SUMMARY

"NUTRITIONAL-METABOLIC IMPLICATIONS OF PROBIOTICS IN PIGS"

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KEYWORDS: *pigs, probiotics, synbiotics, pregnant sows, lactating sows, suckling piglets, weaned piglets, blood biochemical profile, haematological profile.*

The thesis entitled "*Nutritional-Metabolic Implications of Probiotics in Pigs*" comprises 203 pagesand is structured in two parts.

Part I - The bibliographic study accounts for about 25% of the total work and presents the main innovations in prebiotics, probiotics and synbiotics use as growth promoters in pigs, microbiota of the digestive tract in pigs, effects, safety and results of the use of probiotics and synbiotics in therapy and prophylaxis in intensive rearing of pigs. The research is based on 153 references and 4 scientific worksas the first author published in indexed databases and presented in the bibliography. Also, 6 tables of interest in the specific literature are used with regard to prebiotics, probiotics and symbiotics respectively, scientific name, classification, mode of action, and their effects.

Part IIrepresents 75% of the work, and the results of the nutritional-metabolic monitoring, the haematological and biochemical blood profile and the characterization of the performance of piglets' groups are presented in 115 tables and 14 figures.

The purpose of the paper

The ban on the use of antibiotics in animal feed since 1 January 2006 has led to research into the use of natural growth promoters as a viable alternative to the use of antibiotics. As such, the aim of the PhD thesis was to improve knowledge of the use as growth promoters of probiotic and symbiotic in pigs and to evaluate the benefits for breeding sows and piglets until weaning. At the same time, the research sought to improve the hematological and biochemical blood parameters (nutritional-metabolic effects) by growth promoters (probiotics and synbiotics) in breeding sows that are highly productive buthave high metabolic imbalances.

Materials and methods

The researches were carried out during 2010-2015, in a closed circuit selection farm in Ialomita County, which obtains high-performance F1gilts (TOPIGS-MA x Landrace).

The effect of supplementing fodder ratios of the sows with the **BioPlus 2B** probiotic product on the last part of pregnancy and on piglets from birth to weaning was monitored.

Also, the **Biomin IMBO** (probiotic + prebiotic) symbiotic product was studied in order to evaluate the differences between a simple probiotic and a probiotic in combination with the nutritional substrate (prebiotic).

Hematologic (hemoglobin, hematocrit, CHEM, red and white blood cells) and **biochemical** (total proteins, albumin, total globulins, glucose, triglycerides, total cholesterol, HDLP cholesterol, urea, creatinine, bilirubin, Ca, P, Mg, GOT, GPT, GGT, PAL, CPK) determinations have been conducted in sows and piglets, and **performances of sows and piglets**(number of born and dead piglets, number of weaned piglets, weight of the born and weaned piglets, average of piglets' weights at birth and weaning, daily average gain, diarrheal score) were observed.

Results and discussions:

Personal research is structured in four subchapters, namely:

- the effect of supplementation of *pregnant sows'* ratios with BioPlus 2B probiotic preparation;

- the effect of supplementation of *lactating sows'* ratios with BioPlus 2B probiotic preparation;

- the effect of supplementation of *pregnant sows'* ratios with Biomin IMBO synbiotic product;

- the effect of supplementation of *lactating sows'* ratios with the Biomin IMBO synbiotic preparation.

1.The effect of supplementation of pregnant sows' ratios with BioPlus 2B probiotic preparation

In order to see to what extent the metabolism of pregnant sows is influenced by probiotics administered between the 85th day of gestation and farrowing, we used BioPlus 2B commercial product used in intensive livestock rearing, for higher feed conversion, decreasing diarrhea, optimizing daily average growth, developing conception products, decreasing mortality, but also as an immunomodulator. The experiment was organized on homogeneous sow groups, which had the same microclimate conditions and the same fodder.

The experimental batch was fed differently to the control group by adding BioPlus 2B - CHR HANSEN product in feed in quantities of 1kg/ tonne feed. The action of the two

probiotic strains (*Bacillus subtilis* and *Bacillus licheniformis*) in the digestive tract is to select, develop and stabilize a beneficial microflora by acidifying, proliferating andrapid colonization.Blood samples were collected for haematological and biochemical determinations on the 85th day and 110th day of gestation. The obtained data were statistically processed.

The haematological profile of sows on *the 85th day of gestation* (T_1) shows subclinical anemia with Hb, Ht, and number of red blood cells decreased. The protein profile was characterized by normal values of proteins, albumins and globulins but at the lower limit of the reference values. The energy profile was characterized by normal triglycerides, cholesterol and slightly decreased cholesterol HDLP values. The values of uraemia and bilirubinemia are above the baseline. The mineral profile shows hypocalcemia, and the enzymatic profile shows increased transaminases.

At the T_{final}time (*110th day of gestation*) there was an increase in Hb, Ht, insignificant increases in albumin, a decrease in globulins, an insignificant decrease in blood glucose, triglycerides and cholesterol. There is a significant increase in HDLP cholesterol, which demonstrates the regulation of lipid metabolism and liver esterification capacity. Decreased urea and creatinine normalization tendency are observed, but bilirubin is maintained increased. Also the calcemia and enzyme profile values arenormalized.

The performance of pregnant sows: the number of piglets was 4% higher in the experimental group, the number of dead piglets by 75% higher in the control group, the average farrowing weight by 5.69% higher in the experimental group, the total weight of the weaned piglets by 15.60% higher in the experimental group, the average daily increase by 7.1% higher in the experimental group. Differences were also noted with respect to glycemia in pigs in the control and experimental groups. The haematological profile in piglets shows anemia in both groups at farrowing with a tendency to recovery at weaning.

2.The effect of supplementation of *lactating sows'* ratios with BioPlus 2B probiotic preparation

The experiment was conducted on lactating sows using the BioPlus 2B product and started on the first lactation day (T_0time) and ended on the 28th lactation day (T_2time).

During this period, both lactating sows' health condition (T_0 and T_2) and suckling piglets from the first day of life to weaning (28th day -T_2) were monitored. Double blood samples from lactating sows were collected on the first lactation day (T_0), 21th lactation day (T_1) and 28th lactation day (T_2). Suckling piglets were weighed at farrowing, double blood samples were collected for haematological and biochemical determinations. Dosing of blood

glucose was done on 1st, 5th, 8th, 15th, 21th, 24th, and 28th day. The weightings were made on weaning and double blood samples were collected for biochemical and haematological analyzes.

The haematological profile of the sows on *the first day of lactation* (T_0) in the experiment with the probiotic showed the presence of subclinical anemia with values of Hb, Ht, the number of red blood cells below the reference values, along with a slight leukopenia, and the protein profile of lactating sows to T_0time shows severe hypoalbuminemia, and a slight decrease in proteinemia relative to reference values. Uremia was increased, indicating elevated kidney stress and hepatic impairment in the last gestation period. Energy profile of lactating sows at the T_0 time, showed hypoglycemia, hypercholesterolemia and hypertriglyceridemia, and mineral profile was dominated by severe hypocalcemia. Enzymatic profile of lactating sows at the T_0time of the BioPlus 2B experiment shows increased values of GPT, GGT, and CPK and slightly increased to the upper limit PAL values compared to reference values.

At the T_2time (the 28thday of lactation), it was observed recovery from subclinical anemia, the protein profile, and uremia decreased significantly compared to the control group, sows' blood glucose in the experimental group increased compared to control group, and to the first day of lactation, decreased triglycerides and total cholesterol, along with the very significant increase in HDLP-cholesterol. The mineral profile of lactating sows demonstrates the recovery of the values of calcemia, phosphatemia and magnesemia, after 28 days of BioPlus 2B administration. Enhancement of the enzyme profile includes the majority of the enzymes monitored within the reference values.

Administration of probiotics in lactating sows for 28 days, affectsthe average weight at weaning, also increasing the total weight of weaned piglets' groups, while increasing average daily gain during farrowing-weaning, but does not affect the number of piglets weaned, correct hypoglycaemia of piglets, *albuminemia* at weaning was increased in the experimental group compared to the control group. *Subclinical anemia* present at farrowing in both groups was corrected at weaning in the experimental piglet group. The diarrheal score was dominated by the presence of diarrheal syndromes in the first 9 days in piglets in the control group (14), well above the experimental group (4).

3.The effect of supplementation of *pregnant sows'* ratios with Biomin IMBO synbiotic product

To demonstrate the effects of Biomin IMBO synbioticon pregnant sows between the 85th day of gestation and farrowing we used in the experiments symbiotic premix, feed

administered to pregnant sows. The objectives of the experiment with symbiotics on pregnant sows are the same as those with BioPlus 2B on pregnant sows.

Biomin IMBO is a (pro/ prebiotic) product based on the competitive exclusion principle, being an alternative replacing antibiotic, as growth promoters in sows. Biomin IMBO is a combination of synergistically-acting ingredients, between prebiotics (prebiotic "Inulina", of oligo-saccharides category) and probiotics supplemented with phicophytic substances from algae combined with extracts from cell walls of yeasts. The rate of inclusion in feed for pregnant and lactating sows for positive results in the development and feeding of piglets was 1 kg per tonne of feed.

At the T_0 time (85^{th} day of gestation), prior to administering the Biomin IMBO symbiotic to pregnant sows, there was evidence of subclinical, proteiprive anemia, possibly correlated with hypoproteinemia, present in the context of insufficient processing of proteins from the ration. Hyperglycemia of pregnant sows has been associated with hypercholesterolemia and hypertriglyceridemia with decreased HDLP-cholesterol values, uremia exceeded the baseline values on the 85^{th} day of gestation, showing disorders of energy metabolism and possible subclinical hepatopathy correlated with renal overstress. Hypocalcemia associated with slight hypomagnesaemia was present, and phosphorymia was within the reference range, but increased activity of transaminases GOT, GPT, GGT was observed, reflecting the presence of subclinical hepatopathies.

At the T_{final} time after 25 days of symbiotic administration, anemia was recovered, increased proteinemia and albuminemia with total globulin within the reference range. The hypoglycemia of the experimental group at T_{final} time was opposite to the control group hyperglycemia, as total blood cholesterol decreased below reference values and HDLP-cholesterol increased over control group values. After administration of symbiotics, there is a reduction in uremia due to decreases in creatinemia and bilirubinemia, resulting in higher hepatobiliary drainage and better kidney function, concomitantly with regulation of calcemia, phosphatemia and magnesemia, and regulation of enzyme activity for all monitored enzymes.

The synbiotics increased by more than 7.3%, the number of farrowed piglets from the experimentally treated sow group had an average weight by 13.63% higher than that of the control group and a total weight by 21.44% higher. Synbiotics given to pregnant sows indirectly influenced the blood glucose values of suckling piglets during farrowing, assuring energy needs through sows' milk and avoiding hypoglycemia present after farrowing in suckling piglets in the control group, and values for total protein, albumin and total globulins were within the reference range, compared to the piglet control group showing

hypoalbuminemia, hypoproteinemia,hypoglobulinemia and significantly decreased levels of albumin. The diarrheal score was dominated by a very high number of diarrheal episodes in the control group compared to a transient diarrhea episode on the 9th day in the experimental group of suckling piglets.

4.The effect of supplementation of *lactating sows'* ratios with the Biomin IMBO synbiotic preparation

The experiment was conducted on lactating sows using the Biomin IMBO product and started on the first lactation day (T_0 time) and ended on the 28th lactation day (T_2 time).

Double blood samples from lactating sows were collected on the first lactation day (T_0), 21th lactation day (T_1) and 28th lactation day (T_2). Suckling piglets were weighed at farrowing, double blood samples were collected for haematological and biochemical determinations. Dosing of piglet blood glucose was done on the 1st, 5th, 8th, 15th, 21th, 24th, and 28th day.

The average weights at farrowing and weaning were calculated, the average daily gain and the total weight of the piglet groups during farrowing and weaning. The number of smashed piglets in the first 3 days of life for both batches, of lactating sows, and the total number of dead piglets between the first day of life and weaning was quantified, and the diarrheal score was observed during the first 9 days of life.

At the T_0 time (first lactation day) of the Biomin IMBO experiment, sows showed subclinical anemia, hypoalbuminaemia and hyperglobulinaemia, uraemia and bilirubinemia increased beyond baseline values, mild hyperglycemia, hypertriglyceridemia, hypercholesterolemia, hypocalcemia, phosphorymia and magnesemia within normal range, increased activity of the GPT, GGT, and CPK above the reference limits.

At the T_1 time (the 21th day of lactation), it was observed recovery from subclinical anemia of the experimental group, proteinemia, albuminemia and globulinemia in the reference range, decreasing uraemia, creatinemia and bilirubinemia to the reference values, reduced glycemia below baseline values, total cholesterol, triglycerides and increased HDLP-cholesterol values with liver function enhancement and correction of baseline hypocalcemia, together with the enhancement of enzyme activity of all enzymes monitored.

At the end of the experiment (T_2) after 28 days of symbiotic administration, the hematologic profile and the protein profile were optimized, the values of uraemia, creatinemia and bilirubinemia were recovered, and the energy profile at the end of the experiment was characterized by hypoglycemia and significant increase in HDLP-cholesterol. At the T_2time, administration of synbiotics to lactating sows regulated the values of calcemia,

phosphatemia and magnesemia inphysiological limits, and has got the enzymatic activity of lactating sows balanced after 28 days of administration of the Biomin IMBO symbiotic. The productive performances of the lactating sows fed with the addition of synbiotics during the 28 days of feeding were evidenced by the daily average increase of piglets by 10% higher than the control group, with the total weight of weaned piglets by more than 8% higher, and mean weight at weaning by more than 7% higher.

The evaluation of blood glucose dosing showed the regulation of the glycemia curve in the piglets in the experimental group at weaning, the increase in their proteinemia and albuminemia. Hematological analyzes of suckling piglets showed a significant increase in hemoglobin, hematocrit and red blood cell number, recovery of subclinical anemia from farrowing and reduction in diarrheal scores (2 episodes), possibly by their superior immune reactivity.