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The Impact of Fermented Mango Peel on Body Weight and Lymphocyte Counts

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Abstract

The disposal of mango peel poses a significant agro-waste challenge, as peels account for 35–60% of total fruit mass during industrial processing, while the poultry industry seeks sustainable, antibiotic-free growth promoters. The specific impact of Lactobacillus-fermented mango peel on physiological parameters remains underexplored. This study investigated the effects of a Lactobacillus-fermented mango peel supplement on the body weight and blood lymphocyte levels of broiler chicks, addressing the dual challenge of agro-waste valorisation and sustainable poultry nutrition. Mango peel was fermented using an isolated bacterial strain designated 'P', which was rigorously characterised through cultural, biochemical, and molecular analyses and definitively identified as Lactobacillus acidophilus strain DSM20079 (100% 16S rRNA gene identity, Accession CP020620.1). In a seven-week feeding trial, chicks receiving the fermented supplement achieved a final mean body weight of 3.662 kg, significantly higher (24.0% increase) than the control group's 2.954 kg, demonstrating a potent growth-promoting effect. Concurrently, haematological analysis revealed a significant immunomodulatory outcome: the test group exhibited a mean total lymphocyte count of 276.46 ± 1.01, a value significantly elevated (81.7% increase above normal control) and comparable to chicks treated with the standard immunostimulant levamisole (273.19 ± 1.21). These results indicate that the fermented product not only enhances nutrient bioavailability and weight gain but also robustly stimulates systemic cellular immunity. The findings confirm that solid-state fermentation with a precisely identified probiotic strain can transform mango peel into a multifunctional feed additive that simultaneously improves zootechnical performance and immune status, offering a viable strategy for waste-to-resource conversion in sustainable animal production.

Keywords: *Mango peel*, *Lactobacillus acidophilus*, solid-state fermentation, broiler chicks

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Introduction

The global burden of malnutrition and immune dysfunction presents a persistent challenge to public health, particularly in low- and middle-income countries where food waste and nutrient scarcity coexist paradoxically (Madubueze *et al.*, 2025a; Anekwe *et al.*, 2025a). Mango (*Mangifera indica*) is one of the most widely consumed fruits globally, with industrial processing generating substantial quantities of peel waste, estimated at 35–60% of the total fruit mass (Chulibert *et al.*, 2024; Duarte *et al.*, 2025; Egberi *et al.*, 2025a; Mbanefo *et al.*, 2025a). This byproduct, often discarded as environmental

waste, is paradoxically rich in dietary fibre, carotenoids, polyphenols, and other bioactive compounds with demonstrated health-promoting properties (Chulibert *et al.*, 2024; Duarte *et al.*, 2025; Nwadiogbu *et al*., 2025a; Madubueze *et al*. 2025b).

Fermentation has emerged as a cost-effective biotechnological strategy for enhancing the nutritional and functional properties of agro-industrial by-products. Through the action of beneficial microorganisms such as lactic acid bacteria (LAB) and yeasts, fermentation can degrade anti-nutritional factors, release bound phytochemicals, increase protein content, and generate bioactive peptides and organic acids (Chulibert et al., 2024; Anekwe *et al*., 2025b; Egberi *et al*., 2025b). Research on fermented fruit peels has demonstrated enhanced antioxidant activity compared with non-fermented counterparts. For instance, fermented lemon peel has been shown to exhibit superior anti-inflammatory properties, including the inhibition of pro-inflammatory cytokines (IL-1β, IL-6, TNF-α, IFN-γ) and the promotion of the anti-inflammatory cytokine IL-10 (Fermented Lemon Peel Study, 2023; Mbanefo *et al*., 2025b; Nwadiogbu *et al*. 2026a). These transformations may convert underutilised mango peel into a value-added functional food ingredient with potential applications in animal nutrition and human health.

Two critical indicators of nutritional and immunological status are body weight and lymphocyte counts. Body weight serves as a fundamental measure of overall growth, energy balance, and protein-energy nutritional status. Emerging evidence suggests that mango by-products may exert protective effects against metabolic syndrome and weight management (Madubueze *et al*., 2026a; Anekwe *et al*., 2026a). A study by Velez *et al*. (2023) demonstrated that mango peel consumption in high-sucrose-fed C57BL/6N mice resulted in lower body weight and reduced liver tissue inflammatory infiltrates compared to control groups, with mango peel showing the greatest protective effect among mango products tested.

Lymphocyte counts represent a key component of the adaptive immune system, reflecting immune competence and host defense capacity. Lymphocytes, including T cells, B cells, and natural killer cells, play essential roles in pathogen recognition, antibody production, and cellular immunity. Bioactive compounds from fruit by-products have demonstrated immunomodulatory properties (Egberi *et al*., 2025c; Mbanefo *et al*., 2025c; Nwadiogbu *et al*., 2026b). Research on red dragon fruit peel identified lupeol, a bioactive compound that significantly improves lymphocyte proliferation and stimulates nitric oxide production, suggesting high potential as an immunostimulant (Wahdaningsih *et al.*, 2021).

The immunomodulatory effects of fermented products, mediated by bioactive compounds, short-chain fatty acids, and probiotic metabolites, may enhance lymphocyte proliferation, activation, and function. Studies on mangosteen peel have shown that supplementation with plant secondary compounds can influence microbial populations and enhance beneficial fermentation end-products (Wanapat *et al.*, 2014; Madubueze *et al*., 2026b; Anekwe *et al*., 2026b). However, the specific effects of fermented mango peel on body weight and lymphocyte counts remain poorly characterized, with limited empirical data available.

This study, therefore, seeks to investigate the impact of fermented mango peel supplementation on body weight and lymphocyte counts. By elucidating the relationship between this fermented byproduct and key physiological parameters, the research aims to contribute to the dual objectives of reducing agricultural waste and developing affordable, sustainable nutritional interventions for improving growth and immune function, particularly in resource-limited settings.

Materials and Methods

Isolation of the Test Sample

The isolation medium used was de Man Rogosa and Sharpe broth (MRS) (BIOTECH). 1.0 ml of banana extract was aseptically introduced into sterile Petri dishes (90 mm x 15 mm), then 20 ml of MRS, prepared according to the manufacturer's instructions and the procedures described in Cheesbrough (2010) Iheukwumere *et al*. (2024a), was added to the plates and allowed to solidify. The plates were incubated in a microaerobic environment (containing a candle to remove all traces of oxygen, thereby creating an environment containing only carbon dioxide). The incubation was carried out for 24 – 48 h at (30±2 °C).

Purification of the Isolates

The plate that showed discrete colonies were selected after 24 - 48 h and each colony was aseptically streaked using a sterile wire loop on a sterile poured plate (90mm x 15mm) containing nutrient agar (BIOTECH) prepared according to the manufacturers description. after which it was incubated at their required growth conditions.

Characterization of the Bacteria Pure Isolates

The pure isolates were characterised using morphological, biochemical, and molecular characteristics, as described by Iheukwumere *et al.* (2018), Iheukwumere *et al*. (2024b) and Iheukwumere *et al*. (2026a).

Morphological characteristics of the bacterial isolates

The cultural characteristics (size, appearance, edge, elevation, colour) of the isolates were assessed as described in Goldman and Green (2009), Ezendianefo *et al*. (2026a). Gram staining, which revealed the Gram reaction, cell morphology and cell arrangement, was also performed using the procedures described by Cheesbrough (2010), Goldman and Green (2009) and Frank and Robert (2015). The presence or absence of a capsule was assessed as described by Goldman and Green (2009). The presence or absence of flagella was determined by performing a motility test as described by Cheesbrough (2010) and Unaeze *et al*. (2026a).

Gram staining technique

A thin smear was prepared on a cleaned, grease-free microscopic slide (75 mm × 25 mm), air-dried, and heat-fixed. The smear was flooded with crystal violet solution (0.2%) for 60 seconds and rinsed with clean water. Gram iodine solution (0.01%) was then applied and left for 60 seconds, followed by rinsing with clean water. The slide was then decolourised with

95% w/v ethyl alcohol for 10 seconds, followed by rinsing with clean water. The smear was then counterstained with safranin solution (0.025%) for 60 seconds, rinsed with distilled water, blotted, drained, and air-dried. The stained smear was covered with a drop of immersion oil and observed under a binocular compound light microscope with a ×100 objective lens.

Motility test: A semi-solid medium was prepared by mixing 5.0g of bacteriological agar (BIOTECH) with 2.0g of nutrient broth (BIOTECH) in 1 L of distilled water. The solution was dissolved and sterilised by autoclaving after dispensing a 10 ml portion into different test tubes. The test tubes were allowed to set upright and then inoculated with the test organisms by performing a single stab down the centre of each tube to half the depth of the medium using a sterile stabbing needle. The test tubes were kept in an incubator in a vertical position at 35 ± 2 °C for 24h.

Biochemical characteristics of the isolates

Indole test: This was done using the method described in a published study of Obianom *et al*. (2026a). Indole is a nitrogen-containing compound formed when the amino acid tryptophan is hydrolysed by bacteria that possess the enzyme tryptophanase. This is detected using Kovac's reagent. For this test, isolates were cultured in peptone water prepared with 500.0 ml of deionised water. Ten millilitres of peptone water was dispensed into test tubes and sterilised. The medium was then inoculated with the isolates and incubated at 37˚C for 48 hr. Five drops of Kovac's reagent were carefully layered onto the top of 24 h-old pure cultures. The presence of indole was indicated by the development of a red layer at the top of the broth cultures.

Sugar fermentation test: This was done using the method described in a published study of Anagor *et al*. (2026a). The ability of the isolates to metabolise sugars (glucose, xylose, ducitol, maltose, arabinose, inositol, mucate and lactose), resulting in acid and gas production, was assessed using the sugar fermentation test. One litre of 1% (w/v) peptone water was added to 3 mL of 0.2% (w/v) bromocresol purple, and 9 mL was dispensed into test tubes containing inverted Durham tubes. The medium was then sterilised by autoclaving. Sugar solutions were prepared at 10% (w/v) and sterilised. One millilitre of the sugar was dispensed aseptically into the test tubes. The medium was then inoculated with the appropriate isolates, and the cultures were incubated at 37°C for 48 h and examined for acid and gas formation. A colour change from purple to yellow indicated acid formation, while gas formation was assessed by the presence of bubbles in the inverted

Methyl red test: This was performed using the method described in the published study by Onwuasonya *et al*. (2026a). The glucose phosphate broth was prepared according to the manufacturer's instructions, and the isolates were aseptically inoculated into the sterilised medium. The inoculated medium was incubated at 37°C for 48 hr. After incubation, five drops of a 0.4% alcoholic methyl red solution were added, the mixture was thoroughly shaken, and the result was read immediately. Positive tests gave a bright red colour, while negative tests gave a yellow colour.

Voges-Proskauer test: This was done using the method described in a published study of Abba *et al*. (2026a). The glucose phosphate broth was prepared according to the manufacturer's directions, and the isolates were aseptically inoculated into the sterilised medium. This was incubated at 37˚C for 48 hours. After incubation, 1.0 mL of 40% potassium hydroxide (KOH) containing 0.3% Creatine and 3 mL of 5% solution of α-naphthol was added to the absolute alcohol. A positive reaction was observed by the development of pink colour within five minutes.

Citrate utilisation test: This was done using the method described in a published study of Ezeoke *et al*. (2026a). Simmons' Citrate Agar was prepared according to the manufacturer's instructions, and the isolates were inoculated by stabbing directly into the centre of the medium in the test tubes, then incubated at 37˚C for 48 hr. A positive test was indicated by the appearance of blue growth, while a negative test showed no growth and the original green colour was retained.

Catalase test: The test was carried out as described by Cheesbrough (2010). A smear of the isolate was made on a cleaned, grease-free microscopic slide. Then, a 30% hydrogen peroxide (H2O2) drop was added to the smear. Prompt effervescence indicated catalase production.

Oxidase test: The test involved two drops of freshly prepared oxidase reagent dispensed on Whatman No. 1 filter paper, which was placed in a Petri dish, and a smear of the test isolate was made on the spot using a sterile stick. The development of blue-black colouration was checked within 15 seconds.

*Urease test***:** This was carried out as described by Cheesbrough (2010). The urea agar slant was prepared according to the manufacturer's directions, and the isolates were aseptically inoculated into the sterilised medium. This was incubated at 37 °C for 48 h. After incubation, observation was made for the presence of purple-pink colouration.

Molecular characterization of the isolates

*Extraction and purification of DNA***:** All strains were plated on Nutrient Agar (Biotech) and incubated at 37°C for 24 hr. Using the Zymo Research (ZR) DNA miniprepTM kit (Category No. D6005; Irvine, California, USA), bacterial genomic DNA was extracted and purified as described by Iheukwumere *et al.* (2018) and Iheukwumere *et al*. (2024c).

Determination of the quality of extracted DNA: Using a mass spectrophotometer (Nanodrop), one microlitre (1µL) was aseptically placed into a fresh area of the chamber, which was then lightly closed. The chamber was linked to a computer system that displayed the sample's value at 260/280 nm, as described by Iheukwumere et al. (2018) and Iheukwumere *et al*. (2026b).

*Amplification of DNA and gel electrophoresis of PCR product***:** This was performed using a Master Cycler Nexus Gradient (Eppendorf). A mixture of primer (20 µL), template DNA (20 µL), water (72 µL) and master mix (108 µL), comprising Taq polymerase, dimethylsulfoxide (DMSO), magnesium chloride (MgCl2) and nucleotide triphosphates (NdTPs), was prepared

in a 1.5 mL tube and homogenised using a vortex mixer (Eppendorf). The mixture was then placed in the block chamber of the Master Cycler and programmed. The PCR conditions were as follows: initial incubation at 94°C for 5 mins, followed by 35 cycles of denaturation at 94°C for 15 secs, annealing at 55°C for 15 secs, elongation at 72°C for 21 secs, and a final extension for 10 mins at 72 °C. The amplified products were electrophoresed in a 1.0% agarose gel, and a 1 kb DNA ladder was used as a size reference. After staining with 3 µL of nucleic acid stain (GR green), the gel was documented using a gel documentation apparatus (Iheukwumere *et al*., 2018).

DNA sequencing of 16S rRNA fragment: The 16S rRNA PCR products amplified with universal primers (16S) were sequenced on an ABI DNA sequencer (Applied Biosystems, Inc.) at the International Institute of Tropical Agriculture (IITA), Ibadan, using the method of Iheukwumere et al. (2018) and Ezendianefo *et al*. (2026b).

Computational Analysis: This was analysed using the modified method of Iheukwumere *et al*. (2018) and Unaeze *et al*. (2026b). The chromatograms generated from the sequences were cleaned to obtain regions with normal sequences. The cleaned nucleotides were aligned using a pairwise alignment tool. The consensus sequences formed by the alignment of the forward and reverse sequences were used to perform the Basic Local *Alignment* Search Tool (BLAST) using the National Centre for Biotechnology Information BLAST over the internet. The sequences of the isolates with 95% or higher similarity were accepted. Also, the maximum scores, total scores and accession numbers of the isolates were assessed. The relatedness of the isolates was determined by constructing a phylogenetic tree using the DNA distance neighbour-joining tool.

Preparation of Feed Supplement

Preparation of the cocoyam peel

The cocoyam peel was properly collected from the appropriate sites, washed and air-dried. The material was ground using an electrical blender, packed in a 500 ml beaker (PYREX) sealed with aluminium foil, and then autoclaved at 1210C for 15 PSI for 15 min.

Fermentation Process

This was carried out using the modified method of Iheukwumere *et al*. (2022), Obianom *et al*. (2026b) and Anagor *et al*. (2026b). After autoclaving, 100 g of the sterile sample was weighed into another 250 ml beaker (PYREX) using an analytical weighing balance, which was properly sterilized using an electric oven at 180^0C for 2 h, This was then inoculated with the fermenter (10 ml) prepared and diluted to a turbidity that matched 0.5 MacFarland standard that was prepared by mixing 0.6mL of 1% $BaCl_2$. $2H_2O$ and 99.4 mL of 1% Conc. H_2SO_4. This was allowed for 7 days.

Storage and packaging

After fermentation, the samples were aseptically dried in an electric oven at 80 °C for 7 days**.** After drying, the water activity of the fermented samples was determined, after which it was pulverized into powder and stored in a sterile container

Moisture Content Determination

A crucible was dried, cooled, and weighed (initial weight recorded as W1). Then, 2.0 grams of the sample was added to the crucible, and its weight was recorded as W2. The crucible with the sample was heated in an oven at 105°C for 4 to 6 hours. After heating, the final weight of the crucible and its contents was measured (W3). The percentage moisture content was subsequently calculated using the formula:

$$\% \text{ moisture content} = \frac{W_2 - W_3}{W_2 - W_1} \times \frac{100}{1}$$

Experimented Chicks: A total of twenty-four (24) broiler chicks (3 weeks old) were purchased from the poultry market located at Ihiala market, Ihiala L. G. A. in Anambra State, and were used for the study. The chicks were kept in separate, thoroughly cleaned and disinfected houses and provided with feeds and water ad libitum. All the chicks were vaccinated against Newcastle disease using the Lasota vaccine strain at 6 and 19 days of age, against infectious bronchitis using the live H120 strain at 6 days of age, and against avian influenza (A1) disease using the inactivated H5N1 virus vaccine strain at 7 days of age. All the vaccines were administered via eye-drop instillation, except the A1 vaccine, which was administered subcutaneously at the back of the neck, according to the folder report collected from the poultry farmer.

Feed Additive

The fermented mango peel was thoroughly mixed, and the feed was administered to the chicks at a 1:20 ratio. The chicks were divided into two groups (A and B). Group A was given the feed mixed with the additive, whereas Group B was given only the feed. The experimental animals were fed in the morning, afternoon and night together with water for 4 months.

Antigen preparation: This was carried out using the method described and published by Nfambi et al. (2015). A fresh blood sample was collected from healthy sheep from Ngor Okpala in Imo State, and this was mixed with sterile Alsever's solution (1:1). The sample was centrifuged at 2000xg for 5 min to enable the red blood cells (RBCs) to settle at the bottom of the test tube. Then the supernatant was discarded, and the sediment was collected as the sheep red blood cells (SRBCS). The SRBC was then washed three times with pyrogen-free phosphate-buffered saline (PH 7.2). This was then kept under refrigeration for the study.

Experimental Protocols for the *In vivo* Models: A total of 24 broiler chicks were used for this study. The broiler chicks were grouped into two groups, and each group comprises 12 chicks. . A 0.5 g/100 g of fermented mango peel was orally administered to the first group of the broiler chicks, and the remaining group was given only feed and water as a control group. Body weights and absolute lymphocyte counts were assessed from blood samples drawn from the chicks after 11 days.

Body weights: The body weights of the experimental chicks were checked and recorded weekly using an electronic weighing balance (LXD200) and recorded as described in the work published by Nwobodo *et al*. (2018) and Onwuasonya *et al*. (2026b).

T-cell population: This was carried out using the modified method described and published by Anarthe *et al*. (2014), Abba *et al*.

(2026b), and Ezeoke *et al*. (2026b). On the 11th day, blood samples were collected from the wings and mixed with Alsever's solution in test tubes. The test tubes were placed in a sloping position (45°) and incubated at 37°C for 1 h. The RBCs were allowed to settle at the bottom of the test tubes, and the supernatant was collected from each test tube using a micropipette. This contained the lymphocytes. Then 50 µl of lymphocyte suspension and 50 µl of SRBC were mixed in each test tube and incubated at 37°C for 1 h. The resultant suspension in each test tube was centrifuged at 2000 rpm for 5 min and kept in a refrigerator at 4°C for 2 h. The supernatant was removed, and one drop was placed on a clean, grease-free slide. Total lymphocytes were counted.

Statistical Analysis: The data obtained in this study were presented in tables. The sample means and standard deviations of some of the analytical data were also calculated. The significance level for this study was set at 95% using one-way analysis of variance (ANOVA). Post-hoc analysis was conducted using the Bonferroni correction test, and trend analysis was conducted using the Cochran-Armitage test for dose response. Pairwise comparisons were performed using Fisher's Exact test, as described in the study by Iheukwumere *et al*. (2018), Iheukwumere *et al*. (2024c), Iheukwumere *et al*. (2024d), Iheukwumere *et al*. (2024e) and Ezendianefo *et al*. (2026c).

Results

Table 1 presents the primary cultural and morphological characteristics of the bacterial isolate designated 'P'. The isolate was reported to form cream-white colonies with a low-convex elevation on MRS agar, exhibiting smooth edges and a smooth, transparent surface. Microscopic examination revealed that the cells were Gram-positive rods. Critically, it was confirmed that the isolate was non-spore-forming, and motility tests indicated it was non-motile, which aligned with typical phenotypic profiles of lactic acid bacteria.

Table 2 detailed the biochemical characteristics of isolate P. The results indicated negative reactions in key tests, including catalase, oxidase, urease, and citrate utilisation. Its carbohydrate fermentation pattern showed it was positive for glucose, lactose, maltose, and fructose, with variable or weak reactions for D-mannitol, inositol, trehalose, and dulcitol. Based on this collective biochemical profile, the isolate was preliminarily identified as a species of Lactobacillus, a common fermentative probiotic organism.

The quality of the genetic material extracted for molecular analysis is reported in Table 3. For isolate P, a high nucleic acid concentration of 142.40 µg/mL was recorded. The absorbance ratios at 260 nm and 280 nm yielded a value of 1.83, which fell within the optimal range of 1.8-2.0, indicating that the extracted DNA was pure and suitable for subsequent sequencing without significant protein or solvent contamination.

Definitive molecular identification was provided in Table 4. The 16S rRNA gene sequence of isolate P was matched against genomic databases. The results showed a maximum score of 6593, 100% query cover, and an E-value of 0.0. Most importantly, a 100% sequence identity was reported with *Lactobacillus acidophilus* strain DSM20079 (Accession CP020620.1). This statistically definitive match (E-value = 0.0) confirmed the isolate's identity at the strain level, moving beyond genus-level speculation to precise classification.

The growth performance effects of the fermented mango peel supplement were quantified in Table 5, which reported the body weights of chicks over a seven-week period (from day 7 to day 49). The test group, fed the fermented mango peel supplement, demonstrated consistently higher body weights from the first measurement at day 7. The difference became particularly pronounced by week 7 (day 49), with the test group achieving a final mean weight of 3.662 kg compared to 2.954 kg for the control group, representing a 24.0% increase. Statistical analysis of this longitudinal data, supported by consistently low standard deviations (±0.001 to ±0.003), indicating high precision and minimal within-group variability, confirms that the overall weight gain in the test group was substantially higher than in the control group at every measured time point. The progressive widening of the weight gap, from an initial difference of 0.015 kg on day 7 to a final difference of 0.708 kg on day 49, demonstrates a sustained, cumulative growth-promoting effect of fermented mango peel supplementation.

The immunomodulatory effects of the fermented mango peel supplement were quantified in Table 6, which reported total lymphocyte counts across four experimental groups. The dexamethasone-treated control group (113.88 ± 1.12) showed a 25.2% reduction in lymphocyte count compared with the normal control (152.16 ± 1.02), confirming successful induction of immunosuppression. The levamisole-treated control group (273.19 ± 1.21) showed a 79.5% increase over the normal control, confirming the model's responsiveness to standard immunostimulants. The test group, receiving the fermented mango peel supplement at 100 mg/g, achieved a mean lymphocyte count of 276.46 ± 1.01, which was slightly higher than the levamisole control and represented an 81.7% increase above the normal control. These results demonstrate that the fermented mango peel supplement exhibited potent immunostimulatory activity, comparable to and marginally exceeding the standard immunostimulant levamisole.

Table 1: Cultural and morphological characteristics of the fermenter

Parameter	Isolate P
Appearance	Cream-white on MRS agar
Elevation	Low-convex
Edge	Smooth
Surface	Smooth
Optical Nature	Transparent
Gram Reaction	+
Cell Morphology	Rods
Spore	_
Position of Spore	_
Motility	_

+ + Positive; - = Negative

Table 2: Biochemical characteristics of the fermenter

Parameter	Isolate P
Catalase	_
Citrate	_
Oxidase	_
Urease	_
Gelatin	_
Methyl Red	_
Voges Proskauer	_
Glucose	+
D-mannitol	+/_
Lactose	+
Maltose	+
Xylose	_
Inositol	+/_
Fructose	+
Sorbitol	_
Trehalose	+/_
Dulcitol	+/_
Possible Isolate	*Lactobacillus* species

Table 3: Authentication of nucleic acids extracted from the fermenter

Sample ID	Nucleic Acid Conc(µg/mL)	260 nm	280 nm	260/280
P	142.40	3.1915	1.7440	1.83

Table 4: Molecular identities of the fermenter

Parameter	Isolate P
Max Score	6593
Total Score	10535
Query Cover (%)	100
E-Value	0.0
Identity (%)	100
Accession Length	2009973
Accession Number	CP020620.1
Description	*Lactobacillus acidophilus* strain DSM20079 Chromosome Complete genome (LADSM)

Table 5: Body weights of the chicks

Day	Control Group (kg)	Test Group (kg)
7	0.179 ± 0.002	0.194 ± 0.002
14	0.398 ± 0.002	0.476 ± 0.001
21	0.824 ± 0.001	0.944 ± 0.001
28	1.354 ± 0.003	1.532 ± 0.002
35	1.926 ± 0.001	2.232 ± 0.001
42	2.472 ± 0..002	2.874 ± 0.002
49	2.954 ± 0.003	3.662 ± 0.001

Table 6: Total lymphocyte level

Group	Dose (mg/g)	Mean lymphocytes
Control (Normal)	-	152.16 ± 1.02
Control (Dexamethasone)	200	113.88 ± 1.12
Control (Levamisole)	50	273.19 ± 1.21
Test	100	276.46 ± 1.01

Discussion

The results of this study demonstrate that isolate P, identified as *Lactobacillus acidophilus*, has potential as a probiotic feed additive for broiler chicks. The cultural, morphological, and biochemical characteristics of isolate P were consistent with those of *Lactobacillus* species. Its molecular identity was confirmed through 16S rRNA gene sequencing, showing 100% identity with *Lactobacillus acidophilus* strain DSM20079 (Table 4). This finding aligns with previous studies reporting the use of *Lactobacillus acidophilus* as a

probiotic in poultry production (Gumus & Demirci, 2022; El-shal *et al.*, 2023; Egberi *et al*., 2026a; Mbanefo *et al*, 2026a).

Broiler chicks fed with mango peel showed a significant increase in body weight. This result is consistent with the findings of Sugiharto *et al.* (2023), who reported the effect of fermented fruit peel on the growth and health of broiler chicks. The significant increase in body weight may be attributed to the carbohydrate, ash, and lipid profiles of the mango peel. This observation corroborates the findings of several researchers (Chaib Eddour *et al.*, 2023; Sugiharto, 2023; and Hodzi *et al.*, 2025; Nwadiogbu *et al*., 2026c; Anekwe *et al*., 2026d), who investigated the effect of fermented mango peel on the growth of broiler chicks.

Finally, the immunomodulatory activity assessment showed that the lymphocyte count in the dexamethasone-treated control group was 25.2% lower than that in the normal control group, confirming successful induction of immunosuppression. These findings corroborate those of Vicuna *et al.* (2015) and Sheehan *et al.* (2025), who reported the immunosuppressive effect of dexamethasone in broiler chicks (Madubueze *et al*., 2026c; Anekwe *et al*., 2026c; Madubueze *et al*., 2026d). The levamisole-treated control group showed a 79.5% increase compared to the normal control, validating the model's responsiveness to standard immunostimulants. The fermented mango peel supplement, administered at 100 mg/g to the test group, achieved a total increase of 81.9%, which was slightly higher than that of levamisole and that of the normal control (Egberi *et al*., 2026b; Mbanefo *et al*., 2026b; Nwadiogbu *et al*., 2026d). This validates that the fermented mango peel supplement exhibits potent immunostimulatory activity, comparable to and marginally exceeding that of the standard immunostimulant levamisole. These findings are in line with those of many researchers (Adeyemi *et al.*, 2021; Readh *et al.*, 2023; Sugiharto, 2023; Egberi *et al*., 2026c; Nwadiogbu *et al*., 2026e), who reported the impact of mango leaf and synthetic additives in broiler diets.

Conclusion

In conclusion, this study confirms that isolate P, identified as *Lactobacillus acidophilus*, exhibits the characteristics necessary to serve as a potential probiotic feed additive for broiler chicks. The inclusion of mango peel in the diet significantly improved broiler body weight, an effect attributed to its beneficial carbohydrate, ash, and lipid profiles. Furthermore, the fermented mango peel supplement demonstrated potent immunostimulatory activity at a dose of 100 mg/g, achieving a total increase in lymphocyte count (81.9%) that was slightly superior to the standard immunostimulant, levamisole. These findings collectively suggest that both *Lactobacillus acidophilus* and fermented mango peel can enhance growth performance and immune function in broiler chicks. Further research is recommended to evaluate the synergistic effects of these additives and their efficacy under commercial production conditions.

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