



Multi-enzymatic complex on growth performance, blood parameters, and economic viability in piglets

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ABSTRACT. The aim of the present study was to evaluate the dietary effect of an enzymatic complex on the growth performance variables, blood parameters, and economic viability of piglets. To achieve this, we used 80 piglets (40 castrated males and 40 females) in a 2 × 2 factorial design following a randomized block experimental distribution, with two levels of metabolizable energy (adequate: 3206.09 and low: 3005.45 kcal kg⁻¹) and two enzyme levels (0 and 50 g ton⁻¹). The results showed no significant difference between treatments in the growth performance variables or blood parameters of piglets. The economic viability, economic efficiency index, and cost index were improved when we used a diet with a low energy supplemented with the enzyme complex as compared to other experimental diets. Thus, we concluded that enzyme complex supplementation can maintain growth performance and blood parameters in piglets even when they are fed low energy diets. Moreover, this could reduce production costs.

Keywords: xylanase; non-ruminant nutrition; swine production.

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Introduction

The Brazilian swine farming is among the most developed in the world with excellent productivity levels and health status. Generally, the costs of feed account for 70% of total production costs in swine farming. However with the increase costs of soybean meal and corn it is estimated that the costs has currently exceeded 80%. Thus, fluctuations in the prices of these products greatly impact the total price of the diet (Bavaresco et al., 2020). Therefore, an option to reduce these costs is the use of alternative foods and/or feed enzymes.

Pigs are commonly fed high-fiber diets to reduce production costs and non-starch polysaccharide (NSP)-degrading enzymes have been used to increase fiber digestibility (Vila et al., 2018). In addition, feed enzymes can be used to increase the nutrients digestibility of corn and soybean meal diets because the soybean contain in their composition nonstarch polysaccharides (NSP) and oligosaccharide (Bueno et al., 2018). These soluble components are one of the main factors responsible for the antinutritional effects of soybeans (Choct, Dersjant, Mcleish, & Peisker, 2010).

Monogastric animals do not have the enzymes to hydrolyze the nonstarch polysaccharides (NSP), and thus, their digestion occurs by means of bacterial fermentation (Bueno et al., 2018). Thus, the use of feed enzymes can be a nutrition strategy for poultry and swine. Their potential effects include reduction of anti-nutritional factor effects and phytic acid as well as improved digestibility of nutrients (Adeola & Cowieson, 2011), through the reduction in digesta viscosity (Duarte, Zhouc, Dutra, Kim, 2019), and reduction of production costs.

The aim of the present study was to evaluate the dietary effect of an enzymatic complex on the growth performance variables, blood parameters, and economic viability of piglets.

Material and methods

Animals and housing

The experiment was conducted at the Research Unit of Swine Production at the Federal University of Technology - Parana, Dois Vizinhos, Parana, Brazil. All procedures were approved by The Ethics Commission on Animal Use (CEUA) of Federal University of Technology – Paraná (UTFPR) (protocol number 2015-027).

In total, 80 commercial hybrid piglets (40 males, 40 females) and an initial live weight of 16.88 kg (SD 2.69 kg) at 42 days of age were used in this study. The pigs were blocked based on live weight, and within each block, they were assigned to one of four dietary treatments (n = 20). The pigs were grouped in mixed genders (1:1) in each pen. The experimental design was randomized blocks, in a 2x2 factorial scheme, two energy level diets (low and adequate) and two levels of enzyme added to the diet (0 and 50 g t⁻¹).

Diets and feeding

The treatments were as follows: Diet with low energy (3005.45 kcal kg⁻¹) + 50 g ton⁻¹ of Multi-Enzymatic Complex (MEC); Diet with low energy without MEC supplementation; Diet with adequate energy level (3206.09 kcal kg⁻¹) + 50 g ton⁻¹ of MEC and Diet with adequate energy level (3206.09 kcal kg⁻¹) without MEC supplementation. The MEC used for this study had the following composition: xylanase (20,000 u g⁻¹), amylase (120,000 u g⁻¹), beta-glucanase (7,500 u g⁻¹) and mannanase (250 u g⁻¹); the dose recommended by the manufacturer of the product was 50 g ton⁻¹.

Experimental diets (Table 1) were formulated based on the piglets' with high genetic potential (Rostagno, Albino, & Donzele, 2011).

Table 1. Experimental diet compositions.

Ingredients (kg)	Low energy diet + MEC	Low energy diet without MEC	Diet with adequate energy level + MEC	Diet with adequate energy level without MEC
Corn	552.00	552.00	552.00	552.00
Soybean meal 45% PB	346.30	346.30	346.30	346.30
Soybean oil	-	-	22.80	22.80
Kaolin	31.70	31.70	8.90	8.90
Vitamin and mineral premix ¹ with MEC ²	70.00	-	70.00	-
Vitamin and mineral premix	-	70.00	-	70.00
TOTAL	1000.00	1000.00	1000.00	1000.00
Calculated Nutritional Levels				
Crude protein (%)	20.45	20.45	20.45	20.45
Ethereal extract (%)	2.60	2.60	4.86	4.86
Crude fiber (%)	2.79	2.79	2.79	2.79
Metabolizable energy (Kcal kg ⁻¹)	3005.45	3005.45	3206.09	3206.09
Digestible Lysine (%)	1.10	1.10	1.10	1.10
Digestible methionine+cysteine (%)	0.56	0.56	0.56	0.56
Digestible threonine (%)	0.69	0.69	0.69	0.69
Digestible tryptophan (%)	0.22	0.22	0.22	0.22
Digestible arginine (%)	1.30	1.30	1.30	1.30
Calcium (%)	0.72	0.72	0.72	0.72
Total phosphorus (%)	0.58	0.58	0.58	0.58
Disponibile phosphorus (%)	0.35	0.35	0.35	0.35
Colistin	40.00	40.00	40.00	40.00
Multi-enzyme complex (g ton ⁻¹)	50.00	-	50.00	-

¹Vitamin and mineral commercial premix intended for piglets. ²Guaranteed levels: Xylanase (20,000 u g⁻¹), amylase (120,000 u g⁻¹), beta-glucanase (7,500 u g⁻¹) and mannanase (250 u g⁻¹).

Growth performance, blood parameters, and economic viability measurements

The body weight of each animal was measured at the beginning of the experiment (initial live weight were 16,88 + 2,69 kg⁻¹) and was monitored weekly until the end (42 days of age) of the experiment, whereas the average feed intake and feed conversion were monitored weekly. Feed conversion rate was calculated by dividing feed intake by weight gain.

In order to evaluate the economic viability variable, we utilized the prices of raw materials to obtain the cost of each experimental diet in the region of Dois Vizinhos, Paraná State. The premix costs were the values stated by the manufacturer and already included the value of the MEC.

Calculated the cost of feed per kilogram of live weight gain according to (Bellaver, Fialho, Protas, & Gomes, 1985) as follows: $Y_i (R\$ \text{kg}^{-1}) = Q_i \times P_i / G_i$, where: Y_i = feed cost per kg of live weight gained at day i of treatment; Q_i = amount of feed consumed at day i of treatment; P_i = price per kg of the ration used at day i of treatment; and G_i = weight gain at day i of treatment. also calculated the Economic Efficiency Index (EEI) and the Cost Index (CI) (Carvalho et al., 2009), where $EEI (\%) = M_{Ce} / C_{Tei} \times 100$ and $CI (\%) = C_{Tei} / M_{Ce} \times 100$, where M_{Ce} = lowest feed cost per kg gain observed between treatments and C_{Tei} = the considered cost for treatment i.

For the evaluation of blood parameters, the animals were subjected to fasting for 4h, and a ration was then offered for 1h, followed by another fasting period of 1h duration. At the end of this period, 10 mL of blood

was collected by puncturing the anterior vena cava of one animal per experimental unit (Personal communication with Dr. Ivan Moreira and Dr. Paulo Pozza).

The blood was distributed in properly identified evacuated tubes: 5 mL⁻¹ in tubes with EDTA anticoagulant for determination of red blood cell counts (mm⁵ × 10⁵), hemoglobin (%), hematocrit (%), leukocytes (mm⁵), and differential leukocyte count, the percentages of eosinophils, rod neutrophils, segmented neutrophils, lymphocytes, monocytes, and platelet counts; the remaining 5 mL⁻¹, without anticoagulant, was used to determine the levels of immunoglobulins and in biochemical tests (total protein, albumin, urea, cholesterol, triglycerides, phosphorus, calcium, and magnesium) (Schalm & Jain, 1986).

The red blood cell counts, hemoglobin, hematocrit, and platelet counts were determined using an automated hematological analyzer. Alternatively, for differential counting of leukocytes, we performed a blood smear, which was observed using an optical microscope.

Blood samples without anticoagulant were centrifuged at 2,500 rpm for 10 min. to obtain serum, and the aliquots were packed in plastic microtubes, identified, and kept at -20°C until the biochemical tests were performed.

The methods used for these tests were as follows: total proteins were determined by the biuret method, urea by the urease method, albumin by the bromocresol green method, enzyme aspartate aminotransferase by the enzymatic method, total cholesterol by the cholesterol esterase oxidase method, triglycerides by the glycerol-3-phosphate oxidase method, phosphorus by the ammonium molybdate method, calcium by the phthalein purple method, and magnesium by the Magon sulfonate method. All analyses were performed on a Bioplus 200[®] semiautomatic device, using Labtest Diagnostica S.A. (Brazil) commercial kits.

Calculations and statistical analyses

The experimental design was a complete randomized block, with a 2 × 2 factorial design containing two metabolizable energy levels (adequate: 3206.09 and low: 3005.45 kcal kg⁻¹) and two levels of MEC (0 and 50 g t⁻¹), for a total of four treatments with ten replicates per treatment and two piglets per experimental unit.

We performed statistical analysis using Statistix[®] software (2009). Growth performance and blood parameters were subjected to analysis of variance. The effects of the treatments were also compared using factorial analysis, and when statistical significance was observed, Tukey's test was applied at the 5% probability level.

Results and discussion

Based on the results obtained, there was no significant interaction ($p > 0.05$) between dietary energy and MEC level for the performance variables studied. Also did not observe any effect ($p > 0.05$) of energy level or MEC on the animals' performance (Table 2). These results indicate that the diet provided to the animals did not affect their growth performance.

Table 2. Growth performance in piglets supplemented with multi-enzymatic complex (MEC).

Variables Studied	FI ¹ (kg ⁻¹)	BWG ² (kg ⁻¹)	ADFI ³ (kg ⁻¹)	ADBWG ⁴ (kg ⁻¹)	FCR ⁵
Energy and MEC Effect					
Low energy diet + MEC	26.821	12.810	1.276	0.609	2.108
Diet with adequate energy level + MEC	27.814	13.495	1.324	0.642	2.079
Low energy diet without MEC	28.795	12.527	1.370	0.596	2.396
Diet with adequate energy level without MEC	28.661	13.168	1.364	0.626	2.186
<i>P</i> -Value	ns	Ns	Ns	Ns	ns
SE**	1.862	0.956	0.088	0.045	0.165
Energy Effect					
Low energy diet	27.808	12.669	1.323	0.602	2.252
Diet with adequate energy level	28.238	13.331	1.344	0.634	2.128
<i>P</i> -Value	ns [†]	Ns	Ns	Ns	ns
MEC Effect					
Diet with MEC	27.318	13.153	1.300	0.625	2.089
Diet without MEC	28.728	12.848	1.367	0.611	2.291
<i>P</i> -value	ns	ns	Ns	ns	ns
SE	1.316	0.676	0.062	0.032	0.117
***CV (%)	14.860	16.460	14.870	16.480	16.900

[†]ns = Not significant; **SE = Standard Error for comparison; ***CV(%) = Coefficient of Variation; ¹FI = Feed Intake; ²BWG = Body Weight Gain; ³ADFI = Average Daily Feed Intake; ⁴ADBWG = Daily Body Weight Gain; ⁵FCR = Feed Conversion Rate.

The results observed in this study are in agreement with those of Kim, Zhang, Soltwedel, and Knabe (2006), who observed that supplementation of an enzymatic complex based on α -galactosidase, β -1,4-mannanase, and β -1,4-mannanase in piglet diets (corn-soybean meal based diets) with different levels of metabolizable energy (3969; 4187; 4182; and 4174 Kcal kg⁻¹) did not yield any improvement ($p > 0.05$) in digestibility, Feed Conversion (FC), Body Weight Gain (BWG), or Feed Conversion Rate (FCR). These experiments indicate that supplementation of an enzymatic complex improves nutrient digestibility and could reduce at least 3% of ME in swine diets without adverse effects on growth performance.

Xuan et al. (2001) and Nery, Lima, Melo, and Fialho (2000) tested a complex of the enzymes α -amylase, β -amylase, xylanase, β -glucanase, protease, cellulase, and pectinase and observed did not promote a significant difference in the daily feed intake or body weight gain of piglets.

Teixeira et al. (2005) investigated various levels (0, 0.2, 0.4, and 0.6%) of an enzyme complex containing amylase, protease, and cellulase in the piglet diet. They observed that increased inclusion of enzymes caused a linear increase (12.19, 12.36, 12.80, and 13.43 kg⁻¹) in weight gain and in feed intake (491, 489, 514, and 591 g day⁻¹), although the results showed no significant difference among the variables.

In another study, Rodrigues, Freitas, Fialho, Silva, and Gonçalves (2002) tested the supplementation of an enzyme complex (xylanase, amylase, β -glucanase, and pectinase) at 1 kg ton⁻¹ in two treatments with the substitution of corn (3305 kcal.kg⁻¹ of digestible energy) for sorghum (3295 kcal kg⁻¹ of digestible energy). They observed a significant difference ($p < 0.05$) between the diets, and found that diets containing the enzymatic complex increased weight gain by 3.51% and feed conversion by 6.45%.

Kim et al. (2013) evaluated diets with different levels of energy (3272 and 3343 kcal kg⁻¹) and enzyme supplementation (400 u kg⁻¹ β -glucanase), and observed that the diet including the enzyme increased weight gain (81.2 and 82.4 kg versus 79.7 and 81.5 kg⁻¹) as compared to the diet containing the lowest energy level without inclusion of the enzyme.

The results observed by Ruiz et al. (2008) differed from the results of this study and the study conducted by the aforementioned authors. They observed that an energy-deficient diet (3234 kcal kg⁻¹) supplemented with an enzymatic complex presented lower weight gain values, ration intake, and weight gain as compared to a diet with adequate energy levels (3300 kcal kg⁻¹). This indicated that the enzymes did not improve digestibility and that the energy-deficient ration negatively affected the performance of the piglets.

Biochemical parameters in the piglets' blood as observed in Tables 3, 4 and 5, there was no significant relationship ($p > 0.05$) between the energy and MEC levels or between the isolated factors (energy and MEC) and the blood parameter variables analyzed.

Table 3. Biochemical parameters in the piglets' blood.

Variables studied	URE ¹	Ca ²	Mg ³	P ⁴	PROT ⁵	TRG ⁶	COL ⁷	ALB ⁸	GLO ⁹
Reference levels (mg dL ⁻¹)	21.4-64.2	7.10-11.60	2.70-3.70	5.30-9.60	7.90-8.90 (g dL ⁻¹)	-	36.0-54.0	1.90-3.90 (g dL ⁻¹)	-
Energy and MEC Effect									
Low energy diet + MEC	40.87	10.93	2.35	10.21	5.87	36.000	77.5	2.92	2.949
Diet with adequate energy level + MEC	40.40	10.76	2.28	9.87	5.66	41.100	81.6	2.83	2.777
Low energy diet without MEC	45.27	11.08	2.27	9.52	5.64	34.800	79.6	2.92	2.713
Diet with adequate energy level without MEC	40.41	11.09	2.28	10.24	5.94	51.100	76.2	2.95	2.989
P-Value	ns*	Ns	Ns	Ns	ns	ns	ns	ns	ns
SE**	2.65	0.23	0.10	0.53	0.22	7.885	5.1	0.194	0.202
Energy Effect									
Low energy diet	43.07	11.00	2.31	9.86	5.75	35.400	78.5	2.92	2.831
Diet with adequate energy level	40.40	10.92	2.28	10.05	5.80	46.100	78.9	2.91	2.883
P-Value	Ns	Ns	Ns	Ns	ns	ns	ns	ns	ns
MEC Effect									
Diet with MEC	40.63	10.84	2.31	10.04	5.76	38.550	79.5	2.90	2.863
Diet without MEC	42.84	11.08	2.27	9.88	5.79	42.950	77.9	2.93	2.851
P-value	Ns	Ns	Ns	Ns	ns	ns	ns	ns	ns
SE	1.87	0.16	0.07	0.38	0.15	5.766	3.6	0.13	0.143
***CV (%)	14.24	4.87	9.95	12.08	8.58	43.270	14.6	14.88	15.860

¹ = Reference values for biochemical parameters of piglets; * ns = not significant; **SE = Standard Error for comparison; *** CV (%) = Coefficient of Variation; ¹Urea (URE); ²Calcium (Ca); ³Magnesium (MG); ⁴ Phosphorus (P); ⁵Total Protein (PROT); ⁶ Triglycerides (TRG); ⁷ Cholesterol (COL); ⁸ Albumin (ALB), and ⁹ Globulin (GLO)

These results indicate that the diet given to the animals did not alter the biochemical parameters of the blood, which are extremely important to animal health. Thus, can understand the effects of enzyme supplementation on the animals through individual interpretation of each variable.

In the present study, the urea levels were within the expected range of 21.4 to 64.2 mg dL⁻¹. This information is important because it allows to determine the quality of the protein ingested. Furthermore, this analysis indicates the extent to which amino acids are being harnessed for protein synthesis. Therefore, when these levels are high (< 64.2 g dL⁻¹), it indicates that the use of dietary protein is inefficient or that there is an excess of protein in the diet (Lazzeri et al., 2011).

Also observed that the values of Ca, Mg, P, total proteins, and albumin were within the ideal range for piglets at this stage (Kaneko, Harvey, & Bruss, 2008). However, when we evaluated the factors alone, we observed that the enzyme caused an increase in P and Mg levels; this might have been caused by factors in the manufacturer's nutritional matrix (confidential), which was not available for study.

The total proteins and albumin indicate metabolic behavior; from these examinations, can tell if a failure occurs in the ingestion, digestion, absorption, and metabolization of proteins (Lopes, Biondo, & Santos, 2007).

Similarly, there was no significant effect of energy and MEC or isolated factors (energy and MEC) on globulin. The ideal level for piglets was not reported in the literature, but our results are thought to be within normal values, considering that they were very similar to those of dogs (2.7-4.4 g dL⁻¹) (Lopes, Biondo, & Santos, 2007). Globulins are the main factors responsible for transporting lipids, hemoglobin, proteases, and iron. They are also precursors of the immune complement system factors, fibrin, and humoral immunity (Lopes, Biondo, & Santos, 2007). Therefore, any changes in these variables result in alteration of the immune system of the animals, directly affecting their health.

Likewise, cholesterol and triglyceride tests did not present significant differences between treatments. Both are influenced by the energy and fat content of the diet (Pascoal et al., 2008). However, the blood cholesterol levels of the piglets were above the reference values at > 36-54 mg dL⁻¹.

Similarly, there was no significant effect of energy and MEC or isolated factors (energy and MEC) on erythrogram variables (Table 4). The erythrogram examination allowed to determine the red blood cell levels. thus obtained the red blood cell count, hematocrit, and hemoglobin, and the concentration and volume of each (hypochromic or normochromic). Altogether, could determine whether or not the animals were anemic (Lopes, Biondo, & Santos, 2007).

Overall, the health status of the animals was considered appropriate and within the standards, without presenting anemia. Thus, the diets met the animals' nutritional requirements without causing damage to their health, even when the Low Energy Diet without MEC (diet that did not meet the energy requirements + enzymatic complex) was employed for cost reduction.

Table 4. Piglet erythrogram evaluation.

Variables studied	EC (g dL ⁻¹) ¹	HMT (g.dL ⁻¹) ²	HG (g dL ⁻¹) ³	MCV (fl) ⁴	MCH (pg) ⁵	MCHC (%) ⁶	RDW (%) ⁷
Reference levels	-	-	10.0–16.0	50.0–68.0	17.0–21.0	-	-
Energy and MEC Effect							
Low energy diet + MEC	6.670	38.400	12.0	57.8	18.6	32.586	22.900
Diet with adequate energy level + MEC	7.138	39.970	12.0	56.0	16.9	30.181	24.450
Low energy diet without MEC	7.012	40.770	12.2	58.1	17.5	30.136	24.160
Diet with adequate energy level without MEC	7.086	40.520	12.2	57.2	17.3	30.339	22.180
<i>P</i> -Value	Ns	Ns	Ns	ns	ns	ns	ns
SE**	0.311	2.072	0.4	1.6	0.9	1.883	1.422
Energy Effect							
Low energy diet	6.814	39.585	12.1	57.9	18.1	31.361	23.530
Diet with adequate energy level	7.112	40.245	12.1	56.6	17.1	30.260	23.315
<i>P</i> -Value	ns*	Ns	Ns	ns	ns	ns	ns
MEC Effect							
Diet with MEC	6.877	39.185	12.0	56.9	17.8	31.384	23.675
Diet without MEC	7.049	40.645	12.2	57.7	17.4	30.237	23.170
<i>P</i> -value	Ns	Ns	Ns	ns	ns	ns	ns
SE	0.220	1.465	0.3	1.1	0.6	1.331	1.005
***CV (%)	10.010	11.610	7.9	6.5	12.1	13.670	13.580

¹= Reference values for erythrogram parameters of piglets; * ns = Not significant; **SE = Standard Error for comparison; ***CV (%) = Coefficient of Variation; ¹Erythrocytes (EC); ² Mean corpuscular volume (MCV); ³ Mean corpuscular hemoglobin (MCH); ⁴Mean corpuscular hemoglobin concentration (MCHC); ⁵Hemoglobin (HG); ⁶Hematocrit (HMT); ⁷ Red Cell Distribution Width (RDW)

As observed in Table 5, no significant effect ($p > 0.05$) was found for energy and MEC or for the isolated factors (energy and MEC) (control and enzyme levels) on the leukogram variables evaluated. Except for the eosinophil and rod variables, for which the interaction presented a significant effect ($p < 0.05$). In the leukogram results, observed that the values found in this experiment were lower than the reference values. Moreover, there was a reduction in lymphocyte and monocyte counts, which might indicate the occurrence of immune system commitment.

Table 5. Piglet leukogram evaluation.

Variables Studied	BAS ² (μ L)	BST ³ (μ L)	EO ⁴ (μ L)	LEU ⁵ (μ L)	LIN ⁶ (μ L)	MNO ⁷ (μ L)	NEU ⁸ (μ L)	PLQ ⁹ (μ L)	SG ¹⁰ (μ L)
Reference	0.0- 2.0	0.0- 4.0	1.0- 11.0	11.00- 22.00	39.0- 62.0	2.0- 10.0	28.000- 47.000	100.000- 900.000	-
Energy and MEC Effect									
Low energy diet + MEC	0.0	0.1 ^b	1.6 ^a	6.91	57.1	0.3	41.000	299.700	40.900
Diet with adequate energy level + MEC	0.0	1.5 ^a	0.3 ^b	7.53	45.5	0.5	53.700	239.500	52.200
Low energy diet without MEC	0.0	0.9 ^{ab}	0.6 ^{ab}	6.90	48.8	0.6	50.000	282.600	49.100
Diet with adequate energy level without MEC	0.0	0.7 ^{ab}	0.5 ^b	9.41	53.5	0.2	45.800	280.800	45.100
P-Value	Ns	0.0	0.0	Ns	Ns	Ns	ns	ns	ns
SE**	0.0	0.4	0.4	1.11	6.3	0.3	6.490	35.741	6.313
Energy Effect									
Low energy diet	0.0	0.5	1.1 ^a	6.91	52.9	0.4	45.500	291.150	45.000
Diet with adequate energy level	0.0	1.1	0.4 ^b	8.47	49.5	0.3	49.750	260.150	48.650
P-Value	Ns	ns	0.0	Ns	Ns	ns	ns	ns	ns
MEC Effect									
Diet with MEC	0.0	0.8	0.9	7.22	51.3	0.4	47.350	269.600	46.550
Diet without MEC	0.0	0.8	0.5	8.15	51.1	0.4	47.900	281.700	47.100
P-value	Ns	ns	ns	Ns	Ns	s	ns	ns	ns
SE	0.0	0.30	0.2	0.78	4.4	0.2	4.5898	25.273	4.4641
***CV (%)	0.0	118.5	120.4	32.30	27.5	179.7	30.48	28.99	30.15

¹ = Reference values for leukogram parameters of piglets; * ns = Not significant; **SE = Standard Error for comparison; ***CV (%) = Coefficient of Variation; a-b are different in the same column; ² Basophils (BAS); ³Rods (BST); ⁴ Eosinophil (EO); ⁵ Leukocytes (LEU); ⁶Lymphocytes (LIN); ⁷Monocytes (MNO); ⁸Platelets (PLQ); ⁹Segmented neutrophils (SG).

Regarding the eosinophil and rod variables, observed that the interaction of the factors was significant ($p < 0.05$). The diet with adequate energy levels supplemented with MEC yielded a higher rod count compared to that of the other treatments, whereas the energy-deficient diet + MEC generated the lowest rod count compared to the other treatments. However, all levels were within the reference value (0-4 μ L %⁻¹) stipulated by (Kaneko et al., 2008). In contrast, the eosinophil count was lower in the adequate energy level diet supplemented with MEC and the adequate energy level diet without MEC, and presented higher levels in the energy-deficient + MEC diet as compared to the other treatments. However, the treatments presented eosinophil counts within the range of 1–11.0 (μ L %⁻¹), as stipulated by (Kaneko et al., 2008).

Eosinophils and rod counts are important, because they are responsible for promoting immune response. High counts indicate some inflammatory process or parasite in the animal's intestine (Berek, 2016). Counts below the reference values indicate the occurrence of stress (Budiño et al., 2004). In the present experiment, can assume that the significant difference ($p < 0.05$) found in these variables was due to some component of the diet that promoted a stress response in cases when the counts were lower. In the energy-deficient diet + MEC, the counts remained within the standard (1-11.0 μ L), indicating that in this diet, this component was not present and did not affect the animals.

Also observed a decrease in monocyte counts, which might have occurred owing to stress caused by changes in the environment, altered diet, daily management, and/or blood collection procedures. According to (Broom & Molento, 2004), experimental techniques affect the welfare of animals, causing stress, which affects the functioning of the immune system. Under stress conditions, higher secretion of cortisol occurs, which at high concentrations negatively affects immunity by exerting an inhibitory action on the production and functions of granulocytes, lymphocytes, and monocytes (Baptista, Bertani, & Barbosa, 2011). This hormone also decreases leukocyte production and phagocytosis of other defense cells such as eosinophils and affects the secretion of inflammatory cytokines (Tavares, Soares-Fortunato, & Leite-Moreira, 2000). Therefore, can infer that the immune system of the piglets was not compromised owing to the use of the enzymes, but rather was due to the experimental procedures conducted.

The results of economic viability, CI, and EEI are presented in Table 6. observed a higher ration price per kg of product in the diet with adequate energy levels with and without supplementation of MEC. Alternatively, energy-deficient diets with and without supplementation of MEC presented lower ration prices per kg of product. This increase and/or decrease in the price of diets was owing to the inclusion or exclusion of soybean oil and MEC in the diets.

Table 6. Analysis of economic viability with the use of an enzymatic complex in the piglet diet during the initial phase.

	Low energy diet + MEC ⁷	Low energy diet	Diet with adequate energy level + MEC	Diet with adequate energy level
FI ¹	54.43	57.75	55.24	55.97
BWG ²	12.92	12.45	13.48	13.13
Price ³	0.85	0.84	0.91	0.90
EV ⁴	3.62	4.09	3.75	3.85
EEI ⁵ (%)	79.76	70.51	74.42	72.54
CI ⁶ (%)	130.00	147.10	134.92	138.30

¹Ration I; ²Weight Gain; ³Ration Price kg⁻¹; ⁴Economical viability; ⁵Economic efficiency Index; ⁶Cost Index; ⁷Multi-enzymatic complex.

Economic viability increased in the group fed energy-deficient diet supplemented with MEC (3.62 R\$ kg⁻¹), showing a lower cost per kilogram of weight gained by the animal, followed by group fed diet an adequate level of energy supplemented with MEC, diet with adequate energy level, and energy-deficient diet.

Using the economic efficiency index, could confirm the economic viability results. The energy-deficient diet supplemented with MEC was the most efficient, with an economic efficiency of 79.76%; followed by the diet with an adequate energy level supplemented with MEC, which was 74.42% efficient; the diet with an adequate level of energy (72.54%); and the energy-deficient diet (70.51%).

Thus, the energy-deficient diet supplemented with MEC had a lower cost index (130.00%) than other diets; i.e., this diet had a lower cost of use. On the other hand, the energy-deficient diet presented higher FI than other experimental diets.

Conclusion

Multi-enzymatic supplementation in diets with lower energy levels (low energy diets) yielded results similar to those of diets with an adequate level of energy as well as better economic viability, demonstrating that feed costs can be reduced by altering nutritional levels (energy) without negatively affecting the performance and health of the animals.

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