

## Physiological response of weaner pigs fed rice-based indigenous microorganisms in the humid tropics

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### Abstract

Pig production has strong potential to bridge the protein deficiency gap in the humid tropics. Quality nutrition is essential for increased growth and productivity in any pig farming enterprise. This research examined the effect of rice-based indigenous microorganisms (IMOs) solution on the haematological and serum biochemical indices of weaner pigs in the humid tropics. A total of 24 weaner pigs (Aged 12-weeks) with an average body weight of  $10.21 \pm 0.4$  kg were randomly assigned to three treatments (T) group and four replications each in a completely randomized design (CRD). The treatments comprised three levels of rice IMOs in water: T1 (0 mL/L<sup>-1</sup>), T2 (5 mL/L), and T3 (10 mL/L<sup>-1</sup>). The weaner pigs were exposed to the rice IMOs for a period of 20 weeks. Data were collected on haematology and serum biochemical parameters. The results showed that rice IMOs significantly ( $p < 0.05$ ) increased haemoglobin ( $16.59 \pm 0.05$  g/dL), packed cell volume ( $41.80 \pm 0.12\%$ ), red blood cells ( $14.50 \pm 0.16 \times 10^6/\mu\text{L}$ ), and white blood cells ( $19.80 \pm 0.18 \times 10^3/\mu\text{L}$ ) concentrations of pigs in T3. Neutrophil ( $28.00 \pm 0.08 \%$ ), eosinophil ( $8.00 \pm 0.26 \%$ ), and monocyte ( $6.00 \pm 0.10 \%$ ) blood concentrations were higher ( $p < 0.05$ ) at T2, with lowered lymphocyte compared to other treatments. Compared with IMOs-treated pigs, total protein, globulin, alanine aminotransferase, aspartate aminotransferase, and total cholesterol were higher in the control group (T1). Alkaline phosphate ( $37.12 \pm 0.18$   $\mu\text{L/L}$ ) and urea ( $45.00 \pm 0.18$  mg/dL) concentrations were equally higher ( $p < 0.05$ ) for T3 pigs. However, albumin concentration showed a non-significant effect ( $p > 0.05$ ) on the pigs. Based on these findings, the study concluded that rice-based IMOs fed at 10 mL/L improved the physiological health status of weaner pigs in the humid tropics.

**Keywords:** haematology, indigenous microorganisms, probiotics, serum biochemistry.

## Resposta fisiológica de leitões desmamados alimentados com microrganismos indígenas à base de arroz nos trópicos úmidos

### Resumo

A produção de suínos possui grande potencial para reduzir a deficiência proteica nos trópicos úmidos. Uma nutrição de qualidade é essencial para aumentar o crescimento e a produtividade em qualquer sistema de criação de suínos. Esta pesquisa avaliou o efeito de uma solução de microrganismos indígenas (IMOs) à base de arroz sobre os índices hematológicos e bioquímicos séricos de leitões desmamados nos trópicos úmidos. Um total de 24 leitões desmamados (com 12 semanas de idade), apresentando peso corporal médio de  $10,21 \pm 0,4$  kg, foi distribuído aleatoriamente em três tratamentos (T), com quatro repetições cada, em delineamento inteiramente casualizado (DIC). Os tratamentos consistiram em três níveis de IMOs de arroz adicionados à água: T1 (0 mL/L<sup>-1</sup>), T2 (5 mL/L) e T3 (10 mL/L<sup>-1</sup>). Os leitões foram expostos aos IMOs de arroz por um período de 20 semanas. Os dados coletados incluíram parâmetros hematológicos e bioquímicos séricos. Os resultados mostraram que os IMOs de arroz aumentaram significativamente ( $p < 0,05$ ) as concentrações de hemoglobina

( $16,59 \pm 0,05$  g/dL), volume globular ( $41,80 \pm 0,12\%$ ), hemácias ( $14,50 \pm 0,16 \times 10^6/\mu\text{L}$ ) e leucócitos ( $19,80 \pm 0,18 \times 10^3/\mu\text{L}$ ) nos suínos do T3. As concentrações sanguíneas de neutrófilos ( $28,00 \pm 0,08\%$ ), eosinófilos ( $8,00 \pm 0,26\%$ ) e monócitos ( $6,00 \pm 0,10\%$ ) foram maiores ( $p < 0,05$ ) no T2, com redução de linfócitos em comparação aos demais tratamentos. Em comparação aos suínos tratados com IMOs, as concentrações de proteína total, globulina, alanina aminotransferase, aspartato aminotransferase e colesterol total foram maiores no grupo controle (T1). A fosfatase alcalina ( $37,12 \pm 0,18$  IU/L) e a ureia ( $45,00 \pm 0,18$  mg/dL) também apresentaram valores mais elevados ( $p < 0,05$ ) nos animais do T3. Entretanto, a concentração de albumina não apresentou efeito significativo ( $p > 0,05$ ) entre os tratamentos. Com base nesses resultados, o estudo concluiu que os IMOs à base de arroz fornecidos na concentração de 10 mL/L melhoraram o estado fisiológico de saúde dos leitões desmamados nos trópicos úmidos.

**Palavras-chave:** hematologia, microrganismos indígenas, probióticos, bioquímica sérica.

## 1. Introduction

Inadequate food supply is a widespread issue in both industrialized and developing countries, especially in tropical regions (Pawlak; Kołodziejczak, 2020). The rapidly growing global human population has increased the demand for animal protein, raising serious concerns about food security and sustainable livestock production. To meet the growing demand for animal-source protein, it is critical to prioritize the production of short-cycle farm animals such as pigs, poultry, and rabbits (Machebe et al., 2016; Okosun et al., 2019). Pigs (*Sus scrofa domestica*) are among the livestock species that should be promoted to improve global food security because of their numerous zootechnical advantages. These advantages include a short reproductive cycle, high prolificacy, high adaptability, and the ability to survive on diverse feed sources (Food and Agriculture Organization, 2012). Pork is an ideal source of animal protein for human diets (Ahmad et al., 2018). Poor growth due to health challenges hinders the production of pigs, and it stands as a major livestock products which could alleviate protein intake deficit.

Public outcry over potential concerns linked with antibiotic resistance to human health has spurred interest in pig nutrition and the widespread use of antibiotic-free feeding systems. Consequently, feed additives that are used in pig feeding programs as in-feed antibiotic replacements have been developed (Caroline, 2019). Possible alternatives to antibiotics include vaccination programs, proper housing design, production integration systems, biosecurity plans, disease prevention, proper animal management to minimize stress, and the use of high-quality feed and water. Finally, the use of bioactive substances such as plant additives, probiotics, prebiotics, antioxidants, and vitamins has also been reported (Ali et al., 2022; Moreira; Ponce, 2024). Probiotics are living microorganisms that give the host animal health benefits when administered in sufficient doses.

Although a wide variety of bacteria and fungi have been shown to possess probiotic qualities, lactic acid bacteria, *Bacillus*, and yeast strains are the most often utilized probiotics in animal feed (Agregán-Pérez et al., 2021). Probiotics have the ability to combat pathogenic microbes through a variety of mechanisms, such as immune system regulation, anti-adhesive action, and the generation of antimicrobial compounds that strengthen the intestinal epithelial barrier in the animal body system (Gresse et al., 2017). Also, probiotics are capable of modulating the immune system's physiology of the animal (Cardelle-Cobas et al., 2022).

Presently, many pig farmers in the humid tropics have shown interest in the production and use of rice-based IMOs in pig production to enhance production and health conditions of pigs. IMOs are microscopic organisms that are endemic to the environments in which they live (Ikeh et al., 2025). IMOs comprises, fungus and bacteria that coexist symbiotically with their hosts in all known living forms. They are a diverse group of naturally occurring microbes that live in soil and on other surfaces with the capacity to participate in bio-composting, biodegradation, and bioleaching processes. The production of indigenous microorganisms involves encasing living microorganisms that are naturally present in a habitat or ecological niche and adding them to farm animal feed to aid in anaerobic digestion (Kumar; Gopal, 2015).

According to Sanap et al. (2019), when given in sufficient proportions, living organisms improve animal health by promoting the growth and proliferation of beneficial microbiota, which in turn improves the intestinal health of farm animals (Walter et al., 2010). In addition, it produces antimicrobial substances, enhances the antibody-mediated immune response, and aids in digestion and the prevention of enteric pathogen colonization in the intestine of animals (Torres-Maravilla et al., 2024). In view of the aforementioned, IMOs may have the ability to interfere with the physiological health of pigs.

The health status of domestic animals has been evaluated using hematological and serum biochemical analyses.

These evaluations may indicate several abnormal physiological alterations, such as unpleasant or stressful reactions, inflammatory or infectious processes, or alterations in the physiology or cellular makeup of important organs like the kidney and liver (Chen; Luo, 2023). Currently, there is limited scientific information on the haematological and biochemical responses of pigs fed rice-based IMO in the humid tropics. This study aimed to evaluate the haematological and serum biochemical indices of weaner pigs exposed to rice-based IMO solution in drinking water.

## 2. Materials and Methods

### 2.1 Study period and area

This study lasted 22 weeks, comprising a 2-week acclimatization period for the weaned pigs to the new environment and a 20-week feeding trial with the test ingredients. The study was carried out in the piggery unit located at the Department of Animal Science Teaching and Research Farm, University of Nigeria, Nsukka (Figure 1). The study area is located in Nsukka Local Government Area (latitude  $6^{\circ} 51'39''$  N and longitude  $7^{\circ} 21'39''$  E), Enugu State, Nigeria. The microclimate of the area is a humid tropical setting with a bimodal annual rainfall ranging between 1155 and 1955 mm, as reported by Uguru et al. (2011). During the experiment, the average ambient temperature value was  $30.58^{\circ}\text{C}$  with a mean relative humidity of 71.7%. The photoperiodicity of the study area was 12 h light vs. 12-hour dark cycle with no artificial light being applied.

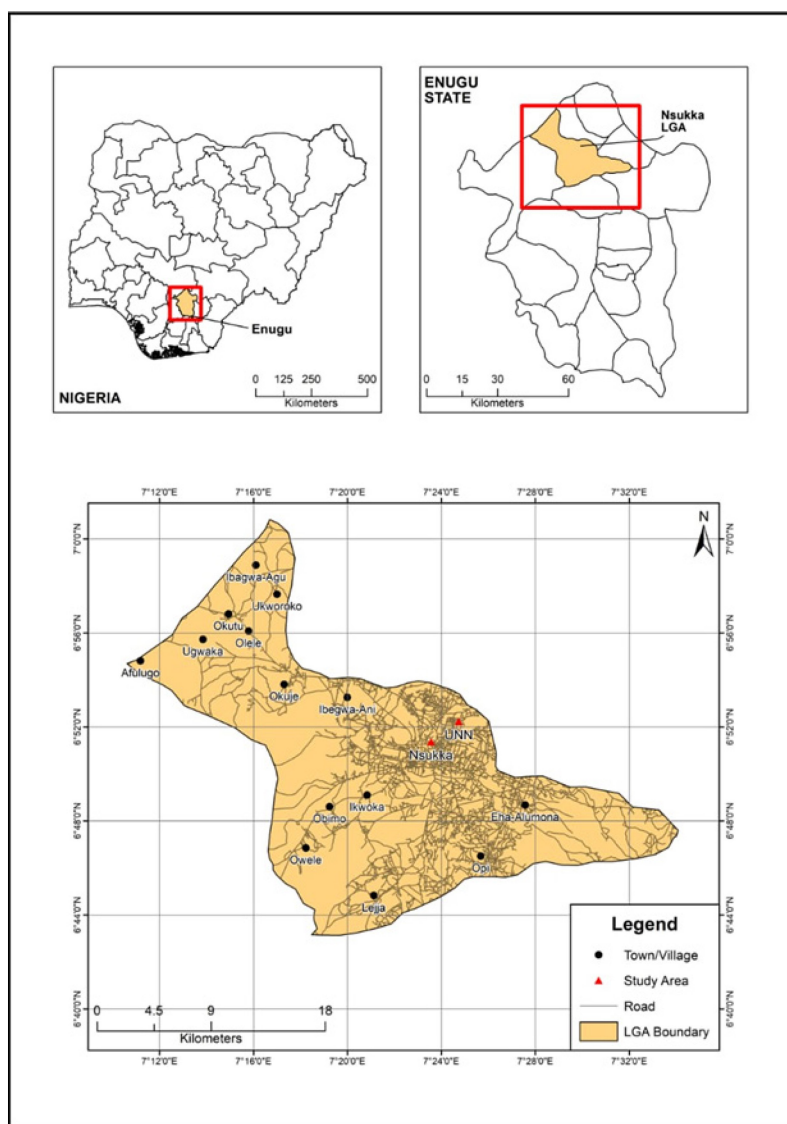


Figure 1. Map showing the study area (Nsukka, Enugu State, Nigeria). Source: Authors, 2026.

### 2.2 Preparation of rice-based IMOs solution

A 10 kg quantity of rice was steamed, allowed to cool, and poured into a sieve. The top of the sieve was wrapped with a paper sheet as the lid before being buried in the soil near the rhizosphere under the shade of trees and allowed to stay for 7 days undisturbed. Afterwards, the sieve was exhumed, and the content (rice, which is expected to be covered with white film as a result of the activities of indigenous microbes) was weighed and mixed with brown sugar in a ratio of (1:1) and homogenized in a 20 L container. The container was covered and protected from direct sunlight by keeping it in a cool environment under room temperature (25 °C) for an extra 7 days before storage in a 25 L gallon (Figure 2).



Figure 2. Preparation of rice-based IMOs. A: Steamed rice covered with a paper sheet, B: Covered rice in a sieve buried, C: Exhumed content covered in white film, D: Weighing of the decomposed rice, E: Weighing of brown sugar, F: Homogenization of rice and brown sugar, G: Homogenized product, H: Air tightly covered, and I: Rice-based IMOs. Source: Ikeh et al. (2025).

### 2.3 Experimental animal, treatments, and diets

A total of 24 crossbreed weaner pigs (aged 12 weeks) with an initial average body weight of  $10.21 \pm 0.4$  kg were procured from reputable farms in the Nsukka area and housed in a pen measuring 2.5 x 1.5 x 1.5 m. On arrival at the study area, the pigs were given 10 g of glucose in 5 L of water and allowed to acclimatize for 2 weeks. Pigs were randomly distributed according to live body weight into three treatments, replicated four times (two pigs per replicate), corresponding to T1 (control; No IMOs), T2 (5 mL IMOs/litre of water), and T3 (10 mL IMOs/litre of water). Pigs were fed at 5% of body weight. The rice-based IMO solution was first prepared in water and then incorporated into the feed at a ratio of (1:2) (1 kg of feed mixed with 2 L of water containing the IMO solution). Table 1 presents the total composition of the experimental diet used in the Nsukka area and housed in a pen measuring 2.5 x 1.5 x 1.5 m in the study.

Table 1. Experimental diet.

<b>Feedstuff</b>	<b>Quantity</b>
Yellow maize	8.50
Cassava chips	28.00
Dried cassava peel	20.00
Soya bean meal	17.00
Bambara nut meal	19.00
Dried blood meal	6.50
Lysine	0.25
Methionine	0.25
Salt	0.25
Mineral vitamin premix	0.25
Total	100.00
<b>Calculated composition</b>	
Crude protein (%)	18.16
Metabolizable energy (kcal/kg <sup>-1</sup> )	2874.21
Calcium (%)	0.79
Available phosphorus (%)	0.41
Lysine (%)	1.29
Methionine (%)	0.53

Note: Source: Authors, 2026.

#### 2.4 Sample collection

At 20 weeks of the study, 6 mL blood samples were taken using a syringe (10 mL) and needle (21 gauge) through the cranial vena cava from 2 pigs per replicate into both 10 mL dry and ethylene diamine tetra acetic (EDTA) tubes for both serum biochemistry and haematological evaluation, respectively. The samples were quickly stored in an ice box and transferred to the Department of Animal Science, Nutrition and Biochemistry laboratory for haematological and serum biochemical analysis, within three hours post-sampling.

##### 2.4.1 Haematological indices

Haematological indices (haemoglobin, packed cell volume, red blood cells, white blood cells, monocytes, eosinophils, neutrophils, and lymphocytes) were examined with the use of an automated Sysmex analyzer as reported by John (2024).

##### 2.4.2 Serum biochemical indices

Serum biochemical variables were determined by using the Universal Clinical Auto Analyzer (Biochemistry Analyzer, BA-A-120, Boeuropeak Ltd, China) as reported by Hagan et al. (2022) and Adegun et al. (2023) for total protein (biuret method), albumin (colorimetric method), alkaline phosphate (ALP) (kinetic method), urea (Berthelot method), total cholesterol (enzyme-colorimetric method), alanine aminotransferase (ALT) and aspartate aminotransferase (AST) (UV-kinetic method). However, Globulin (g/dL) = Total protein (g/dL) – Albumin (g/dL) (Zaitsev et al., 2020).

#### 2.5 Data analysis

The data generated were arranged in a completely randomized design (CRD) and analyzed using one-way

analysis of variance (ANOVA) with IBM Statistical Package for the Social Sciences (SPSS v. 23). The analysis was conducted under the assumptions of approximate normality and homogeneity of variance inherent to the ANOVA model. Significantly varied means were separated using Duncan Multiple Range Test (Duncan, 1955). Although more conservative post-hoc tests exist, the *Duncan* Multiple Range Test was adopted due to its sensitivity in detecting differences among treatment means in animal science studies with multiple experimental groups. The mathematical model applied in the experiment was  $Y_{ij} = \mu + X_i + \epsilon_{ij}$ , where  $Y_{ij}$  is the individual observation,  $\mu$  is the population mean,  $X_i$  is the treatment effect of IMOs, and  $\epsilon_{ij}$  is the experimental error.

### 3. Results

#### 3.1 Haematological indices

Research results showed significant differences ( $p < 0.05$ ) in haemoglobin, packed cell volume, red blood cells, white blood cells, lymphocytes, neutrophils, eosinophils, and monocytes concentrations among treatments (Table 2). The findings indicated a significantly ( $p < 0.05$ ) higher concentration of haemoglobin, packed cell volume, red blood cells, and white blood cells for pigs in 10 mL/L IMOs (T3). However, the concentrations of neutrophils, eosinophils, and monocytes were significantly ( $p < 0.05$ ) higher with lowered lymphocyte count for pigs in T2 (5 mL/L IMOs) compared to those of pigs in T3. Pig in the control group had the lowest neutrophil, eosinophil, and monocyte concentrations.

Table 2. Haematology parameters measured in pigs fed rice indigenous microorganism (IMOs) solution.

Parameters	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	P. Value
	(Control)	(5 mL/L)	(10 mL/L)	
Haemoglobin (g/dL)	13.86 ± 0.26 <sup>b</sup>	14.35 ± 0.23 <sup>b</sup>	16.59 ± 0.05 <sup>a</sup>	0.03**
Packed cell volume (%)	35.50 ± 0.15 <sup>c</sup>	38.50 ± 0.08 <sup>b</sup>	41.80 ± 0.12 <sup>a</sup>	0.02**
Red blood cells (×10 <sup>6</sup> /μL)	7.90 ± 0.04 <sup>c</sup>	9.90 ± 0.26 <sup>b</sup>	14.50 ± 0.16 <sup>a</sup>	0.02**
White blood cells (×10 <sup>3</sup> /μL)	17.80 ± 0.11 <sup>c</sup>	18.50 ± 0.10 <sup>b</sup>	19.80 ± 0.18 <sup>a</sup>	0.01**
Lymphocytes (%)	77.00 ± 0.26 <sup>a</sup>	58.00 ± 0.23 <sup>c</sup>	75.00 ± 0.05 <sup>b</sup>	0.04**
Neutrophils (%)	15.00 ± 0.15 <sup>c</sup>	28.00 ± 0.08 <sup>a</sup>	16.00 ± 0.12 <sup>b</sup>	0.03**
Eosinophils (%)	6.00 ± 0.04 <sup>b</sup>	8.00 ± 0.26 <sup>a</sup>	6.00 ± 0.16 <sup>b</sup>	0.01**
Monocytes (%)	2.00 ± 0.11 <sup>c</sup>	6.00 ± 0.10 <sup>a</sup>	3.00 ± 0.18 <sup>b</sup>	0.02**

Note: <sup>abc</sup> means with different superscripts within rows differ significantly ( $p < 0.05$ ). Source: Authors, 2026.

#### 3.2 Serum biochemical indices

In Table 3, except for albumin index ( $p > 0.05$ ), significant ( $p < 0.05$ ) differences among treatments were reported in total protein, globulin, alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), total cholesterol, and urea of weaner pigs. In comparison with the rice IMOs treated groups (T2 and T3), total protein, globulin, ALT, AST, and total cholesterol showed a higher mean index ( $p < 0.05$ ) in the control group (T1). Furthermore, ALP and urea concentrations in the serum were significantly higher ( $p < 0.05$ ) for T3 pigs.

Table 3. Serum biochemical indices of pigs fed rice indigenous microorganism (IMOs) solution.

Parameters	T <sub>1</sub> (Control)	T <sub>2</sub> (5 mL/L)	T <sub>3</sub> (10 mL/L)	P. Value
Total protein (g/dL)	7.06 ± 0.26 <sup>a</sup>	6.50 ± 0.23 <sup>b</sup>	6.14 ± 0.13 <sup>c</sup>	0.02 <sup>**</sup>
Albumin (g/dL)	3.88 ± 0.62	4.79 ± 0.08	4.33 ± 0.12	0.10 <sup>NS</sup>
Globulin (g/dL)	4.18 ± 0.04 <sup>a</sup>	1.70 ± 0.26 <sup>b</sup>	1.80 ± 0.16 <sup>b</sup>	0.00 <sup>**</sup>
ALP (IU/L)	35.22 ± 0.11 <sup>b</sup>	33.67 ± 0.10 <sup>c</sup>	37.12 ± 0.18 <sup>a</sup>	0.04 <sup>**</sup>
ALT (IU/L)	49.15 ± 0.26 <sup>a</sup>	23.30 ± 0.23 <sup>b</sup>	18.01 ± 0.05 <sup>c</sup>	0.01 <sup>**</sup>
AST (IU/L)	64.50 ± 0.15 <sup>a</sup>	29.01 ± 0.08 <sup>b</sup>	26.17 ± 0.12 <sup>c</sup>	0.03 <sup>**</sup>
Total cholesterol (mg/dL)	174.12 ± 0.04 <sup>a</sup>	149.69 ± 0.26 <sup>b</sup>	147.68 ± 0.16 <sup>c</sup>	0.05 <sup>**</sup>
Urea (mg/dL)	39.54 ± 0.11 <sup>b</sup>	36.54 ± 0.10 <sup>c</sup>	45.00 ± 0.18 <sup>a</sup>	0.03 <sup>**</sup>

Note: <sup>abc</sup> means with different superscripts within rows differ ( $p < 0.05$ ) significantly, ALP: Alkaline phosphate, ALT: Alanine aminotransferase, AST: Aspartate aminotransferase, NS: Not significant. Source: Authors, 2026.

#### 4. Discussion

Haematological parameters serve as essential indicators of physiological status, health, nutritional condition, and responses to environmental or dietary stressors in farm animals (Fanta et al., 2024). According to Schaefer et al. (2055), the majority of the reported haematological indices in this study fell within the reference range for blood obtained from pigs. These findings are in agreement with Harikrishnan et al. (2011) and Parthasarathy & Ravi (2011), who reported that probiotics significantly ( $p < 0.05$ ) increased or contributed to high red blood cells and haemoglobin concentrations. In contrast, Hossein-Ali et al. (2014) showed that feeding of *Bacillus subtilis* and *Bacillus licheniformis* in growing lambs resulted in a significant ( $p < 0.05$ ) decrease in the level of haemoglobin and packed cell volume concentrations.

However, Frimpong et al. (2021) reported a non-significant ( $p > 0.05$ ) effect of different probiotic products on the haematological profile of large white gilts, except in basophil concentration. Elevation in some haematological indices has been linked to the absorption of vitamin B and iron in the small intestine as a result of probiotics (Rusu et al., 2020). Nonetheless, it is important to note that erythropoietic indices improved with the administration of rice-IMOs. These observed increases may have enhanced organ development, maturity, and growth of weaner pigs, as opined by Czech et al. (2017).

The body's defense mechanisms mostly rely on the activities of the white blood cells to fight disease-causing microbes in the body. The findings of this study showed significantly higher concentrations of white blood cells in the T<sub>3</sub> pigs compared to other treatments. Higher white blood cells in the body system could depict diseased situations, conclusion of intense exercise or feed intake, and it could also happen in suckling piglets, as well as in sows in the latter stages of their gestation and right after farrowing (Czech et al., 2017). It may be inferred that the elevated white blood cell concentration in this study could be a result of the body's defense mechanism trying to balance the influx of indigenous microorganisms fed to the animal.

Probiotics have been shown by Mashayekhi et al. (2018) to improve the immune system and white blood cell count. The beneficial effect of IMOs as a probiotic may have resulted from the immunostimulant properties of their outer cell wall components, such as glucan, chitin, and mannan. These components of the outer walls have been reported to stimulate the action of lactic acid bacteria, which is triggered by the production of enzymes that break down bile salts and render them unconjugated (Min et al., 2019). Since probiotics are known to encourage the growth of lactic acid bacteria, which in turn produce antibacterial compounds and stimulate the production of immunoglobulin, it may be concluded that IMOs, as a potent probiotic, enhanced the immune response of the pigs (Sjofjan et al., 2021).

In this study, lymphocyte concentration decreased at 5 ml/L IMOs concentration. It, however, increased slightly at 10 mL/L IMOs concentration. A similar decline in lymphocyte concentration had been reported in pigs fed probiotics (*Lactobacillus acidophilus*) (Sanchez et al., 2019). However, the authors also reported that an oral administration of *B. subtilis* resulted in an up-regulated ratio of intestinal mucosa-associated lymphocytes of pigs. Physiologically, it might seem that feeding rice-IMOs potentially lowered total circulatory immune cells, specifically lymphocytes, as shown in this study. Although the exact mechanism underlying this response

remains unknown, rice-IMOs supplementation likely primed or prepared the pigs, resulting in a lower need for stress or health challenges.

Probiotics could have achieved the above results by various mechanisms, such as depriving pathogens of sites of attachment, the needed nourishment, and also the use of space within the intestinal mucosa, which could have been occupied by pathogens (Lee; Lillehoj, 2017). This mechanism, often referred to as competitive exclusion, leads to the elimination of the pathogens, especially bacteria, along with faeces. These physiological mechanisms lead to a reduction in the quantities of most pathogenic microbes in the gut (Frimpong et al., 2021). The use of rice-IMOs as probiotics in pig feeding is advantageous because of the high rate of survival in the gastrointestinal tract. This could depict that they are capable of resisting the digestion of gastric acid in order to interact with the microorganisms native to the host gut. Also, rice-IMOs meet production requirements, such as suitability for large-scale production, stable shelf storage, and good sensory characteristics. It has no toxic effects on the host and is not pathogenic (Gaggia et al., 2010; Zhange et al., 2023); thus, all these could support the use of rice-IMOs for health improvement in pig production.

The physiological effects of nutrition in an animal's body are often shown by serum biochemical markers. In tandem with the current study, Dong et al. (2013) reported that dietary probiotics (*Lactobacillus plantarum* and *B. subtilis*) in weaned pigs resulted in significant ( $p < 0.05$ ) effects in total protein and globulin, although there was no effect in albumin, urea, and ALP. Also, Cao et al. (2019) reported that in comparison with the control, probiotic-treated piglets exhibited significantly lowered serum ALT and AST, similar to this study. However, in another animal study using weaned pigs, Li et al. (2021) showed that *Bacillus subtilis* had no significant ( $p > 0.05$ ) effect on albumin index. The ranges of mean serological indices reported in this study corroborate those reported by Sanubi et al. (2023).

Increased concentrations of serum ALT and AST indicate hepatocyte leakage and are sensitive markers of liver damage (Lala et al., 2023). A study found that in weaned pigs, probiotics containing *B. subtilis* and *Enterococcus faecium* did not significantly affect the serum profiles of ALT, AST, and ALP, indicating a low hepatic risk (Liu et al., 2018). In the present study, the findings demonstrated a considerable reduction in the levels of serum ALT and AST in the IMOs-treated groups. This suggests that the liver of weaner pigs was not negatively impacted by IMOs treatment. Probiotics such as IMOs are thought to support liver health through a variety of biological mechanisms, including increased insulin sensitivity, decreased endotoxin levels, decreased glucose and low-density lipoprotein cholesterol absorptions, decreased oxidative stress, and decreased inflammatory markers (Manzhalii et al., 2017; Ghavami et al., 2018).

Physiologically, synthesis of vitamin D, hormones, and other compounds depends on cholesterol, which is also necessary for the formation of the cell membrane and other cell structures (Huff et al., 2023). Increased cholesterol levels may cause many negative consequences and health challenges, such as inflammatory pathway activation, which can damage tissue and elevate the probability of infections and inflammatory diseases (Tall & Yvan-Charvet, 2015). Furthermore, the findings in this study showed a significant decline in cholesterol index in the IMOs-treated pigs. This result is in agreement with the report by Aminlari et al. (2019). The potential physiological mechanisms that could be involved in this reduction include bile salt deconjugation by bile salt hydrolase, coprecipitation of cholesterol with deconjugated bile, cholesterol assimilation by growing cells, cholesterol binding to the microbial cellular surface, the binding action of bile by fiber, and the colon's microbial production of short-chain fatty acids (Haroun et al., 2013).

The findings on serum total protein and globulin do not agree with the increased outcome reported by Dong et al. (2013) on the effect of dietary probiotics on the serum profile of weaned piglets. However, their values were within the normal ranges in pigs (total protein: 8.07-8.12 g/dL and globulin: 3.34-4.52 g/dL) according to Adegun et al. (2023). Additionally, Kim et al. (2021) reported improved liver function with respect to a reduction in ALP and urea when pigs were fed multi-probiotics *Lactobacillus*. These outcomes in ALP and urea are not similar to the findings in the current study. These discrepancies in findings could be ascribed to variations in the breed or strain of the animal, genotype of the animal, nutrition, duration of the study, physiological state of the animal, or other environmental factors (Phillip, 2016; Bushby et al., 2018; Cantalapedra-Hijar et al., 2018).

## 5. Conclusions

In conclusion, the increasing demand for animal products has encouraged the use of probiotics as alternatives to improve livestock performance. Although antibiotics have historically been used as growth promoters, their application is associated with adverse effects on animal and human health, particularly the contribution to antimicrobial resistance. Probiotics, such as rice-IMOs (10 mL/L), could be a safer alternative because they

improved some haematological and biochemical indicators. More research is required to assess the effects of rice-IMOs on farm animal performance, as we recommend (10 mL IMOs per liter of water), their usage in pigs and other farm animals. At this point, this study stands to favour animal farmers, animal scientists, veterinarians, consumers, researchers, academicians, etc.

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## 7. Authors' Contributions

*Ndubuisi Samuel Machebe*: conceptualization, supervision, project administration, and funding acquisition. *Luke Chukwudi Ali*: manuscript writing and editing. *Bright Chigozie Amaefule*: data organization. *Nnanna Ephraim Ikeh*: data validation and formal analysis. *Cynthia Ifechukwu Emmanuel*: results and outcome reasoning. *Amarachi Linda Obinna, Paulinus Ikenna Umeugokwe, and Onwuamaeze Gabriel Ugwu*: methodology.

## 8. Conflicts of Interest

No conflicts of interest.

## 9. Ethics Approval

Yes applicable. This study was approved by the Animal Welfare and Ethics Committee, Department of Animal Science, University of Nigeria, Nsukka (Approval Number: UNN/CO24ARO13.28.05.2024).

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**Informed Consent Statement**

Not applicable.

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