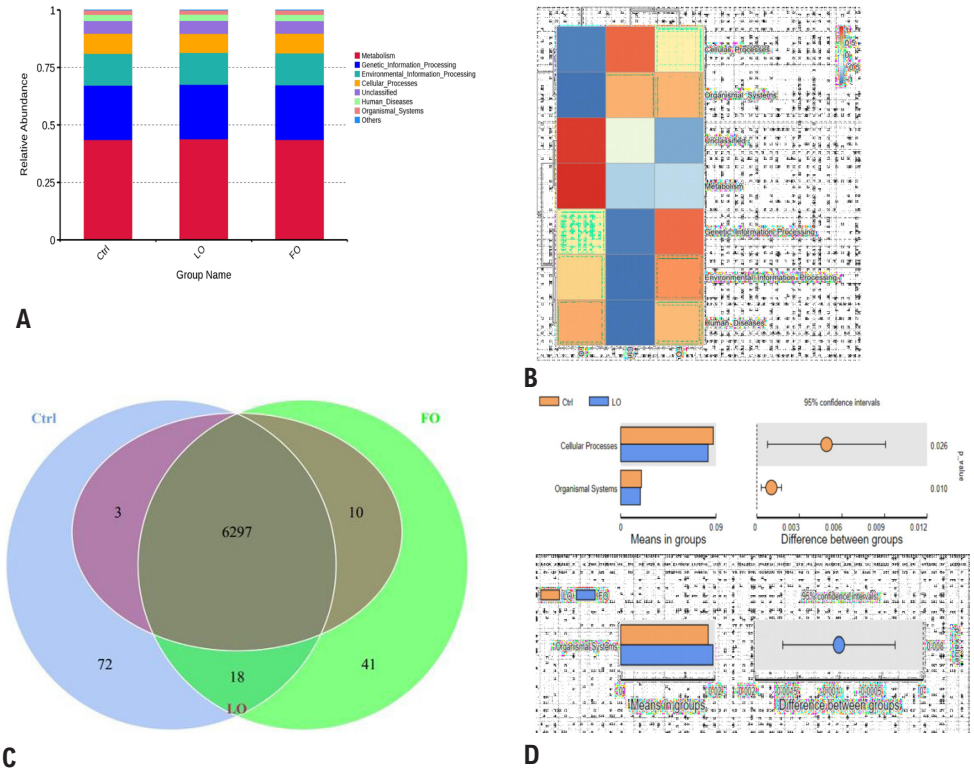

Figure 2 . Effect of dietary supplementation with linseed oil and fish oil on relative abundance of functions. (a) Tax4Fun functional annotation relative abundance histogram at level; (b) Tax4Fun functional annotation clustering heat map at level1; (c)Functional annotation Venn diagram; (d) Relative abundance of significantly.

Discussion

This study found that supplementing sows with dietary fat can increase the weaning litter weight to a certain extent without affecting total litter size and other indicators related to reproductive traits. Among them, linseed oil has a positive effect on the weaning litter weight. Previous studies found that supplementary fat in sow diet can increase the weight of weaning litter to a certain extent [23]. This can be concluded that the addition of fat to the sow's diets improves the sow's energy supplement level, reduces the maternal loss during lactation, and

increases the fat deposition content of the piglet [24-26]. What's more, as the reported described that the mammary gland uses the extra energy provided by fat for milk fat synthesis, increase milk yield, and milk fat content [27-30]. In this research, the milk fat content in oil group was slightly higher than that in the control group. Similar to previous studies, Adding fat to the diets of sows can improve the growth of piglets by increasing the fat and energy secreted by the mammary glands [31,32]. The source of energy and nutrition

for newborn piglets are mainly obtained from colostrum, and the quality of sows' colostrum plays a vital role in the growth and development of the immune system of subsequent piglets. Colostrum is rich in various bioactive substances and nutrients, which can meet the nutritional needs of piglets for growth and development thus promoting the development of piglet's gastrointestinal tract [33]. Intestinal flora plays an important role in digesting diet, maintaining the integrity of the epithelial barrier, regulating immune response, and maintaining intestinal homeostasis [34]. SCFAs (Short-Chain Fatty Acids), the main metabolites of the microbiota, mainly act on energy production, lipogenesis, gluconeogenesis and cholesterol synthesis for host metabolism, and can improve the metabolism of surrounding tissues and stimulate the production of incretin hormones to regulate host lipids metabolism [35]. In this study, LO and FO can increase the content of SCFAs and help maintain intestinal homeostasis. As reported in the previous studies, the intestinal microbiota affects energy balance by influencing the efficiency of calorie acquisition in the diet and storage of energy obtained. Besides, the amount of SCFAs in feces increases, which suggests that the level of microbial energy intake was, improved [36]. Moreover, It was found that dietary intake of n-3 PUFA was associated with the abundance of intestinal micro flora and increased SCFAs content, and the increase in SCFAs content in sows' feces may be related to dietary intake of PUFA [37]. The microbiota, which is involved in the regulation of body metabolism, plays a certain protective role in the body's health and immunity, and affects energy storage and lipid signal transmission [38-41]. What's more, different types of dietary lipids will lead to Differences in intestinal flora [42]. The decrease in microbial diversity implies the occurrence of diseases, and the gut microbial diversity has been considered as a new biomarker of health and metabolic capacity, Alpha Diversity is used to analyze the diversity of the microbial community in the sample, which can reflect the richness and diversity of the microbial community in the sample to a certain extent [43-45]. The results of this study found that the oil group increased the chao 1 index to a certain extent, suggesting that linseed oil and fish oil can improve the abundance of the flora to a certain degree and increase the diversity of the community.

The results of the trial showed that the structure of the fecal microbial flora of sows with different classification levels was adjusted to a certain extent due to the nutritional level of the diet. The sow diet supplemented with oil have some influences on the dominant bacterial in the feces at different classification level, among which FO can increase the abundance of Prevotellaceae to a certain extent, and prevotellaceae is related to the Synthesis of SCFAs, this result reveals that by improving the level of fatty acids in the diet, the body's metabolism can be adjusted and the flora structure has been improved to a certain extent. Under the test conditions, the interaction between dietary microbes and intestinal flora affect organism's metabolism [46].

Conclusion

In conclusion, this study found that supplementing sow diets with linseed oil and fish oil rich in PUFA had no adverse effects on the productive performance and colostrum quality of sow. High-throughput sequencing results show that the microbial abundance in manure has certain differences in the structure of different classification levels to adapt to animal diets changes. The above may be account for the changes in the production performance of the organism under the interaction of the host and the microorganism. In addition, the differences in the metabolic pathways of microorganisms in each treatment group further reveal the adaptive mechanism of microorganisms, and the functional mechanism between different oils present differences, but the mechanism that the effect of microorganisms on sow performance needs to be further explored.

Conflicts of Interest

None of the authors had any personal or financial conflict of interest.

References

1. Duan, X, Tian G, Chen D and Huang L. "Oligosaccharide Supplementation in Diets of Sow and (or) their Offspring Improved Immunity and Regulated Intestinal Bacteria in Piglet1." *J Anim Sci* 97(2019): 4548-4556.
2. Herrera, E, ODuan X, Tian G and Chen D, et al. "Maternal Lipid Metabolism During Normal Pregnancy and its Implications to Fetal Development." *Clin Lipidol* 5(2010): 899-911.
3. Lu, H, Wilcock P, Adeola O and Ajuwon K M. "Effect of Live Yeast Supplementation to Gestating Sows and Nursery Piglets on Postweaning Growth Performance and Nutrient Digestibility." *J Anim Sci* 97(2019): 2534-2540.
4. Prajakta K. "Nutritional and Immunological Importance of Colostrum for the New Born". *J Agric Sci* 143(2010): 469-485.
5. Rooke, JA, Carranca C, Bland, IM and Sinclair, AG, et al. "Relationships between Passive Absorption of Immunoglobulin G by the Piglet and Plasma Concentrations of Immunoglobulin G at Weaning." *Livest Prod Sci* 81(2003): 223-234.
6. Kayama, H, Okumura R and Takeda K. "Interaction Between the Microbiota, Epithelia, and Immune Cells in the Intestine."*Annu Rev Immunol* 38(2020): 23-48.
7. Tremaroli, V, Backhed F. "Functional Interactions between the Gut Microbiota and Host Metabolism."*Nature* 489(2012): 242-249.
8. Tanghe, S and De Smet S. "Does Sow Reproduction and Piglet Performance Benefit from the Addition of n-3 Polyunsaturated Fatty Acids to the Maternal Diet?." *Vet J* 197(2013): 560-569.
9. Lauridsen, C, Theil PK and Jensen SK. "Composition of α-tocopherol and Fatty Acids in Porcine Tissues after Dietary Supplementation with Vitamin E and Different Fat Sources." *Anim Feed Sci Technol* 179(2013): 93-102.
10. Roszkos, R, Tóth T and Mezes M. "Review: Practical Use of n-3 Fatty Acids to Improve Reproduction Parameters in the Context of Modern Sow Nutrition." *Animals* (Basel) 10(2020): 1141.
11. Enke, U, Seyfarth L, Schleussner E and Markert UR. "Impact of PUFA on Early Immune and Fetal Development." *Br J Nutr* 100(2008): 1158-1168.
12. Smits, RJ, Luxford BG, Mitchell M and Nottle MB. "Sow litter size is Increased in the Subsequent Parity when Lactating Sows are Fed Diets Containing n-3 Fatty Acids from Fish Oil."*J Anim Sci* 89(2011): 2731-2738.
13. Greenberg JA, Bell JS, Ausdal WV. "Omega-3 Fatty Acid Supplementation During Pregnancy."*Rev Obstet Gynecol* 1(2008): 162-169.

14. Choo, WS, Birch J and Dufour JP. "Physicochemical and Quality Characteristics of Cold-Pressed Flaxseed Oils." *J Food Compos Anal* 20(2007): 202-211.
15. Raygan F, Mirhosseini N, Akbari E and Bahmani F, et al. "A Comparison Between the Effects of Flaxseed Oil and Fish Oil Supplementation on Cardiovascular Health In Type 2 Diabetic Patients with Coronary Heart Disease: A Randomized, Double Blinded Placebo Controlled Trial." *Phytother Res* 33(2019).
16. Saini, A, Harjai K, Mohan H and Punia R, et al. "Long-Term Flaxseed Oil Supplementation Diet Protects Balb/C Mice against Streptococcus Pneumoniae Infection." *Med Microbiol Immunol* 199(2010): 27-34.
17. Corino, C, Musella M and Mourot J. "Influence of Extruded Linseed on Growth, Carcass Composition, and Meat Quality of Slaughtered Pigs at One Hundred Ten and One Hundred Sixty Kilograms of Live Weight." *J Anim Sci* 86(2008): 1850-1860.
18. Haas BJ, Gevers D, Earl AM and Feldgarden M, et al. "Chimeric 16s Rrna Sequence Formation and Detection in Sanger and 454-Pyrosequenced Pcr Amplicons." *Genome Res* 21(2011): 494-504.
19. Wang Q, Garrity GM, Tiedje JM and Cole JR. "Naive Bayesian Classifier for Rapid Assignment of Rrna Sequences into the New Bacterial Taxonomy." *Appl Environ Microbiol* 73(2007): 5261-5267.
20. Edgar, RC. "Uparse: Highly Accurate Otu Sequences from Microbial Amplicon Reads". *Nat Method* 10(2013): 996-998.
21. Quast, C, Pruesse E, Yilmaz P and Gerken J, et al. "The Silva Ribosomal Rna Gene Database Project: Improved Data Processing and Web-Based Tools." *Nucleic Acids Res* 41(2012): 590-596.
22. Aßhauer, K, Bernd W, Rolf D and Peter MJB. "Tax4Fun: predicting Functional profiles from metagenomic 16S rRNA data." *Bioinformatics* 31(2015), 2882-2884.
23. Rosero, DS, van Heugten E, Odle J and Cabrera R, et al. "Sow and Litter Response to Supplemental Dietary Fat in Lactation Diets during High Ambient Temperatures." *J Anim Sci* 90(2012): 550-559.
24. Brand, H and Kemp B. "Dietary Fat and Reproduction in the Post-Partum Sow". *Soc Reprod Fertil Suppl* 62(2006): 177-189.
25. Lauridsen, C and Danielsen V. "Lactational Dietary Fat Levels and Sources Influence Milk Composition and Performance of Sows and their Progeny."*Livest Prod Sci* 91(2004), 95-105.
26. Shurson, GC, Hogberg MG, DeFever N, and Radecki SV, et al. "Effects of Adding Fat to the Sow Lactation Diet on Lactation and Rebreeding Performance."*Anim Sci J* 62(1986): 672-680.
27. Boyd, RD, Moser BD, Peo ER and Cunningham PJ. "Effect of Energy Source Prior to Parturition and During Lactation on Piglet Survival and Growth And on Milk Lipids." *J Anim Sci* 47(1978): 883-892.
28. Coffey, MT, Yates JA and Combs GE. "Effects of Feeding Sows Fat or Fructose During Late Gestation and Lactation." *J Anim Sci* 65(1987): 1249-1256.
29. Matzat, PD, Hogberg MG, Fogwell RL and Miller ER. "Lactation Performance in High Producing Gilts Fed in Excess of Ad Libitum." (1990).
30. Boyd, RD, Moser BD, Peo ER and Lewis AJ. "Effect of Tallow and Choline Chloride Addition to the Diet of Sows Milk Composition, Milk Yield and Preweaning Pig Performance." *Anim Sci J* 54(1982): 1-7.
31. Farmer, C, Giguere A and Lessard M. "Dietary Supplementation With Different Forms of Flax in Late Gestation and Lactation: Effects on Sow And Litter Performances, Endocrinology, And Immune Response." *J Anim Sci* 88(2010): 225-237.
32. Lauridsen, C and Danielsen. "Lactational Dietary Fat Levels and Sources Influence Milk Composition and Performance of Sows And Their Progeny." *Livest Prod Sci* 91(2004): 95-105.
33. Decaluwé, R, Maes D, Wuyts B and Cools A, et al. "Piglets Colostrum Intake Associates with Daily Weight Gain and Survival Until Weaning." *Livest Prod Sci* 162(2014): 185-192.
34. Kayama, H, Okumura R and Takeda K. "Interaction Between the Microbiota, Epithelia, and Immune Cells in the Intestine." *Annu Rev Immunol* 38(2020): 23-48.
35. Schoeler, M and Caesar R. "Dietary Lipids, Gut Microbiota and Lipid Metabolism." *Rev Endocr Metab Disord* 20(2019): 461-472.
36. Turnbaugh, PJ, Hamady M, Yatsunenko T and Cantarel BL, et al. "A Core Guts Microbiome in Obese and Lean Twins." *Nature* 457(2009): 480-484.
37. Noriega, BS, Sanchez-Gonzalez MA, Daria S and Jonathan C. "Understanding the Impact of Omega-3 Rich Diet on the Gut Microbiota." *Nucleic Acids Res* 41(2016): 1-6.
38. Backhed, F, Ding H, Wang T and Hooper LV, et al. "The Gut Microbiota as An Environmental Factor That Regulates Fat Storage." *Proc Natl Acad Sci* 101(2004): 15718-15723.
39. Yang, H, Yang M, Fang S and Huang X, et al. "Evaluating the Profound Effect of Gut Microbiome on Host Appetite in Pigs." *BMC Microbiol* 18(2018): 215.
40. Besten, GD, Eunen KV, Groen AK and Venema K, et al. "The Role of Short-Chain Fatty Acids in the Interplay between Diet, Gut Microbiota, And Host Energy Metabolism." *J Lipid Res* 54(2013): 2325-2340.
41. Velagapudi, VR, Hezaveh R, Reigstad CS and Gopalacharyulu P, et al. "The Gut Microbiota Modulates Host Energy and Lipid Metabolism in Mice." *J Lipid Res* 51(2010): 1101-1112.
42. Caesar, R, Tremaroli V, Kovatcheva Datchary and Cani P, et al. "Crosstalk Between Gut Microbiota and Dietary Lipids Aggravates Wat Inflammation through Tlr Signaling." *Cell Metab* 22(2015): 658-668.
43. Zhou, P, Zhao Y, Pan Z and Li Y, et al. "Microbial Mechanistic Insight Into the Role of Inulin in Improving Maternal Health in A Pregnant Sow Model." *Front Microbiol* 8(2017): 2242.
44. Clarke SF, Murphy EF, O'Sullivan O and Lucey AJ, et al. "Exercise and Associated Dietary Extremes Impact on Gut Microbial Diversity." *Gut* 63(2014): 1913-1920.
45. Li, b, Zhang X, Guo F and Wu W, et al. "Characterization of Tetracycline Resistant Bacterial Community in Saline Activated Sludge Using Batch Stress Incubation with High-Throughput Sequencing Analysis." *Water Res* 47(2013): 4207-4216.
46. Shen, F, Zheng RD, Sun XQ and Dig WJ, et al. "Gut Microbiotadysbiosis in Patients with Non-Alcoholic Fatty Liver Disease." *Hepatobiliary Pancreat Dis Int* 16(2017): 375-381.

How to cite this article: Chu, Tingting, Guofang Wu, Ye Cheng and Liang Zhong, et al. "Effect of Dietary Supplementation with Linseed Oil and Fish Oil on the Reproductive Traits, Colostrum Composition and Microbial Flora Structure of Sow." *J Anim Health Behav Sci* 5 (2021): 143.