# The effect of a feed additive supplemented with porcine plasma protein on growth performance and selected biochemical indices of nursing piglets

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#### Abstract

The aim of this study was to evaluate the health state, performance, and selected serum biochemical indices in nursing piglets after supplementation of a feed additive containing porcine plasma protein in comparison to the control group fed a standard pre-starter feed mixture. The study was carried out in the conditions of industrial farming. Four sows of the Large White swine breed with their litters were included in the study. Piglets were homogenously distributed into control (22) and experimental (24) groups according to weight and sex. Piglets in both groups suckled their mothers' milk. In addition, piglets in the experimental group were also fed a feed supplemented by porcine plasma protein (5%), starting on the third day post partum, whereas piglets in the control group were offered a standard pre-starter feed mixture. We recorded average daily weight gains calculated per the 27-day trial, and body weight at weaning. Clinical findings, mortality, and selected biochemical indices of protein (total immunoglobulins, total protein, albumin, creatinine, urea), and energy (glucose, total cholesterol, total lipids) metabolism were assessed as well. Results of the study showed a significant difference (P <(0.01) in average daily weight gains in the experimental group compared to the control group ((0.245)kg and 0.177 kg, respectively). Addition of plasma protein in the experimental group also resulted in a significantly higher serum concentration of total immunoglobulins, total protein and creatinine, and a significantly lower concentration of urea and albumin in this group compared to the control group at weaning.

### Protein source, immunity, digestibility, pigs

Efforts of pig breeders in intensive farming conditions are aimed at the most effective production of healthy and perspective animals. In terms of the analysis of critical control points (HACCP) in pig breeding, nutrition has been a key part of the chain. Production of viable and healthy piglets is a *sine qua non* for the subsequent successful performance of the herd. Optimal conditions must be ensured for optimal growth and ontogeny in the category of suckling piglets, and weaners as well. Side by side with standard feed mixtures, numerous other products have been used recently, such as porcine dried blood plasma, high quality pre-starters for piglets, organically bound microelements (iron, copper, manganese, zinc), porcine probiotics (Enterococcus faecium), plant extracts, toxin binders, enzyme products in combination with various vitamins (Figueroa et al. 2012) to reach the optimal performance. Swine diets must contain high levels of easily degradable nutrients due to high intensity of growth and reliance on sufficient enzymatic digestion; intensive growth is directly proportional to the intensity of proteosynthesis in the organism. Pigs are capable of using up to 35% of nitrogen in commercial feed mixtures (Bindas et al. 2013). Feed mixtures must contain balanced levels of proteins and other nutrients; at the same time, they must be used effectively so the producers can decrease the nutrient concentration in feed rations while maintaining their biological value for the animals. Although the order

Phone: +421 907 778 024 E-mail: katarina.bardova@uvlf.sk http://actavet.vfu.cz/ can vary with different dietary ingredient mixtures, typically the first five limiting amino acids for most practical diets are lysine, threonine, methionine, tryptophan and valine (Goodband et al. 2014). However, formulating diets with high levels of synthetic amino acids to the optimal ratio for the first five limiting amino acids often resulted in poorer performance than diets with higher levels of intact protein source (Tokach et al. 2013). On the other hand, current standard diets that often meet the nutritional needs of pigs in terms of the most limited amino acids exceed other amino acids requirements, resulting in excessive nitrogen excretions. Degradation of these excess proteins could result in the production of obnoxious odours (Sutton et al. 1998). Grains contain relatively low levels of proto-plasmatic proteins (albumins and globulins) that are valuable in pig nutrition for their solubility, high digestibility and optimal amino acid composition (Bindas and Maskal'ová 2012). Storage proteins, such as prolamins and glutelins make up the biggest part of the protein content in grains. However, they contain only low levels of some amino acids, and are relatively poorly digestible causing nutritional deficiencies, especially in piglets; supplementation of amino acids is hereby needed in this case to meet the nutritional requirements (Kar et al. 2016). Use of dried plasma as a food additive in piglet nutrition increases the average daily feed intake by enhancing the palatability of feed mixtures; it subsequently improves average daily weight gains, and health state of animals (Van Dijk et al. 2001). It contains a considerable amount of amino acids and immunoglobulins that have a positive effect on gut microflora as well (Heres 2016). One of the key roles of plasma proteins is to stimulate the feed intake, which results in greatly improved weight gains. Increased feed intake in young pigs is known to reduce the incidence and severity of diarrhoea (Owusu-Asiedu et al. 2003). The efficacy of porcine plasma in animal feed seems to be related to an improved barrier function of the gut mucosa and the modulation

Table 1. Composition of the pre-starter feed mixture (Complete compound feed for early weaning piglets OŠ 01, Tajba, a. s., Čaňa, Slovakia).

Ingredient	Unit	Amount	
NS	g·kg <sup>-1</sup>	200.0	
ME	MJ·kg <sup>-1</sup>	13.3	
Dietary fiber max.	g·kg <sup>-1</sup>	40.0	
Lysin	g·kg <sup>-1</sup>	14.0	
Methionine and cystine	g·kg <sup>-1</sup>	7.7	
Threonine	g·kg <sup>-1</sup>	9.1	
Calcium min.	g·kg <sup>-1</sup>	8.0	
Phosphorus	g·kg <sup>-1</sup>	6.7	
Sodium	g·kg <sup>-1</sup>	2.0	
Manganese	mg∙kg⁻¹	30.0	
Iron	mg∙kg⁻¹	125.0	
Copper	mg∙kg⁻¹	10.0	
Zinc	mg∙kg <sup>-1</sup>	100.0	
Retinol	IU·kg <sup>-1</sup>	8000.0	
Calciferol	IU·kg <sup>-1</sup>	1000.0	
Tocopherol	mg∙kg⁻¹	20.0	
Riboflavin	mg∙kg⁻¹	3.0	
Cobalamin	µg∙kg⁻¹	20.0	
Choline	mg∙kg <sup>-1</sup>	600.0	

NS - nitrogenous substances; ME - metabolizable energy

of the mucosal immune response (Pérez-Bosque et al. 2016).

The aim of the study was to evaluate the health, production and selected biochemical indices in nursing piglets after application of a feed additive containing porcine plasma protein in comparison to an unsupplemented control group.

#### Materials and Methods

In terms of production parameters, the swine farm involved in the study belongs to the best breeding facilities for sows in Slovakia, yet one of the smallest ones, with around 50 breeding sows kept at the facility. Purebred sows of the Large White breed, as well as Landrace  $\times$  Large White crossbreeds, and Pietrain  $\times$  Yorkshire crossbreeds were kept at the farm. The number of piglets reaches 27.4 per sow per year. Average daily weight gains in the fattening period reach 0.9 kg, with pre-weaning mortality of around 7%. Nutritional requirements are supplied by complete mixed feed rations based on the Schauer technology, and managed by the Schaumann Company.

The study was carried out during the summer of 2016. Two sows with their litters (n = 24) as the control group and two other sows with their litters (n = 22) as the experimental group were used for the study. Selected sows manifested no clinical signs of impaired health state, or any reproductive failures (abortions or stillborn

Ingredient	Unit	Amount
Crude protein	%	18.8
Fat and oil	%	9.2
Crude fibre	%	2.7
Ash	%	4.6
ME	MJ·kg <sup>-1</sup>	15.2
Calcium	%	0.6
Phosphorus	%	0.5
Sodium	%	0.2
Lysine	%	1.6
Methionine	%	0.6
Retinol	IU	$24 \times 10^3$
Cholecalciferol	IU	$2.0 \times 10^{3}$
Tocopherol	mg	200.0
Iron	mg	250.0
Copper	mg	160.0
Zinc	mg	120.0
Manganese	mg	64.0
Iodine	mg	2.0
Selenium	mg	0.5
Bonvital premix (Enterococcus faecium)	CFU	$1.0  imes 10^9$
Xylanase, glucanase, amylase, pectinase, benzoic acid	%	0.5
Feed acidifiers (formic acid, lactic acid, fumaric acid)	%	0.5
Feed preservants (butylhydroxytoluene, sorbic acid, calcium fumarate	e) mg	21.5
Propyl gallate	mg	7.5
Ethoxyquin	mg	3.3
Neohesperidin dihydrochalcone	mg	1.5
Sodium saccharin	mg	130.0

Table 2. Composition of the experimental diet (SCHAUMA IMMUNO, Schaumann, Germany).

ME - metabolizable energy

piglets). Piglets were marked with coloured ear tags for easier identification. All piglets were suckling their mothers' milk. Piglets in the experimental group were also offered the pre-starter feed mixture supplemented by plasma protein (5%) (SCHAUMA IMMUNO, Schaumann, Germany) starting on the third day *post partum*, whereas piglets in the control group were offered the standard pre-starter feed mixture (Table 1) without any supplementation. The composition of the experimental feed mixture is shown in Table 2. All procedures involving animals followed the guidelines stated in the Guide for the Care and Use of Animals (protocol number 3323/16-221/3) which was approved by the State Veterinary and Food Administration of the Slovak Republic and by the Ethics Committee of the University of Veterinary Medicine and Pharmacy in Košice, Slovakia. The animals were handled in a humane manner in accordance with the guidelines established by the relevant committee. All applicable international, national and institutional guidelines for the care and use of animals were followed.

Blood was drawn from sinus ophtalmicus for the analysis of selected biochemical indices in blood serum before the trial (after birth – day 0), and at weaning ( $28^{th}$  day of age) as well. Piglets were weighed prior to blood sampling. Commercial test tubes without an anticoagulant were used for blood storage until further processing; blood serum was obtained by centrifugation at  $867 \times g$  for 20 min. Serum concentrations of immunoglobulins (TIg) and total lipids (TL) were detected by quantitative, spectrophotometric zinc sulphate turbidity assay (Specol 211, Carl-Zeiss Jena, Germany). Serum concentrations of total protein (TP), albumin (ALB), creatinine (CR), urea (U), glucose (GLU), and total cholesterol (TCH) were determined using an automatic spectrophotometric analyser Alize (Lisabio, France).

Statistical analysis of the results was carried out by assessment of mean values and standard deviations; the level of significance of detected differences between the experimental and control group was assessed using *t*-test, with *post hoc* analysis by Tukey's test in MS Excel 2016.

The average body weight (ABW; Table 3) at birth showed no significant difference between the control and experimental groups (1.860 kg, and 1.862 kg, respectively), whereas at the weaning ABW in the control group (7.147 kg) showed a significant difference (P < 0.01) compared to the experimental group (8.486 kg). Differences in average daily weight gains (ADG), calculated per the 27-day trial in the experimental and control group (0.245 kg, and 0.177 kg, respectively) were significant (P < 0.01) as well.

Concentrations of selected biochemical indices in blood serum of piglets are shown in Table 3. On day 0 they correspond to those of newborn piglets with no significant differences observed between the groups. Regarding the concentration of selected blood biochemistry indices, we found a significantly (P < 0.01) higher mean serum concentration of TP (74.10 g·l<sup>-1</sup>), TIg (25.39 UZST; units of zinc sulphate turbidity), and no significant value of CR (101.54 µmol·l<sup>-1</sup>) at weaning in the experimental group when compared to the control group (56.05 g·l<sup>-1</sup>; 16.06 UZST; 89.94 µmol·l<sup>-1</sup>). On the other hand, the opposite tendency with a significance of P < 0.01 was observed in the mean serum concentration of U (3.09 mmol·l<sup>-1</sup> vs 5.31 mmol·l<sup>-1</sup>) and ALB (31.67 g·l<sup>-1</sup> vs 34.14 g·l<sup>-1</sup>). There were no significant differences between groups in the concentrations of glucose, TL and TCH.

experimental group in piglets at birth and after weaning.								
Indicator [unit]	Reference range	Experimental group		Control group				
		Day 0	Day 28	Day 0	Day 28			
		n = 22	n = 22	n = 24	n = 19			
ABW [kg]		$1.862\pm0.274$	$8.486^{*}\pm 0.513$	$1.860\pm0.488$	$7.147\pm0.971$			
ADG [kg]		0.245*		0.177				
TIg [UZST]	20-35	$18.24\pm2.35$	$25.39* \pm 3.11$	$15.32\pm10.38$	$16.06\pm2.76$			
TP [g·1 <sup>-1</sup> ]	70–90	$50.00\pm3.19$	$74.10^{\boldsymbol{*}}\pm 8.38$	$51.39\pm21.29$	$56.05\pm8.63$			
Albumin [g·l-1]	35–45	$18.31\pm2.25$	$31.67\pm3.41$	$16.26\pm5.27$	$34.14^{\boldsymbol{*}} \pm 5.87$			

 $101.54 \pm 11.13$ 

 $3.09* \pm 0.81$ 

 $5.37 \pm 0.3$ 

 $3.59 \pm 0.5$ 

 $\phantom{0.0}6.9\pm 0.68\phantom{.0}$ 

 $105.06* \pm 56.64$ 

 $4.64 \pm 2.36$ 

 $7.31 \pm 1.37$ 

 $5.03\pm0.68$ 

 $2.44 \pm 0.64$ 

 $89.94 \pm 12.04$ 

 $5.31 \pm 1.01$ 

 $7.18 \pm 2.3$ 

 $6.67\pm0.98$ 

 $4.29 \pm 0.66$ 

 $57.85 \pm 3.24$ 

 $4.42 \pm 1.55$ 

 $6.68 \pm 0.87$ 

 $5.56\pm0.59$ 

 $1.96 \pm 0.38$ 

Table 3. Comparison of selected parameters of performance and serum biochemistry in the control and experimental group in piglets at birth and after weaning.

Results are presented as mean  $\pm$  standard deviation, \* = P < 0.01

141-239

2.5 - 10.7

4.7-8.35

3.5-5.5

2.6 - 3.9

Creatinine [µmol·l-1]

Glucose [mmol·l<sup>-1</sup>] TL [g·l<sup>-1</sup>]

Urea [mmol·l-1]

TCH [mmol·l<sup>-1</sup>]

n – number of animals; ABW - average body weight; ADG - average daily gain; TIg - total immunoglobulins; TP -total protein; TL - total lipids; TCH - total cholesterol; UZST - units of zinc-sulphate test

# Discussion

Numerous authors (Heres 2016; Grinstead et al. 2000; Lawrence et al. 2004) reported that feeding porcine blood plasma as a source of protein increases the feed intake, weight gains, and health performance of piglets when added to their diets in the pre-, and/or post-weaning period. On the other hand, several studies doubt that (Carlson and Veum 2000; Rigueira et al. 2013). The lack of consistent results among different papers may be related to factors such as environmental stress, immune challenge levels, and the complexity of the diets used in these studies (Pereira et al. 2012). The higher weight gain of animals supplemented with blood plasma may be affected by factors such as the age at weaning. In situations of normal challenge, diets supplemented with spray-dried plasma should be used in piglets not older than 35 days, because when used up to 35 days

of age, the supplementation positively influences the average daily weight gain and the feed conversion ratio (Remus et al. 2013). The positive effects of functional feeds such as spray-dried blood plasma on pig performance are more dramatic when the animals are exposed to a higher level of immune challenge (Coffey and Cromwell 2001). The growth-promoting action of plasma protein added to feed mixtures could lie in the observed increase in feed intake due to increased palatability. Plasma proteins could have a direct effect on the intestines, leading to less intestinal diseases and, in turn, higher feed intake and daily gains as well (Perez-Bosque et al. 2016). The immunoglobulin and glycoprotein fractions may prevent attachment of pathogens and thus support functionality of the intestine (Van Dijk et al. 2001). This effect was also observed in different species. The glycoprotein glycans inhibited adhesion of bacteria to the intestine and protected colostrum-deprived calves against lethal doses of enterotoxigenic E. coli. There is some evidence that the feeding of large amounts of spray-dried plasma to weaned piglets offers protection against pathogenic E. coli (Nollet et al. 1999). În our study, we did not observe any health disorders in piglets of the control or experimental group that could have been related to this. Our results regarding performance (average daily weight gains and weight at weaning) were in accordance with a study in piglets fed the starter diet with spray-dried plasma protein (Pujols et al. 2016). Their body weight, average daily weight gain, and average daily feed intake improved during the initial 14 days after weaning, along with feed efficiency which improved during the initial 7 days after weaning. These responses were independent of the applied vaccination. In another study (Zhao et al. 2007), plasma protein and copper supplementation improved pig growth performance during the initial 10 days after weaning. The authors noticed a reduced crypt depth in the duodenum in piglets with only copper supplementation and a tendency for greater villous length with plasma supplementation. These modifications to non-medicated diets acted independently with regard to their impacts on the post-weaning performance and, therefore, could have additive effects. In contrast to these findings, in a study evaluating the effect of diets supplemented with plasma and/or yeast protein on the performance of piglets, no significant differences in intestinal morphology between piglets receiving the different diets were observed. The combination of plasma and yeast extract elevated daily weight gains, but did not affect the intestinal morphology of piglets from 7 to 63 days of age (pre- and post-weaning) (Rigueira et al. 2013). The reason for inconsistencies in the feed intake and subsequent weight gains independently from plasma supplementation might lie in different feeding rations and/or feeding schedule. It is probable that the change in consumption is dependent on the type and origin of ingredients. In this context, it is possible that the diet composition influenced the consumption due to the transit time of the diet in the intestinal tract (Black et al. 2009). Recent studies have mostly dealt with supplementation of porcine plasma to weaners (Grinstead et al. 2000; Torrallardona 2010). Less is known about the effect of plasma protein on nursing piglets, since the preventive measures are aimed at the most challenging periods, such as weaning time. Due to the post-weaning stress and changes in the diet, piglets are becoming more susceptible to pathogens as well as dietary imbalances.

As mentioned above, the analysis of biochemical indices in blood serum of piglets (Table 3) points to the positive effect of feeding dried porcine plasma to piglets during the nursing period on their immune function and nutrient absorption. Plasma urea nitrogen is used as an indicator of protein utilization efficiency because it increases when there is an imbalance of dietary amino acids (Coma et al. 1998).

Hence, the lower urea concentration in the experimental group suggests a positive effect of the plasma protein on the amino acid composition in the piglet feed ration compared to the control group. The significantly higher serum level of TP and TIg in the experimental group may be viewed as the result of improved gut function, as well as increased feed consumption/nutrient intake after supplementation of porcine dried plasma. This was in accordance with numerous studies assessing the effect of different feed additives on the production indices, immunity and blood biochemistry in weaned piglets (Li et al. 2006; Zigger 2011; Xiong et al. 2015; Shenga and Nyachoti 2017). Furthermore, nutrient supplementation in nursing piglets shortly after birth has played an important role in the postnatal development of the immune system (Cunningham-Rundles et al. 2009).

In conclusion, we can assume that the addition of porcine plasma into pre-starter diets for nursing piglets had a positive effect on the production parameters such as average weight gains and weight at weaning by an improved and balanced nutrient intake, and also enhanced the immune functions represented by the serum level of TIg. The results of our study show that the porcine plasma protein can be a part of the strategy to support the health of piglets in critical phases of life, such as the nursing and weaning periods, thereby reducing the need to use of drugs.

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