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Research Article

ASSESSMENT OF AN ANTIMICROBIAL PROPERTY OF SELECTED ANTIBIOTICS AGAINST THE SELECTED BACTERIAL FOOD PATHOGENS

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ABSTRACT

Foodborne bacterial pathogens are a leading cause of food-related illnesses worldwide, posing significant public health and economic challenges. The increasing prevalence of antibiotic resistance among these pathogens necessitates continuous evaluation of antimicrobial agents. The present study investigates the antimicrobial efficacy of selected antibiotics against major bacterial food pathogens, including *Escherichia coli*, *Salmonella enterica*, *Staphylococcus aureus*, *Listeria monocytogenes*, and *Bacillus cereus*. Antibiotics belonging to different classes ampicillin, ciprofloxacin, tetracycline, chloramphenicol, and gentamicin—were assessed using agar disc diffusion, agar well diffusion, and minimum inhibitory concentration (MIC) assays. The results demonstrated differential susceptibility patterns among the tested pathogens. Ciprofloxacin and gentamicin exhibited the highest antimicrobial activity, while reduced sensitivity and resistance were observed against ampicillin and tetracycline, particularly in Gram-negative bacteria. Gram-positive pathogens showed comparatively higher susceptibility to most antibiotics. The observed resistance trends highlight the potential risks associated with the indiscriminate use of antibiotics in food production systems. This study emphasizes the importance of routine antimicrobial surveillance and rational antibiotic use to ensure food safety and mitigate the spread of resistant foodborne pathogens.

Keywords: Foodborne bacteria, Antibiotics, Antimicrobial activity, MIC, Antibiotic resistance, Food safety.

INTRODUCTION

Foodborne diseases remain a significant global public health challenge, affecting millions of people annually and resulting in substantial economic losses due to medical expenses, loss of productivity, and food recalls (https://www.who.int/news-room/fact-sheets/detail/food-safety). Bacterial pathogens are among the most common causative agents of foodborne illnesses, with *Escherichia coli*, *Salmonella spp.*, *Staphylococcus aureus*, *Listeria monocytogenes*, and *Bacillus cereus* frequently implicated in outbreaks associated with contaminated food and water (Yang *et al*., 2025). These microorganisms can contaminate food at various stages of production, processing, storage, and distribution, making effective control measures essential for ensuring food safety (Grace, 2025). Antibiotics have long been used as a primary means of treating bacterial infections in humans and animals (Enshaie *et al*., 2025). In food production systems, antibiotics have also been employed for therapeutic, prophylactic, and growth-promoting purposes, particularly in livestock (Odey *et al*., 2024). However, the widespread and often indiscriminate use of antibiotics has contributed to the emergence and dissemination of antibiotic-resistant bacteria. Foodborne pathogens exhibiting resistance to commonly used antibiotics pose a serious threat, as infections caused by such strains are more difficult to treat and may result in increased morbidity and mortality (Manyi-Loh *et al*., 2018).

Gram-negative bacteria such as *E. coli* and *Salmonella* are particularly concerning due to their intrinsic resistance mechanisms, including the presence of an outer membrane that restricts antibiotic entry and the ability to acquire resistance genes through horizontal gene transfer (Breijyeh *et al*., 2020). Gram-positive pathogens like *S. aureus* and *L. monocytogenes*, although generally more susceptible, have also developed resistance to several antibiotics, including

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β-lactams and tetracyclines. The persistence of resistant strains in food matrices increases the risk of their transmission to humans through the food chain (Punchihewage-Don *et al*., 2024; Galgano *et al*., 2025).

Assessment of the antimicrobial activity of antibiotics against foodborne pathogens is crucial for understanding current resistance trends and guiding appropriate therapeutic strategies (Gajic *et al*., 2022; Farrukh *et al*., 2025). In vitro susceptibility testing provides valuable insights into the effectiveness of antibiotics and helps in identifying emerging resistance patterns. Therefore, the present study aims to evaluate the antimicrobial properties of selected antibiotics against common bacterial food pathogens isolated from food sources (Yamin *et al*., 2023; Belay *et al*., 2024; Bayot *et al*., 2025). The findings of this study will contribute to a better understanding of antibiotic efficacy and support the development of informed policies for antibiotic usage in food safety and public health.

MATERIALS AND METHODS

Bacterial Strains

The bacterial food pathogens used in this study included *Escherichia coli*, *Salmonella enterica*, *Staphylococcus aureus*, *Listeria monocytogenes*, and *Bacillus cereus*. Standard reference strains were obtained from a recognized microbial culture collection and maintained on nutrient agar slants at 4 °C until use.

Antibiotics Tested

The antibiotics selected for this study were ampicillin (β-lactam), ciprofloxacin (fluoroquinolone), tetracycline (tetracycline class), chloramphenicol (phenicol), and gentamicin (aminoglycoside). These antibiotics represent different classes and are commonly used in clinical and veterinary medicine (Nhung *et al*., 2016; Afroze *et al*., 2025).

Preparation of Inoculum

Bacterial cultures were grown overnight in nutrient broth at 37 °C. The turbidity of the cultures was adjusted to match the 0.5 McFarland standard, corresponding to approximately 1×10^8 CFU/mL. Cultures were maintained on nutrient agar and sub-cultured prior to experimentation (Gayathiri *et al*., 2018; Bhrdwaj *et al*., 2025).

Agar Disc Diffusion Method

The antimicrobial activity of antibiotics was evaluated using the Kirby–Bauer disc diffusion method. Mueller-Hinton agar plates were inoculated with standardized bacterial suspensions using sterile cotton swabs. Antibiotic discs were placed on the surface of the agar and plates were incubated at 37 °C for 18–24 hours. Zones of inhibition were measured in millimeters and interpreted according to Clinical and Laboratory Standards Institute (CLSI) guidelines (Bauer *et al*., 1966; Bayot *et al*., 2024).

Agar Well Diffusion Method

Sterile wells of 6 mm diameter were punched into inoculated agar plates. Antibiotic solutions of known concentrations were added into the wells, and plates were incubated under the same conditions. The diameter of the inhibition zones was measured to assess antimicrobial activity (Balouiri *et al*., 2016).

Minimum Inhibitory Concentration (MIC)

The MIC of each antibiotic was determined using the broth dilution method. Serial dilutions of antibiotics were prepared in nutrient broth, inoculated with bacterial suspensions, and incubated at 37 °C for 24 hours. The lowest concentration showing no visible growth was recorded as the MIC (Kowalska-Krochmal *et al*., 2021).

Statistical Analysis

All experiments were performed in triplicate. Results were expressed as mean ± standard deviation. Statistical analysis was carried out using appropriate software, and differences were considered significant at $p< 0.05$.

RESULTS AND DISCUSSION

The results of the antimicrobial susceptibility tests revealed significant variations in the sensitivity of the tested foodborne pathogens to the selected antibiotics. Ciprofloxacin and gentamicin exhibited the highest antimicrobial activity against all tested bacteria, as indicated by larger zones of inhibition and lower MIC values. These findings are consistent with previous studies reporting the broad-spectrum efficacy of fluoroquinolones and aminoglycosides (Shariati *et al*., 2022). Table 1 below represents the zones of inhibitions shown by test antibiotics against the selected pathogens from foods.

Table 1. Zone of inhibition (mm) of antibiotics against foodborne pathogens.

Pathogen	AMP	CIP	TET	CHL	GEN
E. coli	10 ± 0.5	26 ± 1.2	12 ± 0.6	18 ± 0.9	24 ± 1.0
Salmonella	11 ± 0.7	25 ± 1.1	13 ± 0.8	17 ± 0.7	23 ± 0.8
S. aureus	18 ± 0.9	28 ± 1.4	20 ± 1.0	22 ± 1.1	26 ± 1.3
L. monocytogenes	17 ± 0.8	27 ± 1.3	19 ± 0.9	21 ± 1.0	25 ± 1.2
B. cereus	16 ± 0.6	26 ± 1.1	18 ± 0.7	20 ± 0.8	24 ± 1.0

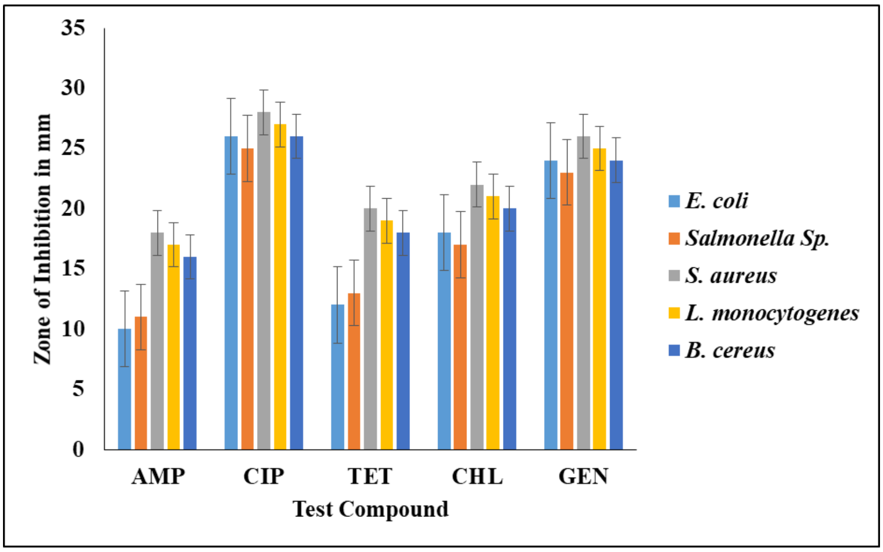

Figure 1. Zone of inhibition in mm for antibiotics against test foodborne pathogens.

The figure 1, illustrates the antimicrobial efficacy of five standard antibiotics ampicillin (AMP), ciprofloxacin (CIP), tetracycline (TET), chloramphenicol (CHL), and gentamicin (GEN) against selected bacterial food pathogens (*Escherichia coli, Salmonella* sp., *Staphylococcus aureus, Listeria monocytogenes,* and *Bacillus cereus*), expressed as zone of inhibition in millimeters. Among the tested compounds, ciprofloxacin exhibited the highest antibacterial activity across all pathogens, with inhibition zones ranging approximately from 25–28 mm, indicating its broad-spectrum and potent efficacy. Gentamicin also showed strong inhibitory effects (22–26 mm), followed by chloramphenicol (15–22 mm), suggesting moderate to high sensitivity of both Gram-positive and Gram-negative bacteria. Tetracycline demonstrated comparatively lower activity, particularly against *E. coli* and *Salmonella* sp., while ampicillin showed the least effectiveness overall, especially against Gram-negative strains. Notably, *S. aureus* and *L. monocytogenes* were generally more susceptible to most antibiotics than *E. coli* and *Salmonella* sp. The presence of error bars indicates experimental variability but does not alter the overall trend, highlighting ciprofloxacin and gentamicin as the most effective antibiotics against the selected foodborne pathogens.

Ciprofloxacin and gentamicin consistently produced the largest inhibition zones, indicating strong antimicrobial activity. Ampicillin and tetracycline showed reduced effectiveness, particularly against Gram-negative bacteria. Ampicillin showed limited effectiveness, particularly against Gram-negative bacteria such as *E. coli* and *Salmonella*, which may be attributed to the production of β-lactamase enzymes. Tetracycline resistance was observed in both Gram-positive and Gram-negative bacteria, reflecting its extensive use in agriculture and clinical settings. Chloramphenicol demonstrated moderate activity; however, its use has declined due to associated toxicity concerns (Kakoullis *et al.,* 2021; Sitohy *et al*., 2024). Table 2 represents the minimal inhibitory concentrations (MIC) against tested foodborne pathogens.

Table 2. MIC values (µg/mL) of antibiotics.

Pathogen	AMP	CIP	TET	CHL	GEN
E. coli	32	0.5	16	8	1
Salmonella	32	0.5	16	8	1
S. aureus	8	0.25	4	4	0.5
L. monocytogenes	8	0.25	4	4	0.5
B. cereus	8	0.5	4	4	1

Lower MIC values for ciprofloxacin and gentamicin confirm their superior efficacy.

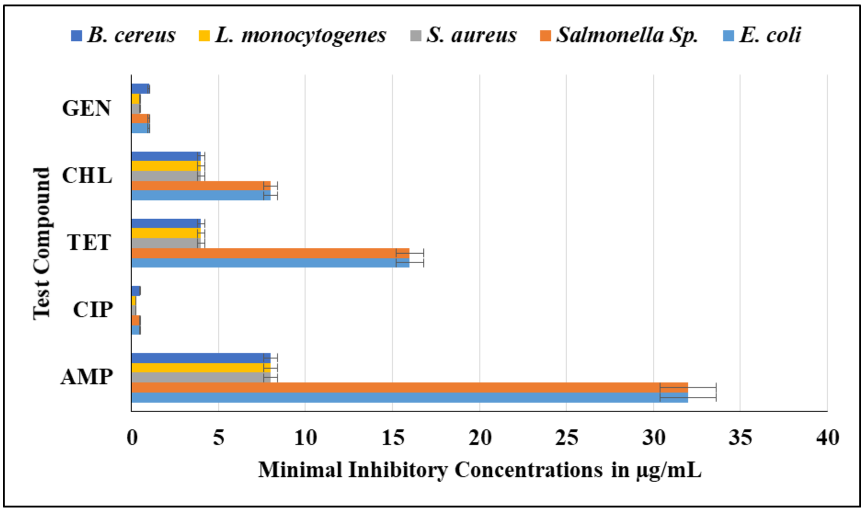

Figure 2. MIC values for test antibiotics against selected foodborne pathogens.

The figure 2, depicts the minimal inhibitory concentrations (MICs, µg/mL) of five antibiotics—ampicillin (AMP), ciprofloxacin (CIP), tetracycline (TET), chloramphenicol (CHL), and gentamicin (GEN)—against major foodborne pathogens (*Bacillus cereus, Listeria monocytogenes, Staphylococcus aureus, Salmonella* sp., and *Escherichia coli*). Ampicillin exhibited the highest MIC values (approximately 30–35 µg/mL), particularly against *Salmonella* sp. and *E. coli*, indicating comparatively lower antibacterial potency. Tetracycline showed moderate MICs, with *Salmonella* sp. and *E. coli* requiring higher concentrations (~15–17 µg/mL) than Gram-positive bacteria. Chloramphenicol demonstrated improved efficacy with lower MICs (around 6–10 µg/mL), reflecting broader activity across both Gram-positive and Gram-negative strains. Ciprofloxacin and gentamicin displayed the lowest MIC values (generally <2 µg/mL) for all tested organisms, highlighting their superior antibacterial effectiveness. Overall, the MIC trends corroborate the strong potency of ciprofloxacin and gentamicin, while ampicillin appears least effective against the selected food pathogens. Gram-positive bacteria, especially *Staphylococcus aureus* and *Bacillus cereus*, were generally more susceptible to the tested antibiotics compared to Gram-negative bacteria. The structural differences in cell walls likely contribute to this observed variation. The presence of resistant strains among foodborne pathogens underscores the need for judicious antibiotic use and regular surveillance (Karaman *et al*., 2020; Alhumaid *et al*., 2021).

The differential susceptibility patterns observed in this study align with earlier reports on foodborne pathogens. The reduced effectiveness of ampicillin may be attributed to β-lactamase production, while tetracycline resistance likely reflects its extensive use in agriculture. Gram-positive bacteria showed higher susceptibility due to simpler cell wall structures, whereas Gram-negative bacteria exhibited increased resistance due to outer membrane barriers. These findings reinforce the urgent need for antimicrobial stewardship and continuous resistance monitoring in food safety systems (Mwansa *et al*., 2022; Grudlewska-Buda *et al*., 2023; Wojnarowski *et al*., 2025). The findings highlight the importance of continuous monitoring of antibiotic resistance patterns in food-associated bacteria to prevent the spread of resistant strains through the food chain.

CONCLUSION

The present study demonstrates that selected antibiotics exhibit varying degrees of antimicrobial activity against common bacterial food pathogens. Ciprofloxacin and gentamicin were the most effective antibiotics, while resistance to ampicillin and tetracycline was evident in several bacterial strains. The emergence of antibiotic-resistant foodborne pathogens poses a serious threat to public health and emphasizes the need for responsible antibiotic usage and robust food safety practices. Regular surveillance and susceptibility testing are essential to guide effective treatment strategies and minimize the risk of resistance development.

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CONFLICT OF INTERESTS

The authors declare no conflict of interest

ETHICS APPROVAL

Not applicable

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AI TOOL DECLARATION

The authors declares that no AI and related tools are used to write the scientific content of this manuscript.

DATA AVAILABILITY

Data will be available on request

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