

# Influence of multi-enzyme (Liptozyme®) addition on performance and haematological parameters of growing pigs fed diets with fermented *Icacina mannii* meal

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**ABSTRACT:** The increasing cost and scarcity of maize in pig production necessitate the exploration of alternative energy feed resources such as fermented and pressed *Icacina mannii* meal (FPIM). A study was conducted to evaluate the effect of multi-enzyme (Liptozyme) supplementation on growth performance, blood profile, and cost benefits of growing pigs fed diets containing fermented and pressed *Icacina mannii* meal (FPIM). Twelve crossbred growing pigs with initial weights of 26–28 kg were randomly allotted to four dietary treatments in a completely randomised design with three replicates per treatment. Fermented and pressed *Icacina mannii* replaced maize at 0%, 10%, 20%, and 30% levels in diets designated as T1, T2, T3, and T4, respectively. Diets T2–T4 were supplemented with Liptozyme at 1 g/kg feed, while T1 served as the control. Data were analysed using one-way ANOVA and Duncan's multiple range test. Results showed that the control diet (T1) recorded the highest final body weight (50523.33 g), which was significantly superior ( $p < 0.05$ ) to the other treatments. Final weights for T2 and T3 were statistically similar (43733.33 g and 43500.00 g, respectively), while T4 had the lowest value. Feed intake followed a similar trend, while T4 recorded the lowest feed conversion ratio. Haematological indices were not negatively affected ( $P > 0.05$ ) by dietary treatments. In conclusion, enzyme supplementation reduced feed cost, but the inclusion of FPIM at higher levels, particularly 30%, was not economically beneficial.

**Keywords:** Growing pigs, haematology, serum, *Icacina mannii*, Liptozyme.

## INTRODUCTION

Maize is the major energy source and the most valued ingredient in monogastric animal diets (Niemi *et al.*, 2013). Research from Udedibie *et al.* (2004) and Assam (2014) reveals that up to 60% of pig diets may be comprised of maize. However, high cost has been imposed on the ingredient through its extensive use in human food, animal supplementary feeds and raw materials for industry. As a result, feed formulation levels are limited (Assam, 2014). The problem of high feed cost is largely associated with the high inclusion level of maize in the diet. Therefore, it is necessary to replace the conventional energy sources, such as maize totally or at least partially with other cheaper

unconventional alternatives in order to cut down the overall cost of feeding (Udedibie *et al.*, 2004). Earthball (*Icacina mannii*) is a shrub with a modified tuber. It is a source of carbohydrate and is not directly used by man (Assam, 2023). *Icacina mannii* has some anti-nutritional factors or components such as phytic acid, oxalic acid, cyanogenic glycosides and galactosamine, which may require it to be processed into fermented and pressed *Icacina mannii* meal.

It has been reported that enzyme supplementation in high-fibre pig feeds has gained attention in recent years due to its potential benefits (Valente *et al.*, 2024; Shipman

*et al.*, 2023). These include improved digestion and absorption of nutrients, increased feed intake, weight gain and better feed conversion ratio, reduced feed cost, improvement of gut health, reduced production of ammonia from excreta, altered population of micro-organisms in the gastrointestinal tract, and reduced output of excreta including N and P (Wang *et al.*, 2015; Shirmohammad and Mehri, 2011). Niemi *et al.* (2013) supported the role of exogenous enzymes in mitigating the limitations of high-fibre unconventional ingredients in pig nutrition.

However, adding multi-enzymes to pig high-fibre feeding with fermented and pressed *Icacina mannii* may be a solution to increase its digestibility. This idea was based on the previous work and provided a more straightforward way to mitigate the cost of pig production by improving the utilisation of nutrients and reducing the cost of production. Considering the contribution of all these techniques, the use of naturally fermented and pressed *Icacina mannii*, additionally supplemented with multi-enzymes, appears as a very promising local alternative to the feed energy component in pigs due to the increasing maize price.

Therefore, the objective of this study is to evaluate the effect of multi-enzyme (Liptozyme) supplementation on growth performance, blood profile and cost benefits of growing pigs fed diets containing fermented and pressed *Icacina mannii* meal (FPIM).

## MATERIALS AND METHODS

### Study location

The experiment was conducted at the Piggery Unit of the Teaching and Research Farm of the Department of Animal Science, Akwa Ibom State University, Obio Akpa Campus, Oruk Anam Local Government Area, Akwa Ibom State, Nigeria. Obio Akpa is located within the southern zone of Nigeria at a latitude 4.660°N to 4.981°N of the equator and longitude 7.521°E to 7.799°E of Greenwich meridian within annual rainfall ranging from 3500 - 5000 mm and a monthly average temperature of 25°C (Mapcarta, 2026).

### Feedstuff and processing of *Icacina mannii* tubers

Fresh *Icacina mannii* tubers were sourced around the university environment in Oruk Anam local government of Akwa Ibom State, Nigeria. The *Icacina mannii* tubers obtained were prepared into fermented and pressed *Icacina mannii* meal (FPIM).

Anaerobic fermentation was adopted as reported by Hahn *et al.* (1987), Udo *et al.* (2024) and Makanjuola *et al.* (2012) and (Assam, 2023). In anaerobic fermentation, peeled, sliced and washed *Icacina mannii* tubers were placed in a capped plastic bucket and allowed to ferment for 3 days (72 hours). The *Icacina mannii* tubers, after fermentation, were grated into mash and packed into

porous jute bags and pressed with a hydraulic jack machine for 12 hours to drain out excess water. The *Icacina mannii* meal was then hand-mashed with a homemade sieve to obtain fermented and pressed *Icacina mannii* meal prior to use in feeding trials.

### Experimental animals and their management

A total of twelve (12) hybrid (Landrace × large white) growing pigs of about 10-12 weeks of age were procured for the study. The pigs were managed in well-ventilated concrete pens equipped with concrete feeding and watering troughs. The pigs were fed daily at 8.00 hours with 5% of the pigs' body weight (Norachack *et al.*, 2004; Tram *et al.*, 2004). These quantities were adjusted weekly based on the highest weight in each treatment. Clean water was provided *ad libitum*, and routine and regular cleaning of the pens was carried out throughout the experiment.

### Experimental diet

Formulated grower's pig ration was given at 0- 1 week of experiment and then fed formulated ration using fermented and pressed *Icacina mannii* meal supplemented with Enzyme as shown in Table 1. The experimental diets were formulated by replacing maize with fermented and pressed *Icacina mannii* meal (FPIM) at 0%, 10%, 20%, and 30% inclusion levels. These graded levels were selected to evaluate the response of growing pigs to increasing dietary FPIM as an alternative energy source. Twelve crossbred growing pigs (26–28 kg) were randomly allotted to four dietary treatments in a Completely Randomised Design (CRD), with three replicates per treatment and one pig per replicate.

### Data collection

#### Determination of growth parameters

The initial live weight of the growing pigs at the beginning of the experiment was taken; subsequent weight was done on weekly basis in the morning hours. Feed intake was obtained by subtracting the feed left after consumption from the feed given. These values were used to obtain the following: total feed intake per pig per day, total weight gain, feed to gain ratio as follows:

$$\text{Daily feed intake} = \frac{\text{Total feed Intake}}{\text{number of days of feed consumed}}$$

$$\text{Daily weight gain} = \frac{\text{mean final body weight} - \text{mean Initial body weight}}{\text{Number of days}}$$

**Table 1.** Composition of experimental diets containing graded level of *laccina mannii* meal supplemented with enzymes.

Ingredients	T <sub>1</sub> (0)	T <sub>2</sub> (10.00)	T <sub>3</sub> (20.00)	T <sub>4</sub> (30.00)
Maize	47.00	37.00	27.00	17.00
FPIM	-	10.00	20.00	30.00
Wheat offal	5.50	5.50	5.50	5.50
Limestone	1.50	1.50	1.50	1.50
Soybean meal	21.50	21.50	21.50	21.50
Bone meal	1.47	1.47	1.47	1.47
Salt	0.25	0.25	0.25	0.25
Vitamin/ premix	0.25	0.25	0.25	0.25
Lysine	0.10	0.10	0.10	0.10
Enzyme	0.1	0.1	0.1	0.1
Methonine	0.10	0.10	0.10	0.10
Total	100	100	100	100
Crude protein	19.23	18.79	18.61	17.92
ME (kcal/kg)	2782.355	2698.395	2657.255	2615.55

$$\text{Feed conversion ratio} = \frac{\text{Average feed intake}}{\text{Average weight gain}}$$

The weighing of the pigs and feed consumed was done using hanging scale/top loading weighing scale.

### Haematology and blood chemistry analysis

At the end of the experiment, blood samples were collected from three weaner pigs per treatment through jugular vein puncture using sterilised needles and syringes into labelled sample bottles. The blood collected was for the determination of haematological indices and serum biochemical parameters.

Blood samples designated for haematological analysis were collected into bottles containing dipotassium ethylenediaminetetraacetic acid (EDTA) as an anticoagulant, while samples for serum biochemical analysis were allowed to clot before serum separation. Mean corpuscular haemoglobin concentration (MCHC), mean corpuscular volume (MCV), and mean corpuscular haemoglobin (MCH) were calculated using standard procedures and formulae as shown below:

$$\text{MCH} = \text{Hb} \times 10 / \text{RBC}$$

$$\text{MCV} = \text{PCV} \times 10 / \text{RBC}$$

$$\text{MCHC} = \text{Hb} \times 100 / \text{PCV}$$

### Experimental design

The design was a Completely Randomised Design. The statistical model of this design was:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where:  $Y_{ij}$  = single observation,  $\mu$  = overall mean,  $T_i$  = effects of treatment, and  $e_{ij}$  = Error of term, which is independently identically normally distributed with a zero mean and constant variance.

The data collected were subjected to analysis of variance (ANOVA) according to Steel and Torie (1980). The means were separated using Duncan's multiple Range Test as described by Duncan from the SPSS (2025) software package.

## RESULTS AND DISCUSSION

### Growth performance

The growth performance of growing pigs fed fermented and pressed *laccina manni* meal supplemented with enzyme is presented in Table 2. There were significant ( $p < 0.05$ ) differences among treatment means for weight gain, daily weight gain, daily feed intake and feed conversion ratio. Pigs fed the control diet (T1) recorded significantly ( $p < 0.05$ ) higher final weight gain than those fed the treatment diets (T2, T3, and T4). However, pigs on T2 and T3 showed comparable weight gain, while the lowest value was observed in T4. A similar trend was recorded for daily weight gain and daily feed intake. In contrast, pigs fed T1, T2, and T3 exhibited comparable and more efficient feed conversion ratios than those fed T4. The lower feed intake observed for pigs on T4 could be attributed to the poor palatability of *laccina manni*. This is in agreement with reports of Essien and Sam (2018), Assam (2014) and Udo *et al.* (2024), who reported that wild tubers are unpalatable.

**Table 2.** Growth performance of growing pigs fed fermented and pressed *Icacina Manni* meal supplemented with multi-enzyme.

Parameters	T <sub>1</sub> (control)	T <sub>2</sub> (10.00)	T <sub>3</sub> (20.00)	T <sub>4</sub> (30.00)	SEM
Initial weight (g)	28350.00	27933.33	28350.00	28250.00	558.89
Final weight gain (g)	50523.33 <sup>a</sup>	43733.33 <sup>b</sup>	43500.00 <sup>b</sup>	36750.00 <sup>c</sup>	233.00
Body weight gain (g)	22023.33 <sup>a</sup>	15591.67 <sup>b</sup>	15000.00 <sup>b</sup>	8333.33 <sup>c</sup>	546.99
Daily weight gain (g)	393.27 <sup>a</sup>	278.42 <sup>b</sup>	267.30 <sup>b</sup>	148.81 <sup>c</sup>	5.83
Feed intake (g)	55533.33 <sup>a</sup>	50673.33 <sup>b</sup>	50493.33 <sup>b</sup>	36316.67 <sup>c</sup>	193.57
Daily feed intake (g/day)	991.67 <sup>a</sup>	904.88 <sup>b</sup>	901.67 <sup>b</sup>	648.18 <sup>c</sup>	3.45
Feed conversion ratio	2.52 <sup>b</sup>	2.76 <sup>b</sup>	3.38 <sup>b</sup>	4.41 <sup>a</sup>	0.26

<sup>abc</sup> mean on the same row bearing different superscripts are significantly different ( $p < 0.05$ ).

**Table 3.** The Haematological indices of weaner pigs diets containing graded levels of fermented and processed *Icacina mannii* meals supplemented with enzymes.

Parameters	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	SEM	Normal Range
RBC ( $\times 10^3/\text{mm}^3$ )	5.80 <sup>c</sup>	6.20 <sup>b</sup>	6.55 <sup>a</sup>	5.77 <sup>c</sup>	0.07	5-8
PCV %	35.67 <sup>c</sup>	38.00 <sup>b</sup>	40.33 <sup>a</sup>	35.67 <sup>c</sup>	0.50	26-43
Hb (g/dl)	11.37 <sup>c</sup>	12.00 <sup>ab</sup>	12.40 <sup>a</sup>	11.77 <sup>bc</sup>	0.15	9-14
MCV	61.76	60.62	61.52	61.94	0.45	42-62
MCH	19.58 <sup>b</sup>	19.35 <sup>b</sup>	18.92 <sup>c</sup>	20.38 <sup>a</sup>	0.12	14-24
MCHC	31.8 <sup>b</sup>	31.5 <sup>b</sup>	30.25 <sup>c</sup>	33.00 <sup>a</sup>	2.64	29.-6
WBC ( $\times 10^3/\text{mm}^3$ )	13.33 <sup>a</sup>	12.56 <sup>b</sup>	12.61 <sup>b</sup>	13.22 <sup>a</sup>	0.19	8.7-37.9
Neutrophils (%)	47.00 <sup>ab</sup>	45.33 <sup>a</sup>	48.00 <sup>ab</sup>	48.67 <sup>a</sup>	0.85	16.6-73.10
Lymphocytes (%)	46.67	45.00	46.00	45.67	1.46	12.5-70.10
Monocytes (%)	4.33	4.00	3.67	3.67	0.29	0-5.0
Eosinophils (%)	2.00	2.33	2.33	2.00	0.24	0-6.0
Basophils (%)	0.00	0.00	0.00	0.00	0.00	0-3.0

<sup>abc</sup> Means bearing different letters of superscript within the same row differ significantly ( $p < 0.05$ ) RBC = red blood cell; PCV = packed cell volume; Hb = Haemoglobin; WBC = white blood cell; MCH = mean corpuscular haemoglobin; MCHC = mean corpuscular haemoglobin concentration; MCV = mean corpuscular volume, SEM = Standard Error of mean.

The lower weight gain and poor feed conversion ratio recorded for T<sub>4</sub> could be associated with reduced feed intake and quantity of enzyme supplemented because the amount of enzyme may not have been enough for the quantity of *Icacina manni*. However, weight gain recorded in this study is higher than the weight obtained by Assam (2023), who reported 5333.00- 9000 g body weight of weaner pigs fed fermented and pressed *Icacina manni*. Also, this study recorded a better feed conversion ratio than Assam (2023). The discrepancies in weight gain and feed conversion ratio could be attributed to the addition of exogenous enzyme, which improves the availability and digestibility of nutrients, thereby enhancing growth. Several studies have shown that the addition of exogenous multi-enzyme in feed has a positive effect on growth performance in non-ruminant animals (Song *et al.*, 2022) for weaned pigs; Rahman *et al.* (2014) for broiler chickens.

### Haematology of growing pigs fed diets with fermented *Icacina mannii* meal

The effect of fermented and *Icacina manni* supplemented with multi-enzyme on haematology and serum biochemistry of growing pigs is presented in Table 3. The result showed that fermented and pressed *Icacina manni* supplemented with multi-enzyme based diets significantly ( $p < 0.05$ ) influenced red blood cell, packed cell volume, haemoglobin, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, white blood cells and neutrophils. White blood cell components, including lymphocytes, monocytes, and eosinophils, were not significantly ( $P > 0.05$ ) affected by the dietary treatments. Nevertheless, all the haematological parameters evaluated remained within the normal physiological ranges reported by Fasuyi *et al.* (2013) and Jelek *et al.* (2018). This indicated the diets were of good quality and had a

**Table 4.** Biochemical parameters of growing pigs fed diets containing graded level of *l-cacina mannii* meal supplemented with Enzyme.

Parameters	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	SEM	Normal Range
Total protein (g/dl)	7.05 <sup>c</sup>	7.36 <sup>a</sup>	7.31 <sup>a</sup>	7.17 <sup>ab</sup>	0.073	4.4 - 7.4
Albumin(g/dl)	3.94 <sup>c</sup>	4.19 <sup>bc</sup>	4.08 <sup>a</sup>	4.07 <sup>ab</sup>	0.08	1.9 - 3.9
Total cholesterol (mg/dl)	92.26 <sup>a</sup>	83.85 <sup>b</sup>	81.30 <sup>bc</sup>	78.34 <sup>c</sup>	1.181	81.4 - 134.1
Alkaline phosphate (g/dl)	66.67 <sup>a</sup>	63.33 <sup>ab</sup>	60.00 <sup>b</sup>	59.33 <sup>b</sup>	1.66	14.2 - 89.10
Globulin (g/dl)	3.11	3.17	3.23	3.10	0.07	2.5 - 3.5

<sup>abc</sup> Means bearing different letters of superscript within the same row differ significantly ( $p < 0.05$ ).

deleterious effect on the immune system of the pigs. The result obtained in this study corroborates the study of Amaefule *et al.* (2020), who reported no significant effect of low-energy and low-protein feed supplemented with multi-enzyme on the haematological parameters of growing pigs. Researchers have reported that blood components are affected by the quality, quantity and toxicity of diets ingested. Any factor that affects blood may surely affect the health, growth and reproduction. Etim *et al.* (2013) noted that haematological parameter values below the reference range is attributed to malnutrition.

#### Serum biochemistry of growing pigs fed diets with fermented *l-cacina mannii* meal

The serum biochemical profile of growing pigs fed fermented and pressed *l-cacina mannii* meal supplemented with multi-enzyme is presented in Table 4. Total protein values (7.05, 7.36, 7.31, and 7.17 g/dL for T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, and T<sub>4</sub>, respectively) differed significantly ( $p < 0.05$ ) among the treatments, with pigs fed the enzyme-supplemented diets recording higher values than those on the control diet (T<sub>1</sub>). This observation suggests adequate protein utilisation and indicates that the experimental diets were of good nutritional quality. The result obtained in this study strengthened the earlier work of Assam (2023), who reported a total protein range of 7.10-7.38 for weaner pigs fed processed *l-cacina mannii* meal. The values for albumin were 3.94g/dl, 4.19g/dl, 4.08g/dl and 4.07g/dl for T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub>, respectively. These values were significantly ( $p < 0.05$ ) higher in treatment groups than control, and were above the normal range values reported by Fusiya *et al.* (2013). This suggested that the animal may have been dehydrated. Total cholesterol values decreased progressively from T<sub>1</sub> to T<sub>4</sub>, with values of 92.26, 83.85, 81.30, and 78.34 mg/dL recorded for T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, and T<sub>4</sub>, respectively. The control group (T<sub>1</sub>) had significantly ( $p < 0.05$ ) higher cholesterol values compared to the treatment groups. A similar trend was observed for alkaline phosphatase, with values of 66.67, 63.33, 60.00, and 59.33 IU/L for T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, and T<sub>4</sub>, respectively. Globulin values (3.11, 3.17, 3.23, and 3.10 g/dL for T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub>, and T<sub>4</sub>, respectively) did not differ significantly ( $p > 0.05$ ) among

the treatments. However, the values obtained for total cholesterol, alkaline phosphatase, and globulin were within the normal physiological ranges reported by Fasuyi *et al.* (2013) and Jekek *et al.* (2018). This indicates that the experimental diets did not adversely affect liver function or normal metabolic activities in the pigs.

#### Conclusion

Fermented and pressed *l-cacina mannii* meal supplemented with Liptozyme can replace maize in growing pig diets at up to 20% inclusion without adverse effects on growth performance, haematological and serum biochemical parameters. However, inclusion levels above 20% reduced feed intake, weight gain, and feed efficiency. Therefore, a maximum inclusion level of 20% is recommended for optimal performance, health, and blood profile of growing pigs

#### CONFLICT OF INTEREST

The authors declares that they have no conflict of interest.

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