IPS Journal of Biotechnology and Applied Biochemistry
ISSN: 3115-5707
IPS J. Biotechnol. Appl. Biochem., 2(1): 101-111 (2026)
DOI: https://doi.org/10.54117/ijbab.v2i1.126

Modulation of Short-Chain Fatty Acid Level of Chicks Gut using *Bacillus subtilis* Fermented Corn and Bean Chaff

Abba, O.[1], Iheukwumere, I. H.[2], Iheukwumere, C. M.[3], Ike, V. E.[4], Mere, C. A.[5], Ezendianefo, J. N.[6], Okongwu, D. J.[7], Nnagbo, P. A.[8] and Ofuani, A. G.[9]

[1]Department of Microbiology, Federal University of Gusau, Zamfara State.
[2]Department of Microbiology, Faculty of Natural Sciences, Chukwuemeka Odumegwu Ojukwu University, Uli, Campus, Anambra State, Nigeria.
[3]Department of Applied Microbiology and Brewing, Faculty of Biosciences, Nnamdi Azikiwe University, Awka, Nigeria.
[4]Department of Microbiology, University of Agriculture and Environmental Sciences, Umuagwo, Imo State, Nigeria.
[5]Department of Biochemistry, Chukwuemeka Odumegwu Ojukwu University, Uli Campus.
[6]Department of Microbiology, Tansian Universiy, Umunya, Anambra State, Nigeria.
[7]Department of Chemistry, Nwafor Orizu College of Education, Nsugbe.
[8]Department of Microbiology, Imo State University, Owerri.
[9]Department of Science Laboratory Technology, Delta State Polytechnic, Ogwashi-Uku, Delta State, Nigeria.

Email: oluchiipan@fufusau.edu.ng / ik.iheukwumere@coou.edu.ng

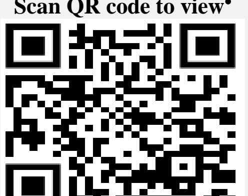
Abstract

The poultry industry faces challenges in optimizing gut health and productivity. Short-chain fatty acids (SCFAs) play a crucial role in gut health, but modulation strategies are limited. Limited studies have explored the impact of *Bacillus subtilis* fermented corn and bean chaff on SCFA levels in the chicks' gut, necessitating an investigation into its potential as a gut health modulator. This study was carried out to evaluate the effect of *Bacillus subtilis* fermented corn and bean chaff on the modulation of short-chain fatty acid (SCFA) levels in the gut of chicks, and to investigate its potential as a gut health modulator. The fermenter used in this study was obtained following the standard microbiological techniques. Corn and bean chaff were collected, processed, homogenized and fermented using solid state fermentation method, and this was incorporated as a feed additive and assessed for the impact on SCFAs of chicks' gut using an *in vivo* technique. The fermenter, identified as *Bacillus subtilis* strain PK5-17, was characterized culturally, morphologically, and biochemically and to the molecular level The study showed significant increase ($p<0.05$) in acetate (69.65% vs 66.22%), propionate (26.11% vs 22.40%), and n-valerate (3.94% vs 3.03%) levels in the test group compared to the control group. The n-butyrate level was similar between groups ($p>0.05$). The study concluded that *Bacillus subtilis* fermented corn and bean chaff modulates SCFA levels in the gut of chicks, suggesting its potential as a gut health modulator.

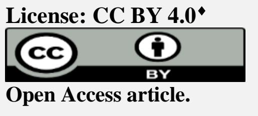
Keywords: *Bacillus subtilis*, Fermented, Short-chain fatty acids, Gut health, Chicks

Article History
Received: 29 Dec 2025
Accepted: 02 Feb 2026
Published: 10 Feb 2026

Scan QR code to view•



License: CC BY 4.0♦



Open Access article.

How to cite this paper: Abba, O., Iheukwumere, I. H., Iheukwumere, C. M., Ike, V. E., Mere, C. A., Ezendianefo, J. N., Okongwu, D. J., Nnagbo, P. A., & Ofuani, A. G. (2026). Modulation of Short-Chain Fatty Acid Level of Chicks Gut using Bacillus subtilis Fermented Corn and Bean Chaff. *IPS Journal of Biotechnology and Applied Biochemistry*, 2(1), 101–111. https://doi.org/10.54117/ijbab.v2i1.126

INTRODUCTION

The use of natural feed to grow chicken has been shown to be of high benefit in terms of quality of meat and health impact to the consumers (Awad *et al.,* 2017; Iheukwumere *et al*., 2025a; Dim *et al*., 2025a). Short-chain fatty acid is an essential component found in the gut of living organisms (Awad *et al.,* 2017). This compound contains mainly fat and acid and confers enormous benefits to the organism. The fat and acid components appear in chains, which do not elongate. The roles of short-chain fatty acids involve regulating the integrity of the gut, stabilizing immune function for warding off pathogens, and minimizing inflammation (Latorre *et al*., 2018). Poor production of short-chain fatty acids in the gut leads to destabilization of body function in both animals and humans, due to loss of coordination (Gharib-Naseri *et al*., 2019; Iheukwumere *et al*., 2022a; and Nwike *et al*., 2017).

It is worthy to not that short-chain fatty acid is produced naturally in the gut by normal flora, which are vital bacterial species that are capable of digesting dietary fibre (Papatsiros *et al*., 2013). Digestion of dietary fibre by the normal flora of the gut releases short-chain fatty acids as byproducts such as butyrate, propionate, and acetate (Correa-Oleveira *et al*.,

2016). This indicates that enhancing short-chain fatty acid production in both animals and humans is essential.

Research has revealed that *Bacillus* species are capable of enhancing the production of short-chain fatty acids in animals such as chicks (Dittoe *et al*., 2018; Ekechukwu *et al.,* 2025a; Obianom *et al*., 2024; Dim *et al*., 2025b). *Bacillus* species are lactic acid bacteria that are capable of fermenting dietary fibre, which resists the action of digestive enzymes (Papatsiros *et al.,* 2013; Amadi *et al*., 2017; Ejike *et al*., 2017). Naturally, indigestible fibre can be found in both plants and animals such as bark of maize and beans chaff. The action of the lactic acid and other metabolites that are produced by *Bacillus* species facilitates the breakdown of the indigestible substances to release short chain fatty acids (Khan and Iqbal, 2016). Regulation of the intestinal mucosa of birds is highly essential to optimum growth and productivity (Timbermont *et al*., 2010) and the product enhances absorption of nutrients and stimulation of vibrant immune response in chicks (Stefanello *et al.,* 2020**).**

Several researchers have worked on the role of organic acid in modulating the integrity of short chain fatty acid such as Awad *et al*. (2017), Latorre *et al*. (2018), and Gharib-Naseri *et al*. (2019), Iheukwumere *et al*. (2025b), Dim *et al*. (2025c) but few studies are available on modulation of short chain fatty acid level of chicks gut using *Bacillus subtilis*-fermented corn and beans chaff. Hence, the aim of this study is to evaluate modulation of short chain fatty acid level of chicks gut using *Bacillus subtilis*-fermented corn and beans chaff.

MATERIALS AND METHODS

Isolation of the Test Sample

The media used for this isolation was de Man Rogosa and Sharpe broth (MRS) (BIOTECH). A 1.0 ml of fermented yoghurt (Aqua yoghurt) and banana extract were aseptically introduced into sterile Petri dishes (90 mm x 15 mm), then 20 ml of MRS which was prepared according to the manufacturers instruction and the procedures described in Cheesbrough (2010), Ekechukwu *et al*. (2025b), Ekesiobi *et al*., (2025), Ezedianafo *et al*., (2025a) was added into the plates, allowed to solidified. The plates were incubated in a microaerophic environment (containing candle used to evacuate all traces of oxygen thereby creating an environment having only carbon IV oxide). The incubation was done for 24 – 48 h at (30±2^{0}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 as described by Iheukwumere *et al*. (2020a), Ezedianafo *et al*. (2025b); Idigo *et al*. (2025a), Iheukwumere *et al*. (2025c).

Characterization of the Bacteria Pure Isolates

The pure isolates were characterized using the morphological, biochemical and molecular characteristics as described by Iheukwumere *et al.* (2017a); Iheukwumere *et al.* (2018a), Ike *et al*. (2025a), Iheukwumere *et al*. (2025d).

Morphological characteristics of the Bacteria isolates

The cultural descriptions (size, appearance, edge, elevation, colour) of the isolates were carried out as described in Goldman and Green (2009); Iheukwumere *et al*. (2017b), Iheukwumere *et al*. (2018b), Iheukwumere *et al*. (2020b). The Gram staining technique which revealed the Gram reaction, cell morphology and cell arrangement were also carried out using the procedure described by Cheesbrough (2010), Goldman and Green (2009) Frank and Robert (2015), Iheukwumere *et al*. (2022b), Iheukwumere *et al*. (2023a). The presence or absence of capsule was also carried out as described by Goldman and Green (2009), Ike *et al*. (2025b), Obiefuna *et al*. (2025a). The presence or absence of flagellum was determined by carrying out motility test as described by Cheesbrough (2010), Iheukwumere *et al*., (2017c), Iheukwumere *et al*. (2018c), Iheukwumere and Iheukwumere (2022a).

Gram staining technique

A thin smear was made in a cleaned grease free microscopic slide (75mm×25mm), air dried heat fixed. The smear was flooded with crystal violet solution (0.2%) for 60 seconds and rinsed with cleaned water. Gram iodine solution (0.01%) was then applied and allowed for 60 seconds. This was rinsed with cleaned water. This was followed by decolourizing the slide content with 95%w/v ethyl alcohol for 10seconds and then rinsed with cleaned water. The smear was then counter stained with safranin solution (0.025%) for 60 seconds, rinsed with cleaned water, blot drained and air dried. The stained smear was covered with a drop of immersion oil and observed under a binocular compound light microscope using × 100 objective lens as described by Iheukwumere *et al*. (2017d); Iheukwumere *et al*. (2020c), Chude *et al*. (2020), Iheukwumere and Iheukwumere (2022b), Iheukwumere *et al*. (2022c).

Motility test: A semi-solid medium prepared by mixing 5.0g of bacteriological agar (BIOTECH) with 2.0g of nutrient broth (BIOTECH) in 1 Litre of distilled water was used. The solution was dissolved and sterilized using autoclaving technique after dispensing 10 ml portion in different test tubes. The test tubes were allowed to set in vertical positions and then inoculate the test organisms by performing a single stab down the centre of the test tube to half the depth of the medium using sterile stabbing needle. The test tubes were kept in an incubator in vertical position at 35±2^{0}C for 24h (Iheukwumere *et al*., 2017e; Iheukwumere and Iheukwumere, 2022c; Iheukwumere *et al*., 2022d; Idigo *et al*., 2025b).

Biochemical characteristics of the isolates

Indole test: Indole is a nitrogen containing compound formed when the amino acid tryptophan is hydrolyzed by bacteria that have the enzyme tryptophanase. This is detected by using KOVAC's reagent. For this test, isolates were cultured in peptone water in 500.0 ml of deionized water. Ten millilitres of peptone water was dispensed into the test tubes and sterilized. The medium was then inoculated with the isolates and kept in an incubator 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 revealed by the development of red layer colouration on the top of the broth cultures as described by Iheukwumere *et al*. (2022e),

Iheukwumere and Iheukwumere (2022d), Iheukwumere *et al.* (2023b), Egbe *et al.* (2025a), Ike *et al.* (2025c).

Sugar fermentation test: The capability of the isolates to metabolize some sugars (glucose, xylose, ducitol, maltose, arabinose, inositol, mucate and lactose) with the resulting formation of acid and gas or either were carried out using 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 in the test tube that contained inverted Durham tubes. The medium was then sterilized by autoclaving. The sugar solution were prepared at 10% (w/v) and sterilized. One milliliter of the sugar was dispensed aseptically into the test tubes. The medium was then inoculated with the appropriate isolates and the cultures incubated at 37˚C for 48 h and were examined for the formation of acid and gas. Change in colour from purple to yellow indicated acid formation while gas formation was assessed by the presence of bubbles in the inverted (Iheukwumere *et al.*, 2022f; Iheukwumere and Iheukwumere, 2022e; Egbe *et al.,* 2025b; Idigo *et al.*, 2025c).

Methyl red test: The glucose phosphate broth was prepared according to the manufacturer's direction and the isolates were aseptically inoculated into the sterilized medium. This was incubated at 37˚C for 48 hr. After incubation, five drops of 0.4 % solution of alcoholic methyl red solution was added and mixed thoroughly, and the result was read immediately. Positive tests gave bright red colour while negative tests gave yellow colour (Ezedianafo *et al.*, 2025c; Ike *et al.*, 2025c).

Voges-Proskauer test: The glucose phosphate broth was prepared in accordance to the manufacturer's direction and the isolates were aseptically inoculated into the sterilized medium. This was incubated at 37˚C for 48hr. After incubation, 1.0 mL of 40% potassium hydroxide (KOH) containing 0.3% Creatine and 3 ml of 5% solution of *α*-naphthol was add ed in the absolute alcohol. Positive reaction was observed by the development of pink colour within five minutes (Egbe *et al.*, 2025b; Ekechukwu *et al.*, 2025c).

Citrate utilization test: The Simmon's Citrate Agar was prepare according to the manufacturer's direction and the isolates were inoculated by stabbing directly at the center of the medium in the test tubes and incubated at 37˚C for 48 hr. Positive test was shown by the appearance of growth with blue colour, while negative test showed no growth and the original green colour was retained (Idigo *et al.*, 2025d; Ezedianafo *et al.* 2025d).

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 drop of 30% hydrogen peroxide (H_2O_2) was added on the smear. Prompt effervescence indicated catalase production (Idigo *et al.*, 2025e; Idigo *et al.*, 2025f).

Oxidase test: The test involved two drops of freshly prepared oxidase reagent dispensed on Whatman No. 1 filter paper which was placed in 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), Idigo *et al.* (2025g) and Idigo *et al.* (2025h). The urea agar slant was prepared in accordance to the manufacturer's direction and the isolates were aseptically inoculated into sterilized medium. This was incubated at 37^0C 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. By means of the procedures of Zymo Research (ZR) DNA miniprepTM kit, bacterial genomic DNA was then extracted and purified (Category No. D6005; Irvine, California, USA) as described by Iheukwumere *et al.* (2018) Iheukwumere *et al.* (2025e; Idigo *et al.*, 2025h).

Determination of the quality of extracted DNA: Using mass spectrophotometer (Nanodrop), One micro litre (1µL) was aseptically dropped into a fresh space in the chamber and the chamber was lightly closed which was then linked to a computer system which showed the window that discovered the value of the sample at 260/280nm as described by (Iheukwumere *et al.,* 2018; Iheukwumere *et al.*, 2025f; Idigo *et al.*, 2025i).

Amplification of DNA and gel electrophoresis of PCR product: This was analysed using Master cycler Nexus Gradient (Eppendorf). A mixture of primer (20 µL), template DNA (20µL), water (72 µL) and master mix (108 µL), which comprises taq polymerase, dimethylsulfoxide (DMSO), magnesium chloride ($MgCl_2$) and nucleotides triphosphates (NdTPs), was made in 1.5 mL tube and homogenized using vortex mixer (Eppendorf). This was then positioned in the block chamber of the master cycler and then programmed. The PCR program for 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 final extension period for 10 mins at 72°C. The amplified products were electrophorezed in 1.0% agarose gel and a1kb DNA ladder was used as a size reference. After staining with 3µL of nucleic acid stain (GR green), the gel was documented with gel documentation apparatus (Iheukwumere *et al.,* 2018; Iheukwumere *et al.*, 2025g; Idigo *et al.*, 2025j; Idigo *et al.*, 2025k).

DNA sequencing of 16s rRNA fragment: The 16S rRNA amplified PCR products generated from universal primer (16S), was used for the sequencing using ABI DNA sequencer (Applied Biosystem Inc) at International Institute of Tropical Agriculture (IITA), Ibadan using the method of Iheukwumere *et al.* (2018), Iheukwumere *et al.*, (2025h), and Idigo *et al.* (2025l), Idigo *et al.*, (2025m).

Computational Analysis: This was analysed making use of the modified method of Iheukwumere *et al.* (2018), Iheukwumere *et al.* (2025i), Idigo *et al.* (2025n), Iheukwumere *et al.*, (2025j). The chromatograms generated from the sequences were cleaned to obtain regions with normal sequences. The cleaned nucleotides were aligned using pair wise 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 National Centre for Biotechnology Information BLAST over the internet. The sequences of the isolates with 95% and above similarities were accepted. Also the maximum scores, total scores and accession numbers of the isolates were assessed. The relatedness of the isolates was determined by tracing their phylogenetic tree using DNA distance neighbour phylogenetic tree tool.

Preparation of Feed Supplement

Preparation of the corn and bean chaff

The corn and bean chaff was properly collected from the appropriate sites, washed and air dried. The material was ground using an electrical blender, packed in 500 ml beaker (PYREX) sealed with aluminium foil and then autoclave at 121^{0}C for 15 min at 15psi.

Fermentation Process

This was carried out using the modified method of Iheukwumere *et al* (2022), Iheukwumere *et al.* (2025k), Iheukwumere *et al*. (2025l). After autoclaving, a 100 g of the sterile sample was weighed into another 250 ml beaker (PYREX) using analytical weighing balance, which was properly sterilized using electric oven at 180^{0}C 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 fermented samples were aseptically dried using an electric oven at 80^{0}C for 7days**.** After drying 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 (final weight recorded as 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 poultry market located at Ihiala market, Ihiala L. G. A. in Anambra State were used for the study. The chicks were kept in separate, thoroughly cleaned and disinfected house and provided with feeds and water ad libitum. All the chicks were vaccinated against Newcastle disease using Lasota vaccine strains at 6 and 19 days of age, against infectious bronchitis using live H120 strain at 6 days old and also against avian influenza (A1) disease using inactivated H5N1virus vaccine strain at 7 days old. All the vaccines were given via eye drop instillation except (A1) vaccine, which was given through the subcutaneous route at the back of the neck from the folder report collected from the poultry farmer.

Feed Additive

The fermented groundnut chaff was mixed with fish meal and the feed in a ratio of 1:20. This mixture was properly and thoroughly mixed and administered to the chicks. 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.

Experimental Protocols for the *In vivo* Models: A total of 36 broiler chicks were used for this study. The broiler chicks were grouped into six groups, and each group comprises 6 chicks. . A 0.5 g/100 g of fermented corn mixed with fish meal was orally administered to each of group of broiler chicks, and the remaining group was giving only feed and water as control group. The body weights and blood absolute lymphocytes were assessed from the blood samples drawn from the chicks after 11 days.

Short chain fatty acids (SCFAs): This was carried out using gas chromatographic (GC) technique as published by AOAC (2019), Iheukwumere *et al.* (2025m), Iheukwumere *et al.* (2025n). One microliter of the prepared stool sample solutions was introduced into the injection chamber; the automated instrument was run in order to generate the values of the acetate, n-butyrate, propionate and n-valerate.

Statistical Analysis: The data obtained in this study were presented in tables and figures. Their percentages were also calculated. The sample means and standard deviations of some of the analytical data were also calculated. The significance of this study was determined at 95% using one way analysis of variance (ANOVA). Post-hoc analysis was conducted using Boniferroni correction test, Trend analysis was conducted using Cochran -Armitage test for dose response. Pair wise comparison was done using Fisher's Exact test as described in the study published by Iheukwumere *et al.* (2018), Idigo *et al.*, (2025o), Idigo *et al.* (2025p), Idigo *et al.* (2025q), Idigo *et al.* (2025r), Idigo *et al.* (2025s), Idigo *et al.* (2025t), Manasseh *et al.* (2025).

RESULTS

The cultural and morphological characteristics of the fermenter is shown in Table 1. The results revealed that isolate Q appeared white on Nutrient agar with a flat elevation, irregular edge and a rough surface. The isolate was a gram-positive cocci bacillus and a spore former with a central positioning of the spore. The biochemical characteristics of the fermenter are shown in Table 2. The results revealed that isolate Q was positive to catalase, citrate, oxidase, gelatin, and Voges-Proskauer, while negative to urease and methyl red. The isolate showed complete utilization of glucose, D-mannitol, maltose, xylose, inositol, fructose, sorbitol, trehalose and dulcitol while unable to utilize lactose. The nucleic acid extracted from the fermenter revealed that it was deoxyribonucleic acid (DNA) at the ratio of 260nm/280nm with a range of 1.80-1.81, as shown in Table 3.The molecular characteristics of the fermenter revealed the presence of the *Bacillus subtilis* strain PK5-17 chromosome with a complete genome (BSPK5) as shown in Table 4.

The modulation of short-chain fatty acid using *Bacillus subtilis* fermented corn and bean chaff and feather fed to the test group showed an increase in the level of short-chain fatty acid, whereby acetate recorded the highest, followed by propionate, N-valerate and n-butyrate, and these effects were statistically significant (P<0.05) as shown in Table 5.

Table 1: Cultural and morphological characteristics of the fermenters

Parameter	Isolate Q
Appearance	Cream/white on Nutrient agar
Elevation	Flat
Edge	Irregular
Surface	Rough
Optical Nature	Opaque
Gram Reaction	+
Cell Morphology	Rods with round ends
Spore	+
Position of Spore	Central
Motility	+

+ + Positive; - = Negative

Table 2: Biochemical characteristics of the fermenter

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

Table 3: Authentication of nucleic acids extracted from the fermenter

Sample ID	Nucleic Acid Conc(µg/mL)	260 nm	280 nm	260/280
Q	126.20		1.7220	1.81

Table 4: Molecular identities of the fermenter

Parameter	Isolate Q
Max Score	4783
Total Score	4783
Query Cover (%)	100
E-Value	0.0
Identity (%)	100
Accession Length	4086149
Accession Number	CP026037.1
Description	*Bacillus subtilis* strain PK5-17 Chromosome Complete genome (BSPK5)

Table 5: Short-chain fatty acids values

Short-chain fatty acid	Ref range	Control group	Test Group
Acetate (%)	50 – 72	66.22 ± 0.11	69.65 ± 0.22
n-butyrate (mg/L)	0.80 – 1.00	2.06 ± 0.01	2.07 ± 0.01
Propionate (%)	15.40 – 30.30	22.40 ± 0.16	26.11 ± 0.11
N-valerate (%)	0.80 – 5.00	3.03 ± 0.01	3.94 ± 0.01

DISCUSSION

Bacillus subtilis is the type species for the genus *Bacillus* which are gram-positive, spore-forming bacilli which are capable of producing enzymes which break down proteins, starch and lipids into amino acids, sugars, fatty acids and glycerol (Bayasal and Yildiz, 2019). Their ability to break down proteins, starch and lipids into amino acids, sugars, fatty acids and glycerol supports gut health and boosts the immune response, has gained significant insight (Kovacs, 2019). This present study focuses on the modulation of short-chain fatty acid levels using *Bacillus subtilis* fermented chicken feather in broiler chicks. The cultural, morphological and biochemical characteristics of the *Bacillus subtilis* corroborated with the findings of many researchers (Das and Prasad, 2010; Vijayalakshmi *et al*., 2019; Baysal and Yildiz, 2019) who isolated and characterized *Bacillus subtilis* from banana. The ability of the isolates to utilize common sugars such as glucose, lactose, sucrose, maltose and some alcoholic sugars such as mannitol, sorbitol and xylitol indicates their potential to produce beneficial metabolites which support gut health and aid food fermentation. These potentials were also described by many researchers (Das and Prasad, 2010; Vijayalakshmi *et al*., 2019; Baysal and Yildiz, 2019). The molecular characterization of the *Bacillus subtilis* strain revealed the presence of *Bacillus subtilis* strain PK5-17 (LADSM). The occurrence of *Bacillus subtilis* strain in banana had been reported by Haroun (2011) and Ragavi *et al.* (2019), who stated that the occurrence may be due to the enzyme richness of the banana.

The results obtained from the study revealed that *Bacillus subtilis* fermented corn and bean chaff and chicken feather significantly increased the short-chain fatty acid levels such as acetate, n-butyrate, propionate and N-valerate. These findings were in line with the findings of most researchers (Omar *et al*., 2021; Chang, 2022; and Zhang *et al*., 2023) who investigated the effect of *Bacillus subtilis* fermented meal on the modulation of short-chain fatty acid levels in broiler chicks.

CONCLUSION

The study has shown that *Bacillus subtilis* fermented corn and bean chaff exhibited modulate the level of short-chain fatty acid in the gut of broiler chicks. Hence could be used as additive for healthy chicks.

Acknowledgment

We are grateful to all our study participants who join the study voluntarily. We are grateful to ZAHARM Analytical and Research Laboratory, Amawbia, Awka Anambra State, Nigeria for providing enabling environment, resources and techniques for this study. We really salute their wonderful efforts.

Conflict of interests: The authors declare that they have no conflict of interests.

Funding: This research did not receive specific grant from any funding agencies.

REFERENCES

Alahyaribeik, S., Nazarpour, M., Tabandeh, F., Honarbakhsh, S., & Sharifi, S. D. (2022). Effects of bioactive peptides derived from feather keratin on plasma cholesterol level, lipid oxidation of meat, and performance of broiler chicks. *Tropical Animal Health and Production*, *54*(5), 271.

Amadi, R.E., Iheukwumere, I.H. and Unaeze, B.C. (2017). Effects Of Crude Alkaloid Extracted From Ocimum Gratissimum On The Activity Of Ciprofloxacin Against Salmonella Enterica Serovar Typhi. *Advances in Life Science and Technology* 58.

Anahtar, M. N., Byrne, E. H., Doherty, K. E., Bowman, B. A., Yamamoto, H. S., Soumillon, M., ... and Kwon, D. S. (2018). Cervicovaginal bacteria are a major modulator of host inflammatory responses in the female genital tract. *Immunity, 42*(5), 965-976. https://doi.org/10.1016/j.immuni.2015.04.019

Awad, W.A., Hess, C. and Hess M. (2017). Enteric pathogens and their toxin-induced disruption of the intestinal barrier through alteration of tight junctions in chickens. *Toxins* 9:60 – 66

Balogu, T. V., Ibrahim, H., & Balogu, D. O. (2017). Nutritionally improved corn mill waste (chaff) with microbial protein: An economic alternative for poultry feed. *Advances in Applied Sciences*, *2*(2), 18-22.

Baysal, O., & Yildiz, A. (2017). Bacillus subtilis: an industrially important microbe for enzymes production. *EC Microbiol*, *5*(4), 148-156.

Bell, V., Ferrão, J., & Fernandes, T. (2017). Nutritional guidelines and fermented food frameworks. *Foods, 6*(8), 65. https://doi.org/10.3390/foods6080065

Cabel, M. C., Goodwin, T. L., & Waldroup, P. W. (1988). Feather meal as a nonspecific nitrogen source for abdominal fat reduction in broilers during the finishing period. *Poultry Science*, *67*(2), 300-306.

Cai, Y., Li, Y., Zhang, Y., & Wang, Y. (2019). The role of *Lactobacillus* in immune regulation: Current status and perspectives. *Frontiers in Immunology, 10*, 1804. https://doi.org/10.3389/fimmu.2019.01804

Chachaj, R., Sembratowicz, I., Krauze, M., & Ognik, K. (2019). The effect of partial replacement of soybean meal with fermented soybean meal on chicken performance and immune status. *Journal of Animal and Feed Sciences*, *28*(3), 263-271.

Chang, W. Y., & Yu, Y. H. (2022). Effect of Bacillus species–fermented products and essential oils on growth performance, gut morphology, cecal short-chain fatty acid levels, and microbiota community in broilers. *Poultry Science*, *101*(8), 101970.

Chen, M., Sun, Q., & Giovannucci, E. (2021). Dairy consumption and risk of type 2 diabetes: The role of fermented dairy products. *American Journal of Clinical Nutrition, 113*(5), 1100–1111. https://doi.org/10.1093/ajcn/nqaa397

Chude,C.O., Iheukwumere, I.H., Iheukwumere, C.M., Nwaolisa,C.N., Egbuna, C.,Nwakoby, N.E.and Egbe, P.A. (2020). Cidal activity of proteins secreted by *Bacillus thuringensis* against *Ascaris lumbricoides. International Journal of Research Publications* 49(1): 1033 – 1045.

Corrêa-Oliveira, R., Fachi, J.L., Vieira, A., Sato, F.T. and Vinolo, M.A.R. (2016). Regulation of immune cell function by short-chain fatty acids. *Clinical Translational Immunology* 5:73 – 80

Cristofori, F., Dargenio, V. N., Dargenio, C., Miniello, V. L., Barone, M., & Francavilla, R. (2021). Anti-inflammatory and immunomodulatory effects of probiotics in gut inflammation: A door to the future. *Frontiers in Immunology,* 12, 578386. https://doi.org/10.3389/fimmu.2021.578386

Das, G., & Prasad, M. P. (2010). Isolation, purification & mass production of protease enzyme from Bacillus subtilis. *Int. Res. J. Microbiol*, *1*(2), 26-31.

Dim, C. N., Iheukwumere, I. H., Iheukwumere, C. M., Ugwu, C. H., Ike, V. E., Ezendianefo, . J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., Oduenyi, P. M., & Ochibulu, S. C. (2025a). Multiple Antibiotic Resistance Bacterial Strains in Frozen Meat Sold at Abagana, Anambra State: A Public Health Concern. *IPS Journal of Applied Microbiology and Biotechnology*, *4*(3), 181–186. https://doi.org/10.54117/ijamb.v4i3.75

Dim, C. N., Iheukwumere, I. H., Iheukwumere, C. M., Ugwu, C. H., Ike, V. E., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., Oduenyi, P. M., & Ochibulu, S. C. (2025b). The Burden of Antibiotic Resistance: Evaluating the Impact of Multiple Antibiotic-Resistant Enteric Bacteria in Academic Environments. *IPS Interdisciplinary Journal of Biological Sciences*, *4*(4), 144–149. https://doi.org/10.54117/iijbs.v4i4.78

Dim, C. N., Iheukwumere, I. H., Iheukwumere, C. M., Ugwu, C. H., Ike, V. E., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., Oduenyi, P. M., & Ochibulu, S. C. (2025c). Antimicrobial resistance in aquaculture: evaluating pseudomonas aeruginosa from fish ponds. *IPS Intelligentsia Multidisciplinary Journal*, *4*(1), 32–36. https://doi.org/10.54117/iimj.v4i1.10

Dimidi, E., Cox, S. R., Rossi, M., & Whelan, K. (2019). Fermented foods: Definitions and characteristics, impact on the gut microbiota and effects on gastrointestinal health and disease. *Nutrients, 11*(8), 1806. https://doi.org/10.3390/nu11081806

Dittoe, D.K., Ricke, S.C., Kiess, A.S. (2018). Organic acids and potential for modifying the avian gastrointestinal tract and reducing pathogens and disease. *Frontiers in Veterinary Science* 5:216 – 223

Egbe, P. A., Umeaku, C. N., Iheukwumere, I. H., Iheukwumere, C. M., Onwuasoanya, U. F., Ezenwata, I. S., Afulukwe, S. C., Ike, V. E., & Ezeumeh, E. N. (2025a). Antibiotic Susceptibility of Helicobacter pylori Isolates from Patients at Nnewi Teaching Hospital, Anambra State. *IPS Journal of Basic and Clinical Medicine*, 2(2), 51–57. https://doi.org/10.54117/ijbcm.v2i2.11.

Egbe, P. A., Umeaku, C. N., Iheukwumere, I. H., Iheukwumere, C. M., Onwuasoanya, U. F., Ezenwata, I. S., Afulukwe, S. C., Ike, V. E., Ezeumeh, E. N., & Egbuna, C. (2025b). Helicobacter pylori Inhibition by Medicinal Plant Extracts: An In Vitro Assessment. IPS Journal of Drug Discovery Research and Reviews, 3(1), 32–37. https://doi.org/10.54117/ijddrr.v3i1.28.

Egbe, P. A., Umeaku, C. N., Iheukwumere, I. H., Iheukwumere, C. M., Onwuasoanya, U. F., Ezenwata, I. S., Afulukwe, S. C., Ike, V. E., & Ezeumeh, E. N. (2025c). Medicinal Plant Extracts Enhance Conventional Antibiotic Activity against Helicobacter pylori: An In Vitro Assessment. IPS Interdisciplinary Journal of Biological Sciences, 4(2), 93–99. https://doi.org/10.54117/iijbs.v4i2.51.

Ejike, C.E., Iheukwumere, I.H. and Armadi, R.E. (2017). Susceptibility of Escherichia coli Isolated from Oligospermia Patient to *Gongronema latifolium* leaves extract. J. *Biol. Agriculture. Healthcare* 7(14).

Ekechukwu, C. C., Umeh, S. O., Iheukwumere, I. H., & Iheukwumere, C. M. (2025a). Bacterial Loads of Smoked Fish and Chicken: Role of pH and Moisture Content. *IPS Applied Journal of Nutrition, Food and Metabolism Science*, 3(1), 44–49. https://doi.org/10.54117/iajnfms.v3i1.102.

Ekechukwu, C. C., Umeh, S. O., Iheukwumere, I. H., & Iheukwumere, C. M. (2025b). Biological Inhibition of Pathogenic Bacteria Isolated from Smoked Fish and Chicken: An In Vitro Study. IPS Interdisciplinary Journal of Biological Sciences, 4(2), 85–92. https://doi.org/10.54117/iijbs.v4i2.50.

Ekechukwu, C. C., Umeh, S. O., Iheukwumere, I. H., & Iheukwumere, C. M. (2025c). Prophylactic Potential of the Most Potent Synergistic Biological Agent against Bacterial Infections from Smoked Fish and Chicken. IPS Journal of Applied Microbiology and Biotechnology, 4(2), 153–160. https://doi.org/10.54117/ijamb.v4i2.57.

Ekesiobi, A. O., Iheukwumere, C. M., Iheukwumere, I. H., Ejike, C. E., Ilechukwu, C. C., Ike, V. E., Okereke, F. O., & Ochibulu, S. C. (2025). Hyping the Inhibitory Activity of Xylopia aethiopica against Vibrio cholerae using Azithromycin. IPS Journal of Basic and Clinical Medicine, 2(3), 93–98. https://doi.org/10.54117/ijbcm.v2i3.16

Ezedianafo, J. N., Iheukwumere, I. H., Iheukwumere, C. M., Okolo, O., Nwike, I., & Ubajekwe, C. C. (2025a). *Musca domestica*: A vector of multidrug-resistant enteric bacteria. *Journal of Veterinary, Allied, and One Health Sciences, 1*(2), 30–38. https://doi.org/10.54117/3vwg0p36

Ezedianafo, J. N., Iheukwumere, I. H., Iheukwumere, C. M., Okolo, O., Nwike, I., & Ubajekwe, C. C. (2025b). Occurrences of meropenem- and imipenem-resistant *Klebsiella pneumoniae* in *Musca domestica* in hospital landfills. *African Journal of Applied Research & Sustainable Development, 1*(2), 25–35. https://doi.org/10.54117/wjmnyv91

Ezendianefor, J. N., Iheukwumere, I. H., Iheukwumere, C. M., Okolo, O., Nwike, I., & Ubajekwe, C. C. (2025). Multiple antibiotic resistance indices of enteric bacteria isolated from *Musca domestica*. *Journal of Public Health, Policy, and Society, 1*(2), 29–37. https://doi.org/10.54117/k8r78723

Ezendianefor, J. N., Iheukwumere, I. H., Iheukwumere, C. M., Okolo, O., Nwike, I., & Ubajekwe, C. C. (2025). *Klebsiella pneumoniae* isolated from *Musca domestica*: Antibiotic susceptibility and resistance patterns. *Journal of Veterinary, Allied, and One Health Sciences, 1*(2), 39–47. https://doi.org/10.54117/vy6y8f94

Ezendianefor, J. N., Iheukwumere, I. H., Iheukwumere, C. M., Okolo, O., Nwike, I., & Ubajekwe, C. C. (2025). *Musca domestica* as vectors of pathogenic enteric bacteria: A public health concern. *African Journal of Applied Research & Sustainable Development, 1*(2), 36–45. https://doi.org/10.54117/s671mk28

Frias, J., Peñas, E., & Martínez-Villaluenga, C. (2017). Fermented foods in health promotion and disease prevention. In V. R. Preedy & R. R. Watson (Eds.), *Fermented Foods in Health and Disease Prevention* (pp. 3–20). Academic Press.

Gharib-Naseri, K., Dorigam, J.C., Doranalli, K., Morgan, N., Swick, R.A. and Choct M. (2021). *Bacillus amyloliquefaciens* CECT 5940 improves performance and gut function in broilers fed different levels of protein and/or under necrotic enteritis challenge. *Animal Nutrition* 7:185–197

Gharib-Naseri, K., Kheravii, S., Keerqin, C., Morgan, N., Swick, R. and Choct, M. (2019). Two different Clostridium perfringens strains produce different levels of necrotic enteritis in broiler chickens. *Poultry Science* 98:6422–6432

Guo, Q., Goldenberg, J. Z., Humphrey, C., El Dib, R., Johnston, B. C., & Lo, C. K. F. (2020). Probiotics for the prevention of pediatric antibiotic-associated diarrhea. *Cochrane Database of Systematic Reviews, 4*(4), CD004827. https://doi.org/10.1002/14651858.CD004827.pub5

Guo, S., Zhang, Y., Cheng, Q., Xv, J., Hou, Y., Wu, X., Du, E., & Ding, B. (2020). Partial Substitution of Fermented Soybean Meal for Soybean Meal Influences the Carcass Traits and Meat Quality of Broiler Chickens. *Animals*, *10*(2), 225. https://doi.org/10.3390/ani10020225

Haroun, M. (2011). Isolation, identification and molecular characterization of a potential probiotic bacterium, Bacillus subtilis PPP 13 from Penaeus monodon.

Hill, C., Guarner, F., Reid, G., Gibson, G. R., Merenstein, D. J., Pot, B., ... and Sanders, M. E. (2014). Expert consensus document: The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. *Nature Reviews Gastroenterology and Hepatology, 11*(8), 506-514. https://doi.org/10.1038/nrgastro.2014.66

Husnain, A., Anwar, U., Fatima, A., Mustafa, R., Farooq, U., Abbas, W., Khalid, M. F., Ashraf, M., & Aziz ur Rahman, M. (2025). Effects of replacement of soybean meal with fermented soybean meal on growth performance, nutrient digestibility and carcass characteristics in broiler. *Livestock Science*, *299*, 105779. https://doi.org/10.1016/j.livsci.2025.105779

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Bacterial symbionts of *Rhynchophorus phoenicis*: Modulation of rat lymphocyte function and immunity. *Journal of Tropical Medicine and Public Health Solutions, 1*(2), 55–63. https://doi.org/10.54117/jtmphs.v1i2.36

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Multidrug-resistant *Klebsiella pneumoniae* in *Musca domestica*: A potential public health threat. *Journal of Tropical Medicine and Public Health Solutions, 1*(2), 46–54. https://doi.org/10.54117/jtmphs.v1i2.35

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Termite-derived antimicrobials: A novel approach to control *Bacillus cereus* in food products. *International Journal of Global Trends and Research, 1*(2), 45–57. https://doi.org/10.54117/2mgwn270

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Bacterial diversity in insect guts and their potential applications in industry. *International Journal of Global Trends and Research, 1*(2), 36–44. https://doi.org/10.54117/2k0d9t18

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Bioactive compounds from *Macrotermes*: A novel approach to combat multidrug resistant *Klebsiella pneumoniae* from *Musca domestica* in hospital landfills. *IPS Journal of Biotechnology and Applied Biochemistry, 1*(2), 82–92. https://doi.org/10.54117/ijbab.v1i2.111

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Assessment of blood indices in rats supplemented with Macrotermes-derived nutrient-enhanced bacteria. *IPS Journal of Basic and Clinical Medicine, 2*(4), 143–151. https://doi.org/10.54117/ijbcm.v2i4.39

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Exploring the potential of termite gut bacteria as growth promoters and organ function enhancers in albino Wistar rats. *Health Science Research International, 1*(2), 43–52. https://doi.org/10.54117/hsri.v1i2.38

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Assessment of hematological parameters in albino Wistar rats fed with *Rhynchophorus phoenicis* larvae enriched with autochthonous bacteria. *Health Science Research International, 1*(2), 34–42. https://doi.org/10.54117/hsri.v1i2.37

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Entomopathogenic bacteria-mediated management of *Delia radicum*: A step towards eco-friendly pest control. *Journal of Agriculture, Food Technology and Sustainability, 2*(2), 112–121. https://doi.org/10.54117/jafts.v2i2.110

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Antimicrobial activity of Macrotermes-derived eluates against multidrug resistant Pseudomonas species: Implications for aquaculture disease management. *Journal of Agriculture, Food Technology and Sustainability, 2*(2), 102–111. https://doi.org/10.54117/jafts.v2i2.109

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Exploring the potential of entomopathogenic bacteria for sustainable management of *Sitophilus zeamais* in maize storage systems. *IPS Journal of Plant, Animal, and Environmental Sciences, 1*(1), 11–20. https://doi.org/10.54117/ijpae.v1i1.108

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Biological control of *Acanthoscelides obteus* using entomopathogenic bacteria. *IPS Journal of Plant, Animal, and Environmental Sciences, 1*(1), 1–10. https://doi.org/10.54117/ijpae.v1i1.107

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Modulation of phagocytic index in albino Wistar rats via bacterial symbionts from *Rhynchophorus phoenicis* larvae. *African Journal of Nutrition and Applied Research, 1*(2), 27–36. https://doi.org/10.54117/fm3vgt16

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Bacterial symbionts of insect larvae: A novel approach to improving micronutrient content. *African Journal of Nutrition and Applied Research, 1*(2), 27–36. https://doi.org/10.54117/960k2266

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Unlocking the potential of termite gut microbiome: Enhancing nutritional value through bacterial symbionts. *IPS Journal of Nutrition and Food Science, 5*(1), 636–645. https://doi.org/10.54117/ae6gj081

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Enhancing nutritional parameters with bacterial symbionts from Macrotermes species: A potential frontier in nutritional biotechnology. *IPS Journal of Nutrition and Food Science, 5*(1), 625–635. https://doi.org/10.54117/a2b7jb52

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Bacterial symbionts of Macrotermes species: Assessing their impact on phagocytic indices of albino Wistar rats. *IPS Interdisciplinary Journal of Biological Sciences, 5*(1), 187–196. https://doi.org/10.54117/iijbs.v5i1.106

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Bacterial symbionts of insects: Exploring their role in insect nutritional composition. *IPS Interdisciplinary Journal of Biological Sciences, 5*(1), 177–186. https://doi.org/10.54117/iijbs.v5i1.105

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Augmenting rat lymphocyte function by bacterial symbiont of Macrotermes species. *IPS Journal of Applied Microbiology and Biotechnology, 5*(1), 281–290. https://doi.org/10.54117/ijamb.v5i1.104

Idigo, M. A., Iheukwumere, I. H., Iheukwumere, C. M., Nnaeze, B. C., Akulue, C. J., Nwakoby, N. E., Ezendianefor, J. N., Ike, V. E., Nnaedozie, A. O., Ezekwueche, S. N., Anagor, I. S., Aniekwe, C. C., Ezeoke, F. C., Okereke, F. O., & Ochibulu, S. C. (2025). Antimicrobial peptides from insects: A study on their efficacy against pathogens. *IPS Journal of Applied Microbiology and Biotechnology, 5*(1), 271–280. https://doi.org/10.54117/ijamb.v5i1.103

Iheukwumere, C. M., Iheukwumere, I. H., Nwakoby, N. E., Idigo, M. A., & Ike, V. E. (2025m). Evaluation of fermented chicken feather meal as a dietary supplement on rat lipid metabolism and immune response. *African Journal of Nutrition and Applied Research, 1*(1), 17–26. https://doi.org/10.54117/qbwbpp15

Iheukwumere, I. H., Nwankwo, A. K., Iheukwumere, C. M., Okorie, N. A., Nwakoby, N. E.,. Ekesiobi, A. O. and Okolo, O. C.. (2025n). Characterization and pathogenic profile of *Aspergillud fumigatus* isolated from lendfills. *Microbes and Infectious Diseases* 6(4)

Iheukwumere,I.H., Iheukwumere,C.M., Chude, C.O., Nwaolisa,C.N. and Egbe, P.A. (2020a). Comparative study of different clinical samples used for the diagnosis of staphylococcal systemic infections in appearent healthy students. *International Journal of Research Publications* 49(1): 1 – 10

Iheukwumere, C. M., & Iheukwumere, I. H. (2022a). Nutritive and Antinutrient Values of Soybean Condiments Produced from Indigenous Fermenters. *IPS Applied Journal of Nutrition, Food and Metabolism Science*, 1(1): 1-5. https://doi.org/10.54117/iajnfms.v1i1.8

Iheukwumere, I.H., Iheukwumere, M.C. and Nwakoby, N.E. (2022b). Synergistic Effects of Probiotics and Autogenous Bacterin against Salmonella enterica Serovar Typhimurium Strain U288. IPS Journal of Nutrition and Food Science, 1(1), 1–5. https://doi.org/10.54117/ijnfs.v1i1.3.

Iheukwumere, I.H. and Iheukwumere, M.C. (2022c). Streptococcus suis in Pigs and Environs: A Cross-sectional Study. IPS Journal of Public Health, 1(2), 9-12. https://doi.org/10.54117/ijph.v1i2.4.

Iheukwumere, I. H., Iheukwumere, M. C., & Nwakoby, N. E. (2022d). Sequential Pathogenicity Study of SOR+ and SOR-Escherichia coli Isolated from Roasted Meat. *IPS Intelligentsia Multidisciplinary Journal*, *1*(1), 1-11.

Iheukwumere, C. M., & Iheukwumere, I. H. (2022e). Hematological indices and sensory quality of fermented soybean condiments. *World Journal of Advanced Research and Reviews*, 14(2), 435-42

Iheukwumere, C. M., Umeaku, C. N., Chukwura, E. N., & Iheukwumere, I. H. (2022f). Characterization of the indigenous fermenters for the production of fermented condiments from soybean seeds. *World Journal of Advanced Research and Reviews*, *14*(2), 423-434.

Iheukwumere, I.H. and Iheukwumere, M.C. (2022g). Cross-sectional Study of Multiple Antibiotic-resistant Streptococcus suis in Pigs and Environments. IPS Interdisciplinary Journal of Biological Sciences, 1(1), 19–21. https://doi.org/10.54117/iijbs.v1i1.4

Iheukwumere, C. M., Iheukwumere, I. H., Okoli, U. O., & Ugwu, C. H. (2023a). Immunological Impact of Fermented Soybean Condiments Produced from Indigenous Fermenters. *Journal of Advances in Microbiology* 23(10): 27-37

Iheukwumere, C. M., Iheukwumere, I. H., Ugwu, C. H., & Okoli, U. O. (2023b). Toxicity of Prepared Fermented Soybean Condiments from Indigenious Fermenters. *Journal of Advances in Microbiology* 23(10): 38 – 51.

Iheukwumere, I.H. , Iheukwumere, C.M. , Nnadozie, H. C. ,Unaeze, C.B. , Obiefuna, O.H. Obianom, A.O. and Ejike, C. E. (2024). Hematotoxicological and mosquito larvicidal studies of crystal proteins secreted by Bacillus thuringiensis and Bacillus sphaericus. *Tropical Journal of Applied Natural Sciences* 2(2): 61 – 92.

Iheukwumere, I. H., Iheukwumere, C. M., Obianom, A. O., Nnadozie, C. H., Okereke, F. O., Onwuasoanya, U. F., … Ihenatuoha, U. A. (2025a). Cross-Sectional Study of Different Strains of Bacillus cereus among Pap Sold in Major Towns in Ihiala LGA, Anambra State. IPS Journal of Public Health, 5(2), 199–204. https://doi.org/10.54117/ijph.v5i2.39.

Iheukwumere, I. H., Iheukwumere, C. M., Obianom, A. O., Nnadozie, C. H., Okereke, F. O., Onwuasoanya, U. F., … Destiny, E. C. (2025b). Cross-Sectional Study of Major Strains of Salmonella enterica Subspecies Enterica Serovar Typhi among Borehole Used in Uli Community. IPS Journal of Public Health, 5(2), 205–210. https://doi.org/10.54117/ijph.v5i2.40.

Iheukwumere, I. H., Ajeh, J. C., Iheukwumere, C. M., Ike, V. E., Obianom, A. O., Ihenatuoha, U. A. ., Igboanugo, E. U., Onwuasoanya, U. F., Okereke, F. O., Nnadozie, C. H., Agbaugo, C. F., Nwike, M. I., Nwakoby, N. E., & Ilechukwu, C. C. (2025c). Exploring the Phytochemical and Antimicrobial Properties of Fruit Vinegar: A Study on Phoenix Dactylifera and Malus Sylvestris. *IPS Journal of Applied Microbiology and Biotechnology*, 4(1), 115–122. https://doi.org/10.54117/ijamb.v4i1.48

Iheukwumere, I. H., Ajeh, J. C., Iheukwumere, C. M., Ike, V. E., Obianom, A. O., Ihenatuoha, U. A., Igboanugo, E. U., Onwuasoanya, U. F., Okereke, F. O., Nnadozie, C. H., Agbaugo, C. F., Nwike, M. I., Nwakoby, N. E., & Ilechukwu, C. C. (2025d). Microbial Quality and Sensory Assessment of Vinegar from Date Palm and Apple Fruits: Implications for Consumer Preference. IPS Journal of Nutrition and Food Science, 4(2), 410–417. https://doi.org/10.54117/ijnfs.v4i2.100.

Iheukwumere, I. H., Iheukwumere, C. M., Obianom, A. O., Nnadozie, C. H., Okereke, F. O., Onwuasoanya, U. F., Udeagbara, O. E., Unaeze, B. C., Obiefuna, O. H., Ike, V. E., Onyemekara, N. N., & Ihenatuoha, U. A. (2025e). Quotidian of Substantial Strain of Shigella dysenteriae among Ready To-Eat Fruit Salad Sold in Uli Community. Journal of Pollution Monitoring, Evaluation Studies and Control, 4(1), 95–99. https://doi.org/10.54117/jpmesc.v4i1.17

Iheukwumere, I. H., Ajeh, J. C., Iheukwumere, C. M., Ike, V. E., Obianom, A. O., Ihenatuoha, U. A., Igboanugo, E. U., Onwuasoanya, U. F., Okereke, F. O., Nnadozie, C. H., Nwike, M. I., Nwakoby, N. E., & Ilechukwu, C. C. (2025f). Safety Evaluation of Vinegar from Phoenix Dactylifera and Malus Sylvestris: Toxicity and Acetic Acid Content. IPS Journal of Applied Microbiology and Biotechnology, 4(1), 123–131. https://doi.org/10.54117/ijamb.v4i1.49

Iheukwumere, C. M., Ekesiobi, A. O., Iheukwumere, I. H., Ejike, C. E., Ilechukwu, C. C., Dim, C. N., & Ochibulu, S. C. (2025g). Dual Approach Therapy: Assessing Xylopia aethiopica and Ciprofloxacin Synergy against Salmonella enterica Serovar Typhi. *IPS Intelligentsia Multidisciplinary Journal*, *4*(1), 27–31. https://doi.org/10.54117/iimj.v4i1.9.

Iheukwumere, C. M., Ekesiobi, A. O., Iheukwumere, I. H., Ejike, C. E., Ilechukwu, C. C., Dim, C. N., Ochibulu, S. C., Unegbu, C. C., & Egbuna, C. (2025h). Food Safety Implications: Assessing the Potential of Desmodium velutinum Leaves Extracts to Control the Most Predominant Fungal Contamination in Ready-To-Eat Fried Chicken. *IPS Journal of Nutrition and Food Science*, *4*(3), 494–500. https://doi.org/10.54117/ijnfs.v4i3.111

Iheukwumere, I.H., Dimejesi, S.A.,Iheukwumere, C.M., Chude, C.O., Nwaolisa, C.N.,Ukoha, C.C., Nwakoby,N.E., Egbuna, C. and Egbe, P.A. (2020) Diversity and molecular characterization of keratinophilic fungi from soil samples. *International Journal of Research Publication* 50(1); 1047 -1062.

Iheukwumere, I. H., Obi, P. C. and Unaeze, B. C. (2017a). A trial to prevent *Vibrio cholerae* in albino mice using autogenous bacterin. *Advances in Life Science and Technology* 58:34–42

Iheukwumere, I. H., & Ejike, C. E. (2017b). Comparative study of the inhibitory activities of Ocimum gratissimum and Nepeta cataria against Salmonella enterica serovar Typhi and their larvicidal effect against Anopheles gambiae. *African Journal of Education, Science and Technology (AJEST)*, *3*(4), 16-24

Iheukwumere, I. H., Amadi, E. R., & Chude, C. (2018b). Synergistic Effects of Probiotics and Autogenous Bacterin Against Inositol Negative Motile Salmonella Species. *Journal of Biology, Agriculture and Healthcare* 8(6).

Iheukwumere, I. H., Amadi, R. E., Unaeze, B. C., & Campus, N. (2017c). Enterotoxigenicity Profile of Salmonella Enterica Serovar Typhimurium in Suckling Albino Mice. Journal of Natural Sciences Research 7(14).

Iheukwumere, I. H., Chukwura, E. I., & Chude, C. (2018c). In vivo activities of some selected antimicrobial agents against enteric bacteria isolated from chicken feeds on broiler layers. *Journal of Biology, Agriculture and Healthcare*, *8*(6).

Iheukwumere, I. H., Ejike, C. E., & Okeke, C. E. (2017d). A trial to prevent sorbitol negative Escherichia coli infections in chicks using autogenous bacteria and probiotics. *Journal of Natural Sciences Research*, *7*, 56-63.

Iheukwumere, I. H., Uneze, B. C., & Ejike, C. E. (2017e). Efficacy of some selected antimicrobial substances in prevention of enteric bacterial infection in broiler chicks. *J. Biol. Agriculture. Healthcare*, *7*, 58-66.

Iheukwumere, I. H., Iheukwumere, C. M., Obianom, A. O., Nnadozie, C. H., Okereke, F. O., Onwuasoanya, U. F., … Ihenatuoha, U. A. (2025i). Cross-Sectional Study of Different Strains of Bacillus cereus among Pap Sold in Major Towns in Ihiala LGA, Anambra State. IPS Journal of Public Health, 5(2), 199–204. https://doi.org/10.54117/ijph.v5i2.39.

Iheukwumere, I. H., Iheukwumere, C. M., Obianom, A. O., Nnadozie, C. H., Okereke, F. O., Onwuasoanya, U. F., … Destiny, E. C. (2025j). Cross-Sectional Study of Major Strains of Salmonella enterica Subspecies Enterica Serovar Typhi among Borehole Used in Uli Community. IPS Journal of Public Health, 5(2), 205–210. https://doi.org/10.54117/ijph.v5i2.40.

Iheukwumere, I. H., Ajeh, J. C., Iheukwumere, C. M., Ike, V. E., Obianom, A. O., Ihenatuoha, U. A. ., Igboanugo, E. U., Onwuasoanya, U. F., Okereke, F. O., Nnadozie, C. H., Agbaugo, C. F., Nwike, M. I., Nwakoby, N. E., & Ilechukwu, C. C. (2025k). Exploring the Phytochemical and Antimicrobial Properties of Fruit Vinegar: A Study on Phoenix Dactylifera and Malus Sylvestris. *IPS Journal of Applied Microbiology and Biotechnology*, 4(1), 115–122. https://doi.org/10.54117/ijamb.v4i1.48

Iheukwumere, I.H., Nwike, M. I., Iheukwumere, C.M., Ike, V.E., Obianom, A.O., Ihenatuoha, U.A., Igboanugo, E.U., Ekesiobi, A.O., Okereke, F.O., Obiefuna, O. H. Nnadozie, C.H., Agbaugo, C.F., Oduoye, O.T., Nwakoby, N.E., Ilechukwu, C. C.,, Ochibulu, S. C. and Ejike, C. E. (2025l). Extraction and Elucidation of Antibiotics from the Mycelia of Aspergillus niger Isolated from Poultry Farm against Enteric Bacterial Pathogens. *IPS Journal of Advanced and Applied Biochemistry,* 1(1), 1–10. https://doi.org/10.54117/ijaab.v1i1.58.

Ike, V. E., Iheukwumere, I. H., Iheukwumere, C. M., Dim, C. N., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., & Ochibulu, S. C. (2025a). Prevalence of Bacillus cereus in Powdered Soybean Sold in Uli

Community, Anambra State: A Cross-Sectional Study. *IPS Journal of Basic and Clinical Medicine*, *2*(3), 108–114. https://doi.org/10.54117/ijbcm.v2i3.18

Ike, V. E., Iheukwumere, I. H., Iheukwumere, C. M., Dim, C. N., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., & Ochibulu, S. C. (2025b). *Bacillus cereus in Uli's cornflour: A prevalence study*. *IPS Journal of Nutrition and Food Science, 4*(3), 544–548. https://doi.org/10.54117/8btte840

Ike, V. E., Iheukwumere, I. H., Iheukwumere, C. M., Dim, C. N., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., & Ochibulu, S. C. (2025c). Pathogenic Profile Analysis: In Vitro Screening of Enteric Bacteria from University Dusters. *IPS Journal of Applied Microbiology and Biotechnology*, *4*(3), 187–191. https://doi.org/10.54117/ijamb.v4i3.76

Ike, V. E., Iheukwumere, I. H., Iheukwumere, C. M., Dim, C. N., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., & Ochibulu, S. C. (2025d). Frozen Fish Pathogens: Antimicrobial Resistance and Public Health Implications. *IPS Interdisciplinary Journal of Biological Sciences*, *4*(4), 138–143. https://doi.org/10.54117/iijbs.v4i4.77

Ike, V. E., Iheukwumere, I. H., Iheukwumere, C. M., Dim, C. N., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., & Ochibulu, S. C. (2025e). Stream water quality assessment: Antibiotic resistance of Lac-positive enteric bacterial isolates. *Journal of Pollution Monitoring, Evaluation Studies and Control, 4*(2), 120–125. https://doi.org/10.54117/jpmesc.v4i2.21.2025

Ke, K., Sun, Y., He, T., Liu, W., Wen, Y., Liu, S., ... & Gao, X. (2024). Effects of feather hydrolysates generated by probiotic Bacillus licheniformis WHU on gut microbiota of broiler and common carp. *Journal of Microbiology*, *62*(6), 473-487.

Khan, S.H. and Iqbal, J. (2016). Recent advances in the role of organic acids in poultry nutrition. *Journal of Applied Animal Research* 44:359–369

Kim, J., Lee, H., & Kim, Y. (2018). Effects of fermented milk products on cholesterol and blood pressure: A review of clinical trials. *Journal of Dairy Science, 101*(4), 2724–2741. https://doi.org/10.3168/jds.2017-13708

Kim, S. K., Kim, T. H., Lee, S. K., Chang, K. H., Cho, S. J., Lee, K. W., & An, B. K. (2016). The Use of Fermented Soybean Meals during Early Phase Affects Subsequent Growth and Physiological Response in Broiler Chicks. *Asian-Australasian Journal of Animal Sciences*, *29*(9), 1287. https://doi.org/10.5713/ajas.15.0653

Kovács, Á. T. (2019). Bacillus subtilis. *Trends in microbiology*, *27*(8), 724-725.

Latorre, J.D., Adhikari, B., Park, S.H., Teague, K.D., Graham, L.E. and Mahaffey, B.D.. (2018). Evaluation of the epithelial barrier function and ileal microbiome in an established necrotic enteritis challenge model in broiler chickens. *Frontier in Veterinary Science* 5:199 – 206

Le Doare, K., Holder, B., Bassett, A., & Pannaraj, P. S. (2020). Mother's milk: A purposeful contribution to the development of the infant microbiota and immunity. *Frontiers in Immunology, 11*, 362. https://doi.org/10.3389/fimmu.2020.00362

Li, Y., Guo, B., Wu, Z., Wang, W., Li, C., Liu, G., & Cai, H. (2020). Effects of Fermented Soybean Meal Supplementation on the Growth Performance and Cecal Microbiota Community of Broiler Chickens. *Animals*, *10*(6), 1098. https://doi.org/10.3390/ani10061098

Lv, W., Ma, Y., Zhang, Y., Wang, T., Huang, J., He, S., Du, H., & Guo, S. (2023). Effects of Lactobacillus plantarum fermented Shenling Baizhu San on gut microbiota, antioxidant capacity, and intestinal barrier function of yellow-plumed broilers. *Frontiers in Veterinary Science*, *10*, 1103023. https://doi.org/10.3389/fvets.2023.1103023

Manasseh, C.O., Logan, C.S.P., Ikeyi, A.P., Ede, K.K., Iheukwumere, I.H., Iheukwumere, C.M. and Ejike, C.E. (2025). Investigating the Effects of the Covid-19 Pandemic and Climate Risks on Trade Balance in Emerging Markets. *The Nigerian Health Journal* 25(2): 1-27. https://doi.org/10.71637/tnhj.v25i2.914

Marco, M. L., Sanders, M. E., Gänzle, M., Arrieta, M. C., Cotter, P. D., De Vuyst, L., ... & Hutkins, R. (2021). The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on fermented foods. *Nature Reviews Gastroenterology and Hepatology, 18*(3), 196–208. https://doi.org/10.1038/s41575-020-00390-5

Melini, F., Melini, V., Luziatelli, F., Ficca, A. G., & Ruzzi, M. (2019). Health-promoting components in fermented foods: An up-to-date systematic review. *Nutrients, 11*(5), 1189. https://doi.org/10.3390/nu11051189

Meyer, B., Bessei, W., Vahjen, W., Zentek, J., & Harlander-Matauschek, A. (2012). Dietary inclusion of feathers affects intestinal microbiota and microbial metabolites in growing Leghorn-type chickens. *Poultry Science*, *91*(7), 1506-1513. https://doi.org/10.3382/ps.2011-01786

Nwike, M.I., Iheukwumere, I.H. and Uneze, B.C. (2017). Effect of Spices, pH and Temperature on the Survival and Multiplication of Staphylococcus aureus in Locally Made Soya Milk Drink. *Journal of Natural Sciences Research* 7(4).

Obianom, A.O. , Iheukwumere, I.H. , Iheukwumere, C.M. , Ochibulu, S.C., Nnadozie, H. C. and Ifenetu, F. C. (2024). Supersizing the inhibitory activity of Xylopia aethiopica extract against Vibrio cholerae using doxycycline. *Tropical Journal of Applied Natural Sciences* 2(2).

Obiefuna, U. G., Umeh, S. O., & Iheukwumere, I. H. (2025c). Micronutrient profile and acceptability of tomato jam: A comparison of high and low glycemic carbohydrate index sweeteners. *IPS Applied Journal of Nutrition, Food and Metabolism Science, 3*(2), 67–74. https://doi.org/10.54117/pr4r6c73

Obiefuna, U. G., Umeh, S. O., & Iheukwumere, I. H. (2025a). Assessing the Impact of Glycemic Index on Microbial Quality and Storage Stability of Tomato Jam. *IPS Journal of Applied Microbiology and Biotechnology*, *4*(3), 192–202. https://doi.org/10.54117/ijamb.v4i3.81

Obiefuna, U. G., Umeh, S. O., & Iheukwumere, I. H. (2025b). Physicochemical and nutritional properties of tomato jam: Influence of sweetener type and glycemic index. *IPS Journal of Nutrition and Food Science, 4*(3), 561–568. https://doi.org/10.54117/yazv0114

Okeke, C. E. Iheukwumere, I. H. Ejike, C.E. (2017). Pathogenicity Study of Dematiaceous Fungi Isolated from Chicken Feeds on Immunoincompetent Chickens. *J. Biol. Agriculture. Healthcare* 7(4).

Omar, A. E., S., H., Ismail, T. A., M., R., M., S. A., Shalaby, S. I., & Ibrahim, D. (2021). Performance, Serum Biochemical and Immunological Parameters, and Digestive Enzyme and Intestinal Barrier-Related Gene Expression of Broiler Chickens Fed Fermented Fava Bean By-Products as a Substitute for Conventional Feed. *Frontiers in Veterinary Science*, *8*, 696841. https://doi.org/10.3389/fvets.2021.696841

Onunkwo, D. N., & Ekine, O. A. (2020). Performance of broiler chickens fed diet containing fermented maize milling waste. *Nigerian Journal of Animal Production*, *47*(1), 214–220.

Ouwehand, A. C., Forssten, S. D., Hibberd, A. A., & Tiihonen, K. (2022). Probiotic and other functional microbes: From foods to health applications. *Current Opinion in Biotechnology, 73*, 237-242. https://doi.org/10.1016/j.copbio.2021.07.001

Park, K. Y., Jeong, J. K., Lee, Y. E., & Daily, J. W. (2021). Health benefits of kimchi (Korean fermented vegetables) as a probiotic food. *Journal of Medicinal Food, 27*(3), 365–375. https://doi.org/10.1089/jmf.2020.0155

Premathilaka, K. T., Nawarathne, S. R., Nambapana, M. N., Macelline, S. P., Wickramasuriya, S. S., Ang, L., Jayasena, D. D., & Heo, J. M. (2020). Partial or complete replacement of fishmeal with fermented soybean meal on growth performance, fecal composition, and meat quality in broilers. *Journal of Animal Science and Technology*, *62*(6), 824. https://doi.org/10.5187/jast.2020.62.6.824

Ragavi, G., Muthamilan, M., Nakkeeran, S., Kumaravadivel, N., Sivakumar, U., & Suganthi, A. (2019). Phenotypic and molecular characterization of endophytic bacteria isolated from banana. *Current Journal of Applied Science and Technology*, *38*(6), 1-10.

Safari, H., & Mohit, A. (2024). Feather meal processing methods impact the production parameters, blood biochemical indices, gut function, and hepatic enzyme activity in broilers. *Journal of Animal Science*, *102*. https://doi.org/10.1093/jas/skae068

Safari, H., Mohit, A., & Mohiti-Asli, M. (2024). Fermented feather meal improves the antioxidant status, meat quality, and immune response of broilers. *Iran J Vet Med*, 1-43.

Salehizadeh, M., Ebrahimi, M. T., Mousavi, S. N., Sepahi, A. A., & Orooji, R. (2025). Transforming Feather Meal Into a High-

Performance Feed for Broilers. *Veterinary Medicine and Science*, *11*(1), e70199. https://doi.org/10.1002/vms3.70199

Savaiano, D. A., & Hutkins, R. W. (2020). Yogurt, cultured fermented milk, and health: A review. *Nutrients, 12*(5), 1256. https://doi.org/10.3390/nu12051256

Soumeh, E., Mohebodini, H., Toghyani, M., Shabani, A., Ashayerizadeh, A., & Jazi, V. (2019). Synergistic effects of fermented soybean meal and mannan-oligosaccharide on growth performance, digestive functions, and hepatic gene expression in broiler chickens. *Poultry Science*, *98*(12), 6797-6807. https://doi.org/10.3382/ps/pez409

Stanton, C., Ross, R. P., Fitzgerald, G. F., & van Sinderen, D. (2021). Fermented functional foods: Trends and challenges. *Current Opinion in Food Science, 40*, 114–122. https://doi.org/10.1016/j.cofs.2021.02.005

Stefanello, C., Rosa, D.P., Dalmoro, Y.K., Segatto, A.L., Vieira, M.S. and Moraes, M.L. (2020). Protected blend of organic acids and essential oils improves growth performance, nutrient digestibility, and intestinal health of broiler chickens undergoing an intestinal challenge. Frontier in Veterinary Science 6:491 – 501

Timbermont, L., Lanckriet, A., Dewulf, J., Nollet, N., Schwarzer, K. and Haesebrouck, F. (2010). Control of Clostridium perfringens-induced necrotic enteritis in broilers by target-released butyric acid, fatty acids and essential oils. Avian Pathology 2: 10 – 16

Ugwu, C. H., Iheukwumere, I. H., Iheukwumere, C. M., Ike, V. E., Dim, C. N., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., Oduenyi, P. M., & Ochibulu, S. C. (2025a). Maternal health and antibiotic resistance: *Klebsiella pneumoniae* isolates analysis. *IPS Journal of Public Health, 5*(3), 290–295. https://doi.org/10.54117/s3tx6v26

Ugwu, C. H., Iheukwumere, I. H., Iheukwumere, C. M., Ike, V. E., Dim, C. N., Ezendianefo, J. N., Egbe, P. A., Oragwu, I. P., Orji, C. C., Ogbonnaya, O. C., Onwuasoanya, U. F., Okereke, F. O., Oduenyi, P. M., & Ochibulu, S. C. (2025b). Ocimum gratissimum Extract's Effectiveness against Vibrio cholerae from Uli Streams. *IPS Journal of Phytochemistry and Medicinal Plant Research*, *1*(2), 15–19. https://doi.org/10.54117/ijpmpr.v1i2.38

Vijayalakshmi, S., Ranjitha, J., & Rajeswari, V. D. (2013). Enzyme production ability by Bacillus subtilis and *Bacillus licheniformis* comparative study. *Asian J. Pharm. Clin. Res*, *6*(4), 29-32.

Wang, J., Yao, L., Su, J., Fan, R., Zheng, J., & Han, Y. (2023). Effects of Lactobacillus plantarum and its fermentation products on growth performance, immune function, intestinal pH, and cecal microorganisms of Lingnan yellow chicken. *Poultry Science*, *102*(6), 102610. https://doi.org/10.1016/j.psj.2023.102610

Wastyk, H. C., Fragiadakis, G. K., Perelman, D., Dahan, D., Merrill, B. D., Yu, F. B., ... & Sonnenburg, J. L. (2021). Gut-microbiota-targeted diets modulate human immune status. *Cell, 184*(16), 4137–4153.e14. https://doi.org/10.1016/j.cell.2021.06.019

Xiao, C., Li, X., Ding, Z., Zhang, H., Lv, W., Yang, C., He, D., & Zhu, L. (2023). Enhancing Growth and Gut Health in Squabs: The Impact of Fermented Mixed Feed. *Animals*, *14*(10), 1411. https://doi.org/10.3390/ani14101411

Yan, L., An, S., Lv, X., Lv, Z., Zhang, B., Choct, M., Guo, Y., Wang, Z., Yan, B., & Li, Y. (2025). Effects of replacing soybean meal with cottonseed meal on growth performance, carcass trait, intestinal development and intestinal microbiota of broiler chickens. *Poultry Science*, *104*(2), 104653. https://doi.org/10.1016/j.psj.2024.104653

Yang, R., Khalid, A., Khalid, F., Ye, M., Li, Y., Zhan, K., Li, Y., Liu, W., & Wang, Z. (2022). Effect of fermented corn by-products on production performance, blood biochemistry, and egg quality indices of laying hens. *Journal of Animal Science*, *100*(5). https://doi.org/10.1093/jas/skac130

Yeh, R., Hsieh, C., & Chen, K. (2023). Two-Stage Fermented Feather Meal Enhances Growth Performance and Amino Acid Digestibility in Broilers. *Fermentation*, *9*(2), 128. https://doi.org/10.3390/fermentation9020128

Yu, L., Zhihui, C., Hongzhi, W., Fengjie, J., Yang, L., Jianing, L., ... & Liangmei, X. (2024). Effects of Fermented Puffed Feather Meal on Growth Performance, Serum Biochemical Indices, Meat Quality, and Intestinal Microbiota in Broilers. *Journal of Northeast Agricultural University*, *31*(3).

Zhang, Y., Liu, M., Ding, Y., Wang, T., Ma, Y., Huang, J., ... & Guo, S. (2023). Effects of perilla seed meal on productive performance, egg quality, antioxidant capacity and hepatic lipid metabolism of Wenchang breeder hens. *Animals*, *13*(22), 3587.

Zhang, Y., Liu, Y., Lv, L., & Wang, J. (2021). Probiotics in the prevention and treatment of colorectal cancer. *Frontiers in Immunology, 12*, 714947. https://doi.org/10.3389/fimmu.2021.714947

Zhao, L., Zhang, F., Ding, X., Wu, G., Lam, Y. Y., Wang, X., ... and Zhang, C. (2022). Gut bacteria selectively promoted by dietary fibers alleviate type 2 diabetes. *Science, 359*(6380), 1151-1156. https://doi.org/10.1126/science.aao5774

.