TRENDS IN ANIMAL AND PLANT SCIENCES
https://doi.org/10.62324/TAPS/2025.087
www.trendsaps.com; editor@trendsaps.com
E-ISSN: 3006-0559; P-ISSN: 3006-0540

RESEARCH ARTICLE

Antibacterial Efficacy of Tulsi (*Ocimum sanctum*) Leaf Extract against Multidrug-resistant Poultry Pathogens

Samia sarfraz[1], Saira Rehman[2], Aamna Shah[3], Amna Iqbal[4], Muhammad Ubaid Ullah Younis[5], Shakeel Sabir[6] and Ayesha Waris[7]

[1]Institute University of Northampton, United Kingdom.
[2]Faculty of Pharmaceutical Sciences, Lahore University of Biological and Applied Sciences, Pakistan.
[3]Department of Pharmacy, The University of Lahore, Sargodha Campus, Pakistan.
[4]Department of Zoology, Division of Science and Technology, University of Education Lahore 54600, Punjab, Pakistan.
[5]Faculty of Veterinary and Animal Sciences, PMAS Arid Agriculture University, Pakistan.
[6]Department of Higher Education, Government of Azad Jammu and Kashmir, Pakistan.
[7]Department of Physiology, Faculty of Sciences, The University of Faisalabad, Pakistan.
***Corresponding author:** samiasarfraz88@gmail.com

Article History: 25-042 Received: 10-Aug-2025 Revised: 15-Sep-2025 Accepted: 22-Sep-2025

ABSTRACT

The current study was conducted to determine the effectiveness of Tulsi (*Ocimum sanctum*) leaf ethanolic extract against poultry-related multidrug-resistant (MDR) bacterial infections. *E. coli*, *S. gallinarum*, *S. pullorum*, and Pasteurella multocida MDR isolates were also defined and tested for their antibiotic resistance. The Soxhlet technique was used to prepare an Ethanolic Tulsi leaf extract and investigate its antibacterial activity to determine the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC). The results demonstrated that antibacterial activity depends on concentration. The extract exhibited the best inhibition against P. multocida (MIC 25 mg/mL, MBC 50 mg/mL), then S. gallinarum and *S. pullorum* (MIC 50 mg/mL, MBC 100mg/ mL), and *E. coli* (MIC 100mg/ mL, MBC 112.5mg/ mL). These results suggest that Tulsi extract has strong bacteriostatic and bactericidal properties on MDR poultry pathogens. Its phytoconstituents that destroy bacterial membranes and prevent protein synthesis are attributed to the observed activity of eugenol, ursolic acid, and rosmarinic acid. The research explains the use of *O. sanctum* as a natural and environmentally friendly antibacterial compound in the health management of poultry as a viable means of fighting antimicrobial resistance.

Key words: *Ocimum sanctum*, Poultry, Pathogens, Tulsi, Antibacterial activity.

INTRODUCTION

The poultry business is important in supplying cheap animal proteins to the world population. Poultry production is a significant source of livelihood for both small and large-scale farmers in most developing nations, including Asia and Africa (Chiekezie et al., 2022). There has been a rapid increase in poultry farming activities, causing the proliferation of infectious bacterial diseases like *colibacillosis, salmonellosis, staphylococcosis*, and *pasteurellosis*, which have a great impact on lowering productivity and raising the death rates. *E. coli*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, and *S. gallinarum* are the most common bacterial pathogens that cause systemic and localized infections in poultry.

Antibiotics have been extensively applied to poultry over the decades as growth factors, therapeutics, and prophylaxis (Rahman et al., 2022). These management practices have yielded beneficial effects on the health of flocks and the efficiency of feed conversion, although the prolonged and uncontrolled use of antimicrobials has led to the emergence of MDR bacterial strains. The transmission of these resistant pathogens occurs as a result of direct contact between poultry and

Cite This Article as: Sarfraz S, Rehman S, Shah A, Iqbal A and Younis MUU, 2025. Antibacterial efficacy of Tulsi (*Ocimum sanctum*) leaf extract against multidrug-resistant poultry pathogens. Trends in Animal and Plant Sciences 6: 137-142. https://doi.org/10.62324/TAPS/2025.087

people, by handling contaminated poultry meat, or eating poultry products that have not been cooked properly, which is a severe threat to human health. The growing problem of antimicrobial resistance (AMR) has thus necessitated an urgent need to identify alternative, natural, and environmentally friendly antibacterial agents to effectively counteract the resistant pathogens to have sustainable health management of the poultry (Ayana and Kamutambuko, 2024).

Medicinal plants have been a long-known effective part of traditional medicine, including Ayurveda, Unani, and Chinese medicine (Mansoori and Chouhan, 2023). Their broad phytochemical profile, which includes alkaloids, flavonoids, tannins, saponins, phenolic compounds, and essential oils, is the reason behind a broad range of biological activities, such as antibacterial, antifungal, antioxidant, and immunomodulatory ones. Over the past few years, a number of plant-based compounds have been examined as promising alternatives to synthetic antibiotics to be used in poultry production or as primary supplements. Tulsi is considered the queen of herbs in Indian traditional medicine because it has a great variety of pharmacological actions. Tulsi has been reported to have antimicrobial activity against a variety of human and animal pathogens, e.g., *E. coli*, *S. aureus*, *Pseudomonas aeruginosa* and *Salmonella* spp (Binte Ehsan, 2022). Its potential with MDR poultry isolates, however, has not been studied. A number of studies have revealed that Tulsi extracts and especially ethanol and methanol extracts, have potent bacteriostatic and bactericidal activity that results in cell wall destabilization, membrane permeability, and destabilization of protein synthesis. The Tulsi phenolic compounds are thought to induce oxidative stress and release of intracellular elements, which consequently results in bacterial death (Arya et al., 2024). These phytochemicals also work by various mechanisms in contrast to the traditional antibiotics, and this decreases the chances of resistance being developed. Moreover, plant-based antimicrobials are biodegradable, economical, and safe to the poultry and consumers thus a viable substitute in reducing disease. Examining the antibacterial efficacy of *O. sanctum* leaf extract against MDR bacterial strains of avian origin has become crucial due to the growing issue of antibiotic resistance among poultry diseases (Olawuwo et al., 2022). Such research is essential for promoting the integration of phytotherapeutic approaches into modern veterinary care as well as for finding sustainable and efficient substitutes for traditional antibacterial drugs. Therefore, investigating bioactive chemicals produced from plants offers a viable and sustainable strategy to address the increasing problem of antibiotic resistance in poultry production systems. The findings of this study are anticipated to offer scientific validation for the use of *O. sanctum* as a natural antibacterial alternative in poultry production systems, contributing to sustainable disease management and reduced reliance on synthetic antibiotics.

MATERIALS AND METHODS

Collection of *O. sanctum* Leaves

The botanical garden of the University of Lahore, Pakistan, was used to collect the fresh *O. sanctum* leaves. The gathered leaves were put in sterile polythene bags, where they were labeled well and taken to the Microbiology Laboratory to prepare the *O. sanctum* leaf extract. The study was conducted on mature disease-free leaves.

Preparation of *O. sanctum* Leaf Extract

The Tulsi leaves (500 g) were washed with sterilized distilled water after running tap water to remove dust and surface taint. Air drying of the leaves was done at 35-40 °C and the samples were dried until a constant weight was obtained, and then finally powdered using an electric grinder. A 96% ethanol extract using a Soxhlet apparatus (3,000 mL) was used to extract the powdered material in 18 hours at 78 °C. To obtain a semisolid residue, the extract obtained was concentrated at low pressure in a rotary evaporator (BUCHI Rota Vap R-114, Switzerland). The dried crude extract was weighed and put into an airtight container, and kept at 4 °C until further use.

Bacterial Strains

This study involved multidrug-resistant (MDR) bacterial isolates of poultry origin, which included *E. coli*, *S. gallinarum*. The bacterial strains were taken out of the culture collection, which was previously isolated in poultry exhibiting signs of *colibacillosis*, *staphylococcosis* and *salmonellosis* clinical signs. The isolates were identified by morphology of the colonies, Gram staining and normal biochemical reactions including catalase, coagulase, indole and triple sugar iron (TSI) tests. The bacterial strains were preserved in the nutrient broth at 4°C and subcultured in selective media before their use.

Antibiotic Susceptibility Test

The antibiotic susceptibility patterns of the bacterial isolates on Mueller-Hinton agar were determined using the disc diffusion method. Fourteen commonly used antibiotics were chosen, which included Azithromycin, Gentamicin, Vancomycin, Cephalexin, Streptomycin, Erythromycin, Nalidixic acid, Ampicillin, Sulphatrimethoprim, Ciprofloxacin, Doxycycline, Oxacillin, Tetracycline, and Chloramphenicol. The bacterial suspensions were each brought to 0.5 McFarland standard (around 1x108 CFU/mL) and brought into spread uniformly on the agar surface. Aseptically, antibiotic discs were put on the plates, and incubation was done at 37°C after 24 hours. According to the recommendations of the Clinical and

Laboratory Standards Institute, the inhibition zone areas were measured in millimeters and classified as susceptible, moderate, or resistant. Multidrug resistance was considered to be an isolate that is resistant to three or more antibiotic classes.

MIC and MBC

To determine the MIC and MBC, the antibacterial activity of the Tulsi leaf extract was determined by the broth dilution method. Two-fold dilution of Tulsi extract was made in sterile Mueller-Hinton broth to make concentrations of 200, 100, 50, 25, 12.5, 6.25, and 3.125 mg/mL. Each bacterial suspension (20 µL) that is equivalent to 0.5 McFarland standard, was added to the test tubes with 1 mL of diluted extract. Mueller-Hinton broth alone in one test tube was left as the negative control and broth plus ciprofloxacin (1 g/mL in P. multocida and 8 µg/mL in *E. coli*, *S. aureus* and *S. gallinarum*) was left as the positive control. The tubes were incubated at 37°C, 24 hours. Bacterial growth was visually determined after incubation in terms of turbidity and spectrophotometrically at 580 nm. The lowest concentration of Tulsi extract that inhibited observable bacterial growth was taken as the MIC.

To calculate the MBC, 50 µL aliquots of each tube that did not turbidise were serially diluted (10^{-1} to 10^{-10}) and plated on selective agar media - Blood agar on *P. multocida*, SS agar on *Salmonella* spp. and EMB agar on *E. coli*. Plates were incubated at 37°C for 24 hours and colony-forming units (CFU/mL) were counted by using the following CFU/mL = Number of colonies × Dilution factor. MBC was used to represent the lowest concentration of extracts with no visible growth of the colonies.

Statistical Analysis

Every experiment was carried out three times. Standard deviation (SD) ± mean was used to express the data. Using SPSS software version 25, the antibacterial effectiveness of various Tulsi extract concentrations was compared using the Student's t-test. The level of $p \leq 0.05$ was considered statistically significant.

RESULTS

Profiles of MDR of Poultry Bacteria

The antibiotic susceptibility test also demonstrated that *E. coli* was most susceptible to five antibiotic classes, then *S. gallinarum* and *S. pullorum* (four and four classes, respectively) and *P. multicida* (three classes) (Table 1). Such outcomes validate the fact that the tested isolates were multidrug-resistant (MDR).

MBC and MIC of the *O. sanctum* Leaf Extract against P. Multocida

The *O. sanctum* ethanolic extract of the leaves showed strong antibacterial activity against P. multocida. At concentrations of 100 and 50mg/mL, total growth inhibition was realized ($p < 0.001$). Growth of bacteria was less at 25 mg/mL (log 8.12 ± 0.48 CFU/mL) than in the untreated control (log 11.05 ± 0.42 CFU/mL). The inhibitory effect was not noticed at 12.5 mg/mL and below. MIC and MBC values were 25 and 50 mg/ mL, respectively (Table 2).

MBC and MIC of *O. sanctum* Leaf Extract on *S. pullorum*

Tulsi extract at 100mg/mL was able to totally inhibit the growth of *S. pullorum* ($p < 0.001$). A large decrease in the bacterial load (log 8.24 ± 0.35 CFU/mL) was observed in 50 mg/mL compared to the control (log 12.12 ± 0.27 CFU/mL). Partial inhibition occurred at 25 mg/ml, with no effect noted at lower concentrations of 12.5 mcg/mL and below. The MIC and MBC were determined to be 50 and 100 mg/mL, respectively (Table 3).

Antimicrobial Effects of MIC and MBC of *O. sanctum* Leaf Extract Versus *S. gallinarum*

At the concentration of 100 mg/ mL ($p < 0.001$), the ethanolic extract of Tulsi leaves prevented the growth of *S. gallinarum*. Significant reduction of bacteria was found at 50 mg/mL (log 8.09 ± 0.41 CFU/mL) as compared to the control (log 12.20 ± 0.33 CFU/mL). There was no inhibition at the concentrations of 6.25 and 3.125 mg/mL. The MIC and MBC were 50 and 100mg/mL, respectively (Table 4).

Table 1: Antimicrobial groups and tested agents used for evaluating MDR profiles of bacteria isolated from poultry

Bacterial species	Category of antimicrobial agent	Specific antimicrobial compound	Antimicrobial susceptibility result (S/I/R)
Salmonella pullorum	Cephalosporin	Cephalexin	R
	Aminoglycoside	Streptomycin	R
	Macrolide	Erythromycin	R
	Glycopeptide	Vancomycin	I
Pasteurella multocida	Aminoglycoside	Streptomycin	R
	Sulfonamide	Trimethoprim–sulfamethoxazole	I
	Quinolone	Nalidixic acid	R
S. gallinarum	Macrolide	Erythromycin	R
	Glycopeptide	Vancomycin	R
	Cephalosporin	Cefotaxime	R
	Aminoglycoside	Gentamicin	I
E. coli	Aminoglycoside	Streptomycin	R
	Glycopeptide	Vancomycin	R
	Cephalosporin	Cephalexin	R
	Tetracycline	Doxycycline	R
	Macrolide	Azithromycin	R

Table 2: MIC and MBC values of *Ocimum sanctum* leaf extract on *Pasteurella multocida*

Concentration (mg/mL)	Log CFU/mL	Log reduction
0	11.05 ± 0.42	0
6.25	11.02 ± 0.31	0.03
12.5	10.92 ± 0.29	0.13
25a	8.12 ± 0.48	2.93
50b	0	11.05
100	0	11.05

a MIC; b MBC

Table 3: MIC and MBC values of *Ocimum sanctum* leaf extract against *Salmonella pullorum*

Concentration (mg/mL)	Log CFU/mL	Log reduction
0	12.12 ± 0.27	0
6.25	12.08 ± 0.31	0.04
12.5	10.82 ± 0.36	1.30
25	9.42 ± 0.19	2.70
50a	8.24 ± 0.35	3.88
100b	0	12.12

a MIC; b MBC

Table 4: MIC and MBC values of *Ocimum sanctum* leaf extract against *S. gallinarum*

Concentration (mg/mL)	Log CFU/mL	Log reduction
0	12.20 ± 0.33	0
6.25	12.18 ± 0.28	0.02
12.5	10.90 ± 0.30	1.30
25	9.33 ± 0.26	2.87
50a	8.09 ± 0.41	4.11
100b	0	12.20

a MIC; b MBC

MBC and MIC of *O. sanctum* Leaf Extract on *E. coli*

At a concentration of 112.5 mg/mL ($p<0.001$), *E. coli* was fully inhibited from growing. The growth of bacteria at 100 mg/mL was significantly lower (log 8.22 ± 0.52 CFU/mL) as compared to the control (log 13.31 ± 0.29 CFU/mL). MIC and MBC of Tulsi leaf extract against *E. coli* were established as 100 mg/mL and 112.5 mg/mL, respectively (Table 5).

Table 5: MIC and MBC values of *Ocimum sanctum* leaf extract against *E. coli*

Concentration (mg/mL)	Log CFU/mL	Log reduction
0	13.31 ± 0.29	0
6.25	13.28 ± 0.21	0.03
12.5	12.95 ± 0.24	0.36
25	11.22 ± 0.33	2.09
50	9.34 ± 0.27	3.97
100a	8.22 ± 0.52	5.09
112.5b	0	13.31

a MIC; b MBC.

DISCUSSION

The current research results prove that ethanolic extract of *O. sanctum* has a phenomenal antibacterial effect against MDR bacterial causes of poultry diseases such as *Pasteurella multocida*, *Salmonella pullorum*, *S. gallinarum*, and *E. coli*. The observed inhibitory and bactericidal effects were concentration dependent with increase in the concentration of Tulsi extract (50 mg/mL) resulting in total inhibition of bacteria. These findings indicate that Tulsi may be used as a good substitute of synthetic antibiotics in managing MDR infections in poultry. Past research has emphasized the antimicrobial multistage effects of Tulsi which is mostly due to its high concentration of phytochemical constituents. The current findings are supported by the conclusions of Prakash and Gupta (2005) that reported Tulsi essential oil to have a strong antibacterial effect against Gram-positive and Gram-negative bacteria, including *S. aureus* and *E. coli*. Singh et al. (2018) determined that ethanolic Tulsi extracts reduced the growth of *Salmonella typhi* and *E. coli* when used in similar concentrations as those that were used in the current study. Tulsi antimicrobial activity has been associated with the presence of bioactive compounds that use the system of bacterial cell membrane disruption, the interference of enzyme activity, and nucleic acid production inhibition, including eugenol, ursolic acid, linalool, and β-caryophyllene (Bhattarai et al., 2024).

The current findings revealed that *P. multocida* was very sensitive to Tulsi extract, the MIC and MBC of 25 and 50mg/mL, respectively. *O. sanctum* extract suppressed *Pasteurella multocida* that caused respiratory infections in poultry, and attributed the results to the presence of phenolic and flavonoid compounds that destabilize the bacterial cell wall (Orimaye et al., 2024).

The prevention of Salmonella species in this research is also consistent with the results of Das et al. (2020), who argued that the ethanolic extract of Tulsi leaves has had a significant impact in minimizing populations of *S. enterica* in poultry meat samples, highlighting its importance in food safety and preventing infections.

The inhibitory activity towards *E. coli* showed that the leaf extracts of Tulsi produced a significant decrease in *E. coli* viability by disrupting the bacterial membrane potential (Imran et al., 2021). Tulsi extracts had synergistic activity with antibiotics such as ciprofloxacin and tetracycline on resistant *E. coli* strains to increase their vulnerability. This may indicate that Tulsi extract not only possesses an independent antibacterial action but also enhances the efficacy of traditional antibiotics, reducing the dosage required and reducing the development of resistance.

Explaining the difference in MIC and MBC values in the bacterial species in this study, differences in the cell wall structure and resistance mechanisms can be supported. Gram-negative bacteria like *E. coli* have their outer lipopolysaccharide layer that restricts the penetration of hydrophobic substances, and thus higher concentration of the extract is needed to completely inhibit it. The resistance of *E. coli* is slightly more than that of *P. multocida* could also be attributed to the efflux pumps and plasmid-mediated resistance factors common in enteric bacteria.

Some past studies attested the antimicrobial effect of Tulsi against veterinarian and poultry. Tulsi leaf extract had a significant effect of lowering bacterial load in broilers infected with *E. coli*, which resulted in positive growth performance and decreased mortality (Hasan et al., 2016). Tulsi extract added to the poultry feed improved their immunity and decreased the colonization of enteropathogenic bacteria in the intestine, which supports the findings that Tulsi is a potential natural growth promoter and disease prevention agent (Dhama et al., 2015). These findings are supported by the present findings, which highlight the potential of Tulsi as a poultry industry alternative antibacterial compound that is safe and sustainable. Tulsi extract has several benefits over synthetic antibiotics, as it is non-toxic, biodegradable, and less likely to cause microbial resistance because of its complex effect. Its numerous phytochemicals have synergistic actions, which render bacteria hard to adapt and develop cross-resistance (Hanumanthaiah et al., 2020). The increasing interest in the world regarding the production of poultry free of antibiotics has been in line with the application of herbal antimicrobials like Tulsi, which can guarantee food safety as well as the welfare of the animals.

The current research work confirms the emerging body of evidence that *O. sanctum* is a very promising natural source of antibacterial compounds that can be used to curb MDR poultry pathogens. It's repeated anti-*P. multocida*, *Salmonella*, and *E. coli* properties point to its potential as a natural substitute for conventional antibiotics in treating diseases in poultry. However, it is suggested to conduct additional research that includes in vivo experiments, toxicity tests, and determination of active components to confirm its practical uses in poultry farming systems.

Conclusion

The current experiment indicated that a high ethanolic extract of *O. sanctum* leaves has high antibacterial properties against MDR poultry pathogens such as *P. multocida*, *S. gallinarum*, *S. pullorum*, and *E. coli*. The inhibitory effect was dose-dependent, with *P. multocida* having the best susceptibility. These findings support the potential therapeutic use of Tulsi as a natural antibacterial substitute for synthetic antibiotics in the production of poultry. The wide-spectrum activity of Tulsi is probably explained by the presence of a wide range of phytochemical components that can work in a variety of mechanisms, reducing the development of resistance.

DECLARATIONS

Funding: There was no funding to carry out this research.

Acknowledgments: The authors gratefully acknowledge the Faculty of Pharmaceutical Sciences, Lahore University of Biological and Applied Sciences, for providing the necessary facilities, resources, and technical support to carry out this research.

Conflict of Interest: The authors declare that there is no known conflict of interest associated with this publication.

Data Availability: The data will be available upon request to the corresponding authors.

Ethics Statement: All animal studies have been approved by the relevant ethics committee or institutional supervisory board and have been conducted in accordance with the ethical standards.

Author Contributions: SS and SR conceptualized and designed the study. SS conducted the experiments and primary data collection. SR assisted in data analysis, interpretation of results, and supervision of laboratory work. AS, AI, MUUY, SSab, and AW contributed to literature review, experimental support, and manuscript drafting. All authors critically reviewed the manuscript and approved the final version.

Generative AI Statement: The authors declare that no Gen AI/DeepSeek was used in the writing/creation of this manuscript.

Publisher's Note: All claims stated in this article are exclusively those of the authors and do not necessarily represent those of their affiliated organizations or those of the publisher, the editors, and the reviewers. Any product that may be evaluated/assessed in this article or claimed by its manufacturer is not guaranteed or endorsed by the publisher/editors.

REFERENCES

Arya, R., Faruquee, H.M., Shakya, H., Rahman, S.A., Begum, M.M., Biswas, S.K. and Kim, J.J., 2024. *Harnessing the antibacterial, anti-diabetic and anti-carcinogenic properties of Ocimum sanctum Linn (Tulsi)*. *Plants*, 13(24), p.3516.

Ayana, G.U. & Kamutambuko, R., 2024. *Probiotics in disease management for sustainable poultry production*. *Advanced Gut & Microbiome Research*, 2024(1), p.4326438.

Bhattarai, K., Bhattarai, R., Pandey, R.D., Paudel, B. and Bhattarai, H.D., 2024. *A comprehensive review of the phytochemical constituents and bioactivities of Ocimum tenuiflorum*. *The Scientific World Journal*, 2024(1), p.8895039.

Binte Ehsan, T., 2022. *Study of antibacterial activities of the extracts of Neem, Pomegranate, and Tulsi and essential oils against Salmonella Typhi*. Doctoral dissertation, Brac University.

Chiekezie, N.R., Nwankwo, E.C. and Ozor, M.U., 2022. *Analysis of small scale broiler poultry production in South East Nigeria, West Africa*. *International Journal of Animal and Livestock Production Research*, 6(1), pp.1–16.

Dhama, K., Latheef, S.K., Mani, S., Samad, H.A., Karthik, K., Tiwari, R. and Tufarelli, V., 2015. *Multiple beneficial applications and modes of action of herbs in poultry health and production – a review*. *International Journal of Pharmacology*, 11(3), pp.152–176.

Hanumanthaiah, P., Panari, H., Chebte, A., Haile, A. and Belachew, G., 2020. *Tulsi (Ocimum sanctum) – a myriad medicinal plant, secrets behind the innumerable benefits*. *Arabian Journal of Medicinal and Aromatic Plants*, 6(1), pp.105–127.

Hasan, M.N., Mostofa, M., Sorwar, M.G., Hasan, M.T., Das, K. and Hossain, D.M.N., 2016. *Effects of Tulsi leaf extract on body weight gain in broiler production*. *Bangladesh Journal of Veterinary Medicine*, 14(1), pp.21–25.

Imran, M., Khan, A.S., Khan, M.A., Saeed, M.U., Noor, N., Warsi, M.H. and Qadir, A., 2021. *Antimicrobial activity of different plant extracts against Staphylococcus aureus and Escherichia coli*. *Polymers in Medicine*, 51(2), pp.69–75.

Mansoori, M.A. and Chouhan, B., 2023. *Recent growth of herbal drug as over-the-counter products*. *BR Nahata Smriti Sansthan International Journal of Pharmaceutical Sciences & Clinical Research*, 3(1).

Olawuwo, O.S., Famuyide, I.M. and McGaw, L.J., 2022. *Antibacterial and antibiofilm activity of selected medicinal plant leaf extracts against pathogens implicated in poultry diseases*. *Frontiers in Veterinary Science*, 9, p.820304.

Orimaye, O.E., Ekunseitan, D.A., Omaliko, P.C. and Fasina, Y.O., 2024. *Mitigation potential of herbal extracts and constituent bioactive compounds on Salmonella in meat-type poultry*. *Animals*, 14(7), p.1087.

Prakash, P. and Gupta, N., 2005. *Therapeutic uses of Ocimum sanctum Linn (Tulsi) with a note on eugenol and its pharmacological actions: A short review*. *Indian Journal of Physiology and Pharmacology*, 49(2), pp.125–131.

Rahman, M.R.T., Fliss, I. and Biron, E., 2022. *Insights in the development and uses of alternatives to antibiotic growth promoters in poultry and swine production*. *Antibiotics*, 11(6), p.766.

Singh, S., Taneja, M. and Majumdar, D.K., 2018. *Biological activities and medicinal properties of Ocimum sanctum Linn*. *Journal of Pharmacy and Pharmacology*, 70(3), pp.301–315.