A fermented rapeseed meal additive: Effects on production performance, nutrient digestibility, colostrum immunoglobulin content and microbial flora in sows

Eugeniusz R. Grela a, Anna Czech b, *, Martyna Kiesz b, Łukasz Wlazło c, Bożena Nowakowicz-Dębek c

a Institute of Animal Nutrition and Bromatology, University of Life Sciences in Lublin, Lublin, 20-950, Poland
b Department of Biochemistry and Toxicology, Faculty of Biology, Animal Sciences and Bioeconomy, University of Life Sciences in Lublin, Lublin, 20-950, Poland
c Department of Animal Hygiene and Environment, Faculty of Biology and Animal Breeding, University of Life Sciences in Lublin, Lublin, 20-950, Poland

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Abstract
This study was to assess the effect of fermented rapeseed meal (FRSM) in the diet of sows, taking into account the physiological period (pregnancy or lactation) and reproductive cycle (primiparous or multiparous sows), on production performance, nutrient digestibility, colostrum immunoglobulin content, and microbial flora in sows. The experimental material included 30 primiparous gilts and 30 multiparous sows after their second lactation. The animals in the control groups CG (gilts) and CS (sows) received a standard diet for pregnant or lactating sows, depending on the reproductive period. Experimental groups EG and ES comprised gilts and multiparous sows, respectively, receiving a diet with a 4% share of FRSM in place of soybean meal up to 100 d of gestation. In addition, from 100 d of gestation to 7 d of lactation, the sows in experimental groups received a diet with a 9% share of FRSM, and then again a diet with a 4% share of FRSM until the end of lactation. The addition of 4% to 9% share of a FRSM component in feed significantly improves production parameters, mainly in primiparous gilts, leading to an increase in litter size and in litter weight at 28 d of age. It also helps to improve the digestibility of crude protein, fat, and crude fiber, and positively affects the gut microbiota of sows. Fermentation of rapeseed meal is an effective way to reduce anti-nutrients and to increase the level of lactic acid in the diet. It also stimulates the immune system, which improves piglet health, reducing the severity of diarrhoea and mortality.

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1. Introduction

Researchers are searching for alternative fodder protein sources to soybean meal, especially containing genetically modified organisms, for use in animal diets (Florou-Paneri et al., 2014). One such alternative may be rapeseed meal (Nega and Woldes, 2018), but it has a lower content of protein and certain amino acids — mainly lysine, and increased crude fiber content. Furthermore, it contains glucosinolates and other anti-nutrients (Tripathi and Mishra, 2007). Various technological procedures are used to reduce these unfavourable properties. Fermentation of feed has been shown to be a promising solution (Navarro et al., 2017; Tomaszewska et al., 2019). Owing to its nutritional value (as a valuable source of sulphur-containing amino acids and digestible protein with reduced content of anti-nutrients) and health-promoting properties (as a source of microbes, digestive enzymes, and antioxidant compounds), rapeseed meal that has been fermented and then dried can improve production parameters and
nutrient digestibility, modify the gut microbiota, stimulate metabolic processes, and above all improve animal health (Canibe and Jensen, 2012). The reaction of growing gilts to this feed component in comparison to that of mature sows seems to be an interesting question (Shi et al., 2016). In contrast to multiparous sows, the immune functions of primiparous sows are not yet fully developed (Nega and Woldes, 2018). The quantitative and qualitative composition of the diet of pregnant gilts should meet the nutrient requirements of the foetus and also ensure weight gain in the gilts. Significant differences in somatic development, the maturity of the gastrointestinal tract, and the stability of the microbiota may cause different reactions to fermented protein components in diets of gilts and sows (Yang et al., 2015).

The aim of the study was to assess the effect of fermented rapeseed meal (FRSM) in compound feeds for sows, taking into account the physiological period (pregnancy or lactation) and reproductive cycle (primiparous vs. multiparous sows), on production parameters, nutrient digestibility, colostrum immunoglobulin (Ig) content, and gastrointestinal microbial flora.

2. Materials and methods

The experimental procedures used throughout this study were approved by the II Local Ethics Committee on Animal Experimentation of University of Life Sciences in Lublin, Poland (Resolution No. 21/2016).

2.1. Experimental design

The experimental material comprised 60 Yorkshire sows mated with Danish Landrace boars. These included 30 primiparous gilts and 30 multiparous sows after their second lactation. They were randomly divided into 2 groups of equal size — control and experimental. The animals in control groups Cg (gilts) and Cs (sows) received a standard diet for pregnant or lactating gilts, depending on the reproductive period. Experimental groups Eg and Es comprised gilts and multiparous sows, respectively, receiving a diet with a 4% share of FRSM up to 100 d of gestation. In addition, from 100 d of gestation to 7 d of lactation, the sows in these groups received a diet with a 9% share of FRSM, and then again a diet with a 4% share of FRSM until the end of lactation. During gestation, the sows stayed in pens with 5 animals apiece (group feeding), and from 2 wk before parturition until weaning they were housed in individual boxes (individual feeding). Fermented rapeseed meal was obtained from European Protein AS (Bække, Denmark).

2.2. Animal diets

The sows were fed dry mixtures in accordance with NRC (2012). The composition of the diets for the animals in the control and experimental groups is given in Table 1.

| Item                              | Early pregnancy | Mid-pregnancy to late lactation | Late pregnancy to early lactation |
|-----------------------------------|-----------------|---------------------------------|----------------------------------|
| Cg                                | CgP             | CgPL                            | CgLPL                            |
| Wheat                             | 36.0            | 36.0                            | 36.0                             |
| Triticale                         | 30.0            | 30.0                            | 30.0                             |
| Barley                            | 29.6            | 29.0                            | 29.34                            |
| Oat                               | 29.34           | 29.34                           | 4.04                             |
| Soybean meal, 44% CP              | 6.0             | 3.0                             | 1.0                              |
| Rapsed oil                        | 0.4             | 0.4                             | 0.7                              |
| Salt                              | 0.7             | 0.7                             | 0.7                              |
| Limestone                         | 0.4             | 0.7                             | 0.7                              |
| Monocalcium phosphate             | 0.02            | 0.26                            | 0.26                             |
| L-lysine                          | 0.12            | 0.12                            | 0.12                             |
| DL-methionine                     | 0.08            | 0.08                            | 0.08                             |
| Mineral-vitamin premix            | 2.1             | 2.0                             | 2.7                              |
| Acidiifer                         | 0.4             | 0.4                             | 0.4                              |
| FRSM                              | 4.0             | 4.0                             | 9.0                              |

Cg — control sows in early pregnancy; Eg — experimental sows in early pregnancy; CgL — control sows in mid-pregnancy and late lactation; Es — experimental sows in mid-pregnancy and late lactation; CgL — control sows in late pregnancy and early lactation; EL — experimental sows in late pregnancy and early lactation; FRSM — fermented rapeseed meal. 1 Sows up to 84 d of pregnancy. 2 Sows from 85 to 100 d of pregnancy and from 8 to 28 of lactation. 3 Sows from 101 to 114 d of pregnancy and up to 7 d of lactation. 4 One kilogram of mineral-vitamin premix contained the following: vitamin A 620,000 IU, vitamin D3 80,000 IU, vitamin E 800 mg, vitamin K3 100 mg, vitamin B1 80 mg, vitamin B2 280 mg, vitamin B6 200 mg, vitamin B12 60 mg, vitamin B6 1,000 mg, vitamin H 10,000 mg, niacin 800 mg, vitamin B12 600 mg, choline 15,000 mg, iron 4,000 mg, copper 800 mg, zinc 4,000 mg, manganese 2,000 mg, selenium 10 mg, iodine 30 mg, L-lysine 30,000 mg, L-methionine 10,000 mg, threonine 10,000 mg. 5 One kilogram of acidiifer contained the following: orthophosphoric acid, 320 g, citric acid 110 g, fumaric acid 50 g, propionic acid 45 g, formic acid 45 g, carrier (silicon dioxide) 430 g.

Piglets in the group (as severity of diarrhoea), the duration of diarrhoea in days, and faecal score were calculated.

A 10-ml sample of colostrum was taken from the teats of 6 sows in each group 5 to 6 h after parturition. Samples were stored at about –20 °C until analysis.

Digestibility tests were performed during 3 periods of the reproductive cycle: early pregnancy (34 to 40 d), late pregnancy (104 to 110 d), and late lactation (19 to 25 d).

Before the experiment, chemical and microbiological analysis of FRSM was performed, and then samples of the diets were collected twice in each feeding period for chemical analyses.

2.4. Laboratory analyses

Dry matter, crude ash, crude protein, crude fat, and crude fiber were determined in the diets, FRSM and faeces (AOAC, 2012). The energy value of the diets was calculated according to a formula given by Kirchgessner and Roth (1983). The content of calcium, sodium, iron, copper and zinc was determined by atomic absorption spectrometry, and total phosphorus content was determined according to Fiske and Subbarow (1925). Also determined in the diet and fermented components were the content of phytate phosphorus according to Oberleas (1971), lactic acid content according to Taylor (1996), glucosinolates according to PN-ISO 10633-1, 2000 and tannins according to Canbaj et al. (2001).
About 0.3 kg of faeces was collected in the morning from 6 animals in each group. The faeces were weighed and samples from 3 consecutive days were collected in a covered container with a few drops of sulphuric acid. Next, the faeces were mixed and 10 g of each sample was taken for determination of total protein content (AOAC, 2012). The remainder was oven-dried (60 °C/72 h) to a constant weight. From each such portion of dried faeces, finely ground in a mill, 2 samples were weighed out for analysis. The concentrations of nutrients and 4 mol/L HCl-insoluble ash were determined (AOAC, 2012).

The calculation was made as follows:
Nutrient apparent total tract digestibility = 100 – 100 × [(Indicator content in diet × Nutrient content in faeces)]/[(Indicator content in faeces × Nutrient content in diet)]

2.4.2. Colostrum analysis

The titres of IgA, IgG and IgM antibodies and interleukin 6 (IL-6) in the colostrum were quantified using ELISA kits specifically for porcine IgA, IgG and IgM (Bethyl Laboratories, Inc., Montgomery, TX) and IL-6 (Elabscience Biotechnology Co., Ltd).

2.4.3. Microbiological analysis of gastrointestinal contents

During the digestibility tests, faeces from 6 sows from each group were sampled twice (2 and 5 d of the experiment), directly from the anus, for microbiological analyses. The material was cooled to 6 to 8 °C and transported to the laboratory. Then one gram was weighed out from each sample and inoculated into 9 mL of Ringer’s solution with Tween 80 and homogenized. Decimal dilutions of each material were made according to ISO 6887-1: 2017. Each dilution was plated on previously prepared sterile solid media in the amount of 100 μL and then incubated according to standards.

The following were determined in the material: total bacterial number (PN-EN ISO 4833-2:2013/AC:2014), total number of yeasts and moulds (PN-ISO 21527-1/2; PN-ISO 4832, 2007; PN-ISO-16649-2), total number of coliform bacteria (PN-ISO 4832, 2007), total number of Escherichia coli bacteria (PN-ISO-16649-2) and total number of Clostridium perfringens bacteria (PN-EN ISO 7937). Each culture on solid substrates was conducted in duplicate. The number of microorganisms was expressed as colony forming units (cfu) per gram of test material. The result for one animal was expressed as the mean of replicates of the cfu number per g of faeces.

2.5. Statistical analysis

The data on production parameters, nutrient digestibility, and microbial flora of faeces in sows were subjected to statistical evaluation by a 2-factor analysis with interaction, taking into account the following factors:

\[ y_{ijk} = C_i + F_j (C \times F)_{ij} + e_{ijk}, \]

where: \( y_{ijk} \) observations; \( C_i \) effect of reproductive cycle (primiparous gilts or multiparous sows); \( F_j \) effect of animal feeding group (control or experimental — the effect of dried FRSM); \( (C \times F)_{ij} \) effect of interaction between reproductive cycle and diet. Statistical significance between treatments was based on \( P < 0.05 \). Analyses were performed in the GLM procedure of SAS 9.4 (SAS Institute, Cary NC).

3. Results

The content of nutrients and bioactive substances in the FRSM and diets for sows are given in Table 2.

The content of basic nutrients in the sow diets in each feeding period (early pregnancy, late pregnancy and late lactation) was similar for animals in the control and experimental groups and was consistent with NRC (2012). Phytate phosphorus content in the diets containing the fermented component FRSM was significantly lower than that in the diet for the control groups. The reverse was noted for lactic acid content. Only in the diet with a 9% share of FRSM, glucosinolate content was significantly higher than that in the control (\( C_{\text{UL}} \) \( P < 0.05 \)).

3.1. Production parameters and nutrient digestibility

The addition of FRSM significantly increased the number of piglets at 28 d of age, particularly in the case of primiparous gilts (\( P = 0.029 \)). However, it caused a reduction in the weight of piglets at 28 d of age (\( P = 0.084 \)), with significant differences noted in the case of multiparous sows (\( P = 0.041 \)). Litter weight during this period was significantly higher in the gilts receiving the fermented component (\( E_C \)) in comparison to in the control group (\( C_C \)). The mortality of piglets at 28 d of age from sows in the \( E_2 \) and \( E_5 \) groups, whose diet contained FRSM, was significantly lower than that in the animals in groups \( C_2 \) and \( C_5 \), respectively (Table 3). Piglets from sows in the experimental groups (\( E_2 \) and \( E_5 \)) had significantly less severe diarrhoea and significantly fewer days with diarrhoea than the piglets from the control groups. In the piglets from the experimental groups, faecal score was improved relative to the control group (Table 3).

The results of the experiment showed that the reproductive cycle (\( R \) – primiparous vs. multiparous sows) had a significant effect on piglet weight at birth and litter weight at 28 d, as it was higher in piglets from multiparous sows. It also affected piglet mortality and the severity of diarrhoea, which were lower in piglets from multiparous sows (Table 3).

There was an interaction between the reproductive cycle and the addition of the fermented component (\( F \times R \)) for the number of piglets at 28 d of age (\( P = 0.025 \)), litter weight at 28 d of age (\( P = 0.048 \)), mortality (\( P = 0.039 \)), diarrhoea severity (\( P = 0.026 \)), days with diarrhoea (\( P = 0.032 \)) and faecal score (\( P = 0.027 \)) (Table 3).

The FRSM improved the apparent total tract digestibility of fat and crude fiber in sows during late pregnancy and lactation (Table 4). A significantly higher digestibility coefficient for crude protein and crude fiber was noted for gilts during late pregnancy and lactation whose diet included FRSM compared with the control group (\( C_C \)). In multiparous sows receiving a diet with FRSM, a higher digestibility coefficient was found for crude fiber in late pregnancy (\( P = 0.041 \)) and for crude protein during lactation (\( P = 0.046 \)) (Table 4).

The reproductive cycle had a significant effect on the apparent total tract digestibility of crude fiber during early pregnancy, crude fat and crude fiber during late pregnancy, and crude protein, crude fat and crude fiber during lactation. There was an interaction between the addition of the fermented component and the reproductive cycle (\( F \times R \)) for the digestibility of crude fat in sows in late pregnancy and lactation and for digestibility of crude fiber during lactation (Table 4).

3.2. Immunoglobulins in colostrum

The content of IgG and IgA in the colostrum of sows receiving a diet with FRSM, irrespective of the reproductive cycle, was significantly higher than that in the animals in the control groups. An interaction was found between the addition of the fermented component and the reproductive cycle (\( F \times R \)), as well as an effect of the reproductive cycle on the content of IgG in the colostrum (their titre was lower in multiparous sows) (Table 5).
Table 2
Content of nutrients and bioactive substances in diets (g/kg).

| Item                      | FRSM                  | Early pregnancy<sup>1</sup> | Mid-pregnancy to late lactation<sup>2</sup> | Late pregnancy to early lactation<sup>3</sup> |
|---------------------------|-----------------------|-----------------------------|---------------------------------------------|---------------------------------------------|
| Dry matter                | 882.7                 | 883                         | 882                                         | 889                                         |
| Crude ash                 | 78.9                  | 50.6                        | 49.8                                        | 52.4                                        |
| Crude protein             | 291.8                 | 150.3                       | 150.1                                       | 170.4                                       |
| Ether extract             | 31.7                  | 32.2                        | 32.3                                        | 27.2                                        |
| Crude fiber               | 91.5                  | 73.5                        | 74.4                                        | 48.5                                        |
| Metabolizable energy, MJ/kg | 12.27               | 12.3                        | 12.3                                        | 12.6                                        |
| Total phosphorus          | 9.09                  | 5.23                        | 5.19                                        | 5.63                                        |
| Phytate phosphorus        | 5.73                  | 3.14<sup>a</sup>            | 2.31<sup>b</sup>                            | 3.79<sup>a</sup>                            |
| Calcium                   | 8.05                  | 7.45                        | 7.43                                        | 8.62                                        |
| Sodium                    | 2.26                  | 2.04                        | 2.03                                        | 1.98                                        |
| Zinc, mg/kg               | 66.98                 | 142.5                       | 144.3                                       | 148.7                                       |
| Copper, mg/kg             | 6.67                  | 18.61                       | 18.42                                       | 20.30                                       |
| Iron, mg/kg               | 149.2                 | 145.8                       | 145.2                                       | 165.3                                       |
| Glucosinolates, μmol/g    | 6.37                  | 0.001                       | 0.002                                       | 0.001                                       |
| Tannins                   | 4.76                  | 2.19                        | 2.34                                        | 2.21                                        |
| Milk acid, mmol/kg        | 50.4                  | 17.1<sup>b</sup>            | 51.2<sup>b</sup>                            | 16.9<sup>a</sup>                            |

FRSM = fermented rapeseed meal; C<sub>E</sub>P = control sows in early pregnancy; E<sub>E</sub>P = experimental sows in early pregnancy; C<sub>MPL</sub> = control sows in mid-pregnancy and late lactation; E<sub>EMPL</sub> = experimental sows in mid-pregnancy and late lactation; C<sub>CLPL</sub> = control sows in late pregnancy and early lactation; E<sub>ELPL</sub> = experimental sows in late pregnancy and early lactation.

<sup>a, b, c</sup> Different superscripts within a row denote statistically significant differences at <i>P</i> < 0.05.

1 Sows up to 84 d of pregnancy.
2 Sows from 85 to 100 d of pregnancy and from 8 to 28 d of lactation.
3 Sows from 101 to 114 d of pregnancy and up to 7 d of lactation.

Table 3
Production performance of gilts and sows.

| Item                      | Gilts | Sows | P-value | F  | R  | F × R |
|---------------------------|-------|------|---------|----|----|-------|
| Number of piglets in litter | 16.25 | 15.77 | 0.082   | 88.23 | 89.31 | 0.053           |
| Number of live born piglets | 14.25 | 14.43 | 0.089   | 87.54 | 89.64 | 0.042           |
| Number of piglets at 28 d of age | 11.25 | 13.17 | 0.029   | 94.54 | 95.11 | 0.058           |
| Weight of piglets at birth, kg | 1.12  | 1.17  | 0.012   | 12.27 | 13.25 | 0.014           |
| Litter weight at 28 d of age, kg | 67.39 | 73.36 | 0.031   | 57.04 | 59.02 | 0.016           |
| Mortality up to 28 d of age, % | 21.05 | 21.99 | 0.005   | 85.14 | 86.71 | 0.019           |
| diarrhoea incidence, % | 28.2  | 11.5  | 0.001   | 95.12 | 95.38 | 0.033           |
| Days with diarrhoea | 6.2   | 3.4   | 0.014   | 2.1   | 1.4   | 0.006           |
| Faecal score | 2.1   | 1.4   | 0.048   | 1.6   | 1.3   | 0.056           |

CG = control gilts; EG = gilts receiving feed with fermented rapeseed meal (FRSM); CS = control sows; ES = sows receiving feed with FRSM; F = effect of FRSM; R = effect of reproductive cycle; F × R = interaction between experimental factor (FRSM) and reproductive cycle.

Table 4
Coefficients of apparent total tract digestibility (%) of nutrients in sows.

| Item                      | Gilts | Sows | P-value | F  | R  | F × R |
|---------------------------|-------|------|---------|----|----|-------|
| Crude protein             | 88.23 | 89.31 | 0.089   | 88.94 | 90.83 | 0.053           |
| Ether extract             | 78.15 | 78.82 | 0.010   | 80.27 | 81.56 | 0.017           |
| Crude fiber               | 67.48 | 68.19 | 0.075   | 78.35 | 78.94 | 0.025           |
| Nitrogen-free extract     | 95.12 | 95.38 | 0.017   | 95.89 | 96.13 | 0.019           |
| Late pregnancy (104 to 110 d) | 87.54 | 89.64 | 0.042   | 88.05 | 88.49 | 0.094           |
| Ether extract             | 80.29 | 81.33 | 0.096   | 81.72 | 83.92 | 0.103           |
| Crude fiber               | 54.41 | 57.47 | 0.004   | 63.32 | 66.73 | 0.035           |
| Nitrogen-free extract     | 94.54 | 95.11 | 0.010   | 95.05 | 95.64 | 0.084           |

CG = control gilts; EG = gilts receiving feed with fermented rapeseed meal (FRSM); CS = control sows; ES = sows receiving feed with FRSM; F = effect of FRSM; R = effect of reproductive cycle; F × R = interaction between experimental factor (FRSM) and reproductive cycle.
3.3. Results of microbiological analysis of the gastrointestinal tract

The addition of FRSM caused a change in the microbiological composition of the digestive tract contents, particularly in pregnant gilts (Table 6). A significant reduction was noted in the total bacterial number, including coliform bacteria and anaerobic *C. perfringens*, as well as a reduction in the number of *E. coli* in the total number of fungi, as compared to the control group (Cg). In multiparous sows, only the total bacterial number in the faeces was reduced (P = 0.027). The reproductive cycle affected the total number of *E. coli* (P = 0.0039) only in the period of late pregnancy (with a lower number in multiparous sows). An interaction was noted between the addition of the fermented component and the reproductive cycle (F × R), as well as an effect of the reproductive cycle (R) on the total number of coliforms (Table 6).

### 4. Discussion

Due to its high content of essential amino acids, including sulphur-rich methionine, as well as its relatively high content of phosphorus, rapeseed meal is a good protein component of diets for monogastric animals. Research indicates that it can partially replace soybean meal in the diet of sows and piglets (Florou-Paneri et al., 2014). However, its use is limited by the presence of numerous anti-nutrients, such as glucosinolates, tannins and phytate compounds (Tripathi and Mishra, 2007), whose effects include reduced digestibility and nutrient utilization. Fermentation has been shown to be an effective way to reduce undesirable substances in rapeseed meal, even by over 80% (Chiang et al., 2009; El-Batal and Abdel Kareem, 2001; Vig and Waila, 2001; Wang et al., 2010). This effect was also observed in our experiment. The diets with FRSM had a relatively low level of glucosinolates and tannins. Also, the content of phytate phosphorus in the diets with FRSM was significantly lower than that in the control group. According to Wang et al. (2010), this can be attributed to microorganisms accompanying the fermentation process, as they are a source of the enzyme phytase, which breaks down phytate complexes (Tripathi and Mishra, 2007). According to Schone et al. (2001), low content of anti-nutrients in diets for lactating sows is associated with a reduction in their content in the milk. In our experiment, this resulted in an improvement in the condition of newborn piglets from sows fed a diet with FRSM, i.e. a reduction in the incidence and severity of diarrhoea and in mortality. The increase in litter size and in litter weight at 28 d of age of piglets from sows whose feed contained FRSM (mainly from group Eg) may also have been due to stimulation of immune processes in the sow through an increase in the titre of Ig (IgG and IgA) in the colostrum. Such a relationship has been reported by Krakowski et al. (2002).

According to Quesnel et al. (2012), the increased level of Ig in the colostrum of sows receiving a fermented diet is the immune system's response to a foreign antigen of microbial origin. The fermentation process enriches the diet with short-chain fatty acids, vitamins and enzymes, thereby stimulating the gut environment of pigs to develop beneficial gut microflora (including *Lactobacillus*, lactic acid bacteria or *Bifidobacterium*) (Scholten et al., 1999). This review deals with the properties of fermented diets and their

### Table 6

**Microbiological composition (cfu/g) of sow faeces.**

| Item                  | Gilts          |  | Sows          |  | P-value | F  | R  | F × R |
|-----------------------|----------------|---|----------------|---|---------|----|----|-------|
|                       | Cg             | Eg | Cg             | Eg |         |    |    |       |
| Total number of bacteria Early pregnancy | 2.6 × 10^6 | 3.3 × 10^6 | 0.543 | 2.6 × 10^7 | 2.4 × 10^7 | 0.768 | 0.436 | 0.098 | 0.333 |
| Late pregnancy | 9.7 × 10^6 | 4.5 × 10^6 | 0.035 | 3.3 × 10^7 | 5.6 × 10^7 | 0.027 | 0.032 | 0.111 | 0.129 |
| Late lactation | 1.5 × 10^7 | 9.3 × 10^7 | 0.234 | 1.2 × 10^8 | 2.9 × 10^8 | 0.123 | 0.187 | 0.454 | 0.206 |
| Total number of fungi Early pregnancy | 3.8 × 10^4 | 4.7 × 10^3 | 0.053 | 1.4 × 10^4 | 2.0 × 10^4 | 0.435 | 0.311 | 0.074 | 0.101 |
| Late pregnancy | 3.8 × 10^4 | 2.0 × 10^3 | 0.037 | 1.0 × 10^4 | 4.9 × 10^4 | 0.209 | 0.069 | 0.542 | 0.322 |
| Late lactation | 5.0 × 10^3 | 3.0 × 10^2 | 0.222 | 4.7 × 10^2 | 1.3 × 10^2 | 0.541 | 0.407 | 0.265 | 0.201 |
| Total number of coliforms Early pregnancy | 1.3 × 10^4 | 3.1 × 10^3 | 0.294 | 3.5 × 10^5 | 3.0 × 10^6 | 0.656 | 0.381 | 0.045 | 0.276 |
| Late pregnancy | 4.2 × 10^7 | 1.6 × 10^7 | 0.041 | 2.9 × 10^7 | 4.6 × 10^7 | 0.099 | 0.089 | 0.143 | 0.078 |
| Late lactation | 1.0 × 10^3 | 4.0 × 10^2 | 0.657 | 8.0 × 10^2 | 1.0 × 10^3 | 0.735 | 0.657 | 0.444 | 0.231 |
| Total number of *Escherichia coli* Early pregnancy | 1.7 × 10^3 | 1.2 × 10^4 | 0.098 | 2.5 × 10^7 | 4.7 × 10^8 | 0.407 | 0.139 | 0.121 | 0.137 |
| Late pregnancy | 3.8 × 10^4 | 1.6 × 10^4 | 0.035 | 3.5 × 10^5 | 4.5 × 10^6 | 0.188 | 0.044 | 0.039 | 0.089 |
| Late lactation | 1.0 × 10^4 | 3.4 × 10^3 | 0.412 | 7.3 × 10^4 | 1.3 × 10^5 | 0.344 | 0.213 | 0.411 | 0.210 |
| Total number of anaerobic *Clostridium perfringens* bacteria Early pregnancy | 1.0 × 10^4 | 2.0 × 10^4 | 0.071 | 5.0 × 10^4 | 7.1 × 10^4 | 0.289 | 0.293 | 0.100 | 0.109 |
| Late pregnancy | 1.3 × 10^5 | 3.3 × 10^4 | 0.048 | 4.9 × 10^5 | 4.4 × 10^6 | 0.456 | 0.105 | 0.182 | 0.117 |
| Late lactation | 3.0 × 10^5 | 1.9 × 10^4 | 0.426 | 1.0 × 10^6 | 2.3 × 10^7 | 0.109 | 0.230 | 0.187 | 0.222 |

Cg – control gilts; Eg – gilts receiving feed with fermented rapeseed meal (FRSM); Cs – control sows; Es – sows receiving feed with FRSM; F – effect of FRSM; R – effect of reproductive cycle; F × R – interaction between experimental factor (FRSM) and reproductive cycle.
effects on growth performance and gastrointestinal environment of pigs. In addition, some possible modes of action are hypothesized. Starch- and sugar-rich liquid co-products have a high potential for fermentation during storage. Soaking compound feed in water is another means of achieving a fermented diet. These diets have a pH between 3.5 and 4.5, high levels of lactic acid, and, to a lesser extent, acetic acid and alcohol (Scholten et al., 1999). Fermented diets seem to improve the growth performance of pigs compared with non-fermented diets (Canibe and Jensen, 2010). The precise reasons for this are not yet clear; however, some hypotheses have been advanced. A limited number of studies indicate that fermented diets reduce the gastric pH and the number of coliform bacteria in the gastrointestinal tract compared with non-fermented diets (Hong et al., 2009). Furthermore, there are some indications that fermented diets may positively affect pancreatic secretion, villus architecture, and the digestibility and absorption of dietary nutrients (Canibe and Jensen, 2012). Fermented diets may reduce the physical activity of pigs. More specific studies on the effect and modes of action of fermented diets are needed so that firmer conclusions can be drawn (Scholten et al., 1999). The presence of a large amount of lactic acid or other organic acids in fermented feed causes acidification of the intestinal contents, stimulates production of antibacterial bacteriocins, increases intestinal enzyme activity, and, above all, stimulates local immunity in the gastrointestinal mucosa (Krakowski et al., 2002). The increase in IgG content in the colostrum of sows, both primiparous and multiparous (groups E2 and E3), indicates that passive immunity is provided to the neonate, improving its chance of survival. Class G Ig account for about 75% to 80% of all Ig and are the core of the active humoral response (van de Ligt et al., 2002). Immunoglobulins of this class and class A are able to penetrate the placenta into the body of piglets through active transport mechanisms, and can also enter the colostrum (van de Ligt et al., 2002; Czech et al., 2010). This is important in the postnatal development of piglets, as antibody synthesis during this period is naturally impaired. Low IgM levels and nearly undetectable levels of IgA and IgG are observed in them (Quesnel et al., 2012). During this period, maternal class G Ig perform a protective function. The role of class A Ig, similar to that of lactic acid bacteria, is local protection of the mucosa against pathogens, as well as participation in the systemic response (Canibe et al., 2007; Dlamini et al., 2017).

The proper functioning of the digestive system is assumed to be largely dependent on the maintenance of balanced bacterial flora in the gut (Song et al., 2010). According to Grela et al. (2018), feeding pigs with fermented liquid feed improves gastrointestinal function relative to animals receiving a dry diet. This was confirmed in our experiment. The faeces of sows receiving a diet with FRSM, especially primiparous gilts in late pregnancy, contained significantly fewer total bacteria and fungi, as well as coliform bacteria (including E. coli), and anaerobic C. perfringens bacteria. This is due in part to the reduction in pH and increase in the amount of lactic acid and other volatile fatty acids in the intestinal contents, as well as to the reduction in the number of Enterobacteriaceae (Canibe and Jensen, 2012a,b). In addition, probiotic microorganisms accompanying the fermentation process, as natural modifiers of intestinal microflora, are able to block the receptor sites located on the intestinal wall surface and on pathogenic bacteria, such as Salmonella, and potentially pathogenic bacteria, such as E. coli. As a result, microorganisms entering the gastrointestinal tract cannot bind to specific receptors and are excreted from the body with no adverse effect on the animal’s health (Perdigón et al., 2001; Sugiharto et al., 2015). No such phenomenon was observed in sows in early pregnancy, possibly because the share of the fermented component in the diet was too small (4%). It should also be noted that this component was not as effective in the multiparous sows as in the gilt. This may have been due to the maturity of the gastrointestinal tract and its microflora in the older sows, which is too stable to be modified by this amount of the fermented component. Differences in gastrointestinal function between primiparous and multiparous sows have been reported by Yang et al. (2009).

The reduction in the content of anti-nutrients (phytate phos- phorus) in the sow diets during pregnancy and lactation through the use of FRSM, as well as the development of beneficial gut microflora, improved the digestibility of nutrients, especially crude protein, crude fat and crude fiber. This effect was evident during late pregnancy and lactation, when the share of FRSM in the diet was increased by more than 50% (from 4% to 9%). According to Cho et al. (2011) and Upadhaya et al. (2015), improved nutrient digestibility in pigs receiving feed with a fermented component, which is a source of lactic acid bacteria, may largely be due to increased production and activity of hydrolytic enzymes such as amylase in the gut (Canibe and Jensen, 2012a,b; Liao and Nyachoti, 2017).

Probiotic microorganisms involved in the fermentation process may enhance digestion (Sugiharto et al., 2015). The microorganisms appearing in the fermentation process also synthesize microbial phytase, which improves the bioavailability not only of minerals (Czech and Grela, 2004), but also of certain amino acids (Song et al., 2010). The reduction in phytic acid as a result of fermentation was in line with results obtained by El-Batal and Abdel Karem (2001), who found that the microorganisms accompanying the fermentation process (mainly Aspergillus niger) were able to produce phytase, thereby reducing the content of phytate compounds by as much as 86%. Vig and Walia (2001) also drew attention to the significant reduction (by over 25%) in crude fiber in FRSM, which is particularly beneficial in the diet of young monogastric animals. This is confirmed by the improved weight gain in piglets from the experimental groups.

5. Conclusions

The addition of 4% to 9% share of a FRSM component in diets significantly improves production parameters, mainly in primipa- rous gilts, leading to an increase in litter size and in litter weight at 28 d of age. It also helps to improve the digestibility of crude protein, fat and crude fiber and positively affects the gut microbiota of pigs (especially gilts during late pregnancy).

Fermentation of rapeseed meal is an effective way to reduce anti-nutrients (phytate phosphate and glucosinolates) and to increase the level of lactic acid in compound feeds, as well as to stimulate the immune system, which improves the health of pig- lets, reducing diarrhoea severity and mortality.

Conflict of interest

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work, there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the content of this paper.

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6. ACKNOWLEDGEMENTS

7. REFERENCES