Volume 4 • Issue 1 • 2022

Research Article

Archives of Veterinary and Animal Sciences

Increased Levels of Crude Proteins in Pre-fattening Pig Feeds Fermented with Multipurpose Autochthonous Microorganisms

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Received: March 30, 2022; **Published:** May 12, 2022

Summary

The purpose of this research was to determine crude protein increase in pre-fattening pig concentrate fermented with activated multipurpose autochthonous microorganisms (MAM). The study comprised three groups: 1) Pre-fattening pig concentrate (control); 2) 0.5 kg of feed/120 ml of MAM, till 3 kg, the mix was soaked in water in a 20 L bucket and fermented 12 hours; and 3) Same as described above, but following a 24-hour-fermentation. The presence of Salmonella, fungus, and yeast was determined according to the norms for non-lactic products. Crude protein was determined using a Kjeltec system 1. Each variant had three repetitions. Only the 24-h fermented concentrate reached and went above (26.5) the protein contents recommended for pre-fattening animals (23.7). This result may be linked to higher microbial growth over time. It is an easy sustainable option for weaned pigs' nutrition in swine production.

Keywords: *Autochthonous Multipurpose Microorganisms; Efficient Microorganisms; Fermented Feeds; Pigs; Protein Increase*

Introduction

Fermented liquid feed technology in pig nutrition is an ideal option to replace antibiotics as growth promoters after weaning and without their adverse effects. The product helps to repair the anatomical and physiological damage to the intestinal microvilli typical of the stage and promotes the establishment of *Lactobacillus* spp., *Streptococcus* spp., and other bacterial species with probiotic action on them. The reduction of the pH and the production of lactic acid and alcohol in the fermentation reduce the enteropathies (Rodríguez et al., 2021).

This simple variant ranges from proposals as simple as mixing the food with water to those that incorporate *Lactobacillus* spp. and preselected yeasts. The fermentation time takes from a few hours to days depending on the proposed purpose; it must always be stopped before the production of acetic acid begins, since the smell and taste that it gives to the feed is unpleasant for pigs (Missotten et al., 2015).

The final fermented product, in addition to the advantages mentioned, presents protein increases. A more digestible and better absorbed protein (Polyorach et al., 2018). However, perhaps due to

a refusal to decline antibiotics or hormones, some growers argue that the acquisition of suggested microbial cultures and their maintenance are beyond their resources and capabilities (Barreto et al., 2020).

Microbial mixtures of the type "efficient microorganisms" (EM) or "multipurpose autochthonous microorganisms" (MAM), are easy to prepare and preserve. They are composed of lactic acid bacteria (*Lactobacillus plantarum, L. casei* and *Streptoccus lactis*), phototrophs (*Rhodopseudomonas palustris* and *Rhodobacter spaeroides*), yeasts (*Saccharomyces cerevisiae* and *Candida utilis*), actinomycetes (*Streptomyces albus* and *S. griseus*) and filamentous fungi (*Aspergillus oryzae, Penicillium spp.* and *Mucor hiemalis*). These consortia are readjusted in a system in which one depends on the other and achieve a synergy for the exclusion of transient pathogens (Rodríguez et al., 2021; Barreto et al., 2021).

The microbial diversity of these mixtures makes it difficult to guide the time that fermentation should last to achieve fermented liquid feed with higher crude protein content. Therefore, this research aimed to determine the increase in crude protein in pre-fattening feeds fermented with MAM for 12 and 24 hours.

Materials and Methods

Preparation of indigenous multipurpose microorganisms (MAM)

The liquid mother, acquired at the Pastures and Forages Experimental Station Indio Hatuey, was propagated and activated as suggested by Barreto et al. (2021).) After two weeks, a product with a bittersweet odor was achieved, typical of lactic acid fermentations, with a pH of less than 3.5. This activated form was used in the experiment.

Concentrates

The experiment comprised three variants: 1) Pre-fattening pig concentrate (acted as control); the composition declared by the manufacturer is attached (Table 1). 2) A homogeneous mixture of 120 mL of MAM/0.5 kg of CP to a final weight of approximately 3 kg. The procedure was carried out in a 20 L bucket. The mixture was covered with water and fermented for 12 hours. 3) Same as above but fermented for 24 hours.

Components	Porcentage
Corn	54.55
Soja	37.50
Calcium	0.80
Phosphate	0.90
Salt	0.30
Nucleous porcine	2.50
Methionine	0.10
Lysine	0.40
Coline	0.15
Biotonic	0.10
Glucosil expanded	0.20
Sugar	2.50

Table 1: *Composition of pre-fattening pig concentrates according to the manufacturer.*

Collection and processing of feed samples

Dry feed as well as feed fermented with MAM for 12 and 24 hours was sampled by the quarter method. For this purpose, the corresponding tank was divided into four parts to take portions from two opposite quarters. The procedure was repeated successively to guarantee in each case the quantity equivalent to three replicates. They were transported to the laboratory in 1 kg polyethylene bags duly identified and sealed to avoid loss of humidity. They were worked at the time so as not to violate the action time of the MAMs.

Determination of the percentage of crude protein (CP)

It was established by the Kjeldahl method, using a Kjeltec I system. The CP contents were expressed as CP = N x 6.25 as established by Association of Official Analytical Chemist (AOAC, 1995). Each type of feed had three replicates. From these, statistical evaluation was carried out by means of a simple ANOVA. The HSD Tukey multiple comparison test was used to compare the means for each type of feed.

Results and Discussion

As can be seen, only the concentrate treated with the MAM for 24 hours reached the protein content required for pre-fattening (Table 2), as established in the Manual of technical procedures for pig rearing (Macías et al., 2015).

The criteria regarding nutritional requirements have been established under feeding conditions, racial crossbreeding and ownership of each country, so they differ when comparing them. In the case of Cuba, it has been regulated that growing pigs weighing between 5-10 kg should be provided with concentrates with 23.70% crude protein, a value that will decrease as the animals increase their weight (Macías et al., 2015). The one supplied by the manufacturer for these experiments did not meet the requirement.

Boonnop et al. (2009) and Polyorach et al. (2013), in independent experiments aimed at improving crude protein (CP) content in foods based on cassava (Manihot esculenta Crantz) through fermentation with Saccharomyces cerevisiae, achieved values between 30.4% and 47.0%, respectively. Remarkable results if one takes into account that before the treatments the CP percentages ranged between 2% and 3%. Polyorach et al. (2013) attributed the former change to high yeast growth (3.0 x 1011 cells/mL). Phenomenon associated with the ability of Saccharomyces cerevisiae to secrete extracellular enzymes (amylases, linamarase and cellulase) in the cassava mass and degrade starches and other polymers that contribute to the yeast growth increment.

Study groups	CP content (%) Replicates	CP average (%)
Dry concentrate (control)	16,4	16,1[a]
	16,5	
	15,6	
12 hours fermented concentrate	21,8	20,6[b]
	20,2	
	20,0	
24 hours fermented concentrate	25,8	26,5[c]
	27,0	
	26,7	

Legend: CP = crude protein. Different letters differ significantly ($p < 0.05$)

Table 2: *Crude protein (CP) values determined in the variants analyzed.*

More recently, Polyorach et al. (2018) carried out an experiment with fresh pulp and dry grated cassava (with CP contents equal to 3.1f% and 3.5f%, respectively) that they subjected to fermentations with: yeasts (Y), efficient microorganisms (EM) and a mixture of both (EMY). After three days of interactions microorganisms - substrate in the shade, followed by drying in the sun for 48 h, the CP contents of the fresh ground substrate were: 28.7e % (Y), 30.4d % (EM) and 31 .8c% (EMY). Meanwhile, the values obtained for dry scratches rose to: 42.1b% (Y), 44.2a% (EM) and 45.3a% (EMY). These authors attributed such high increases to the growth and proliferation of yeast, as well as to the implicit bacterial complex that conforms the EM mixture, acting as single cell proteins.

To the previous discussed, it is worth adding what is contributed by filamentous fungi, both in terms of cellular protein content and in the abundant production of extracellular enzymes. More detailed and current information about the advantages associated with the production of unicellular proteins by various routes is discussed in the proposal by Akwu Omede et al. (2018).

Concentrates fermented with MAM 24 hour is a sustainable option for weaned pigs' nutrition in swine production (Rodríguez et al., 2022). In a parallel experiment, in which the three variants previously analyzed were supplied to homogeneous groups of post-weaning pigs, it was found that fermented with MAM 24 hours promoted positive responses in blood parameters (Rodríguez et al., 2021).

Conclusions

The highest percentages of crude protein were obtained in the concentrates fermented with MAM for 24 hours, which may be due to a higher microbial population growth, with an emphasis on yeasts. It is a sustainable option and easy to apply in pig production.

Conflict of interests

They do not exist.

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