**Meat Research**
Vol 5, Issue 2: 1-8, Article 114, April 2025
ISSN: 2790-1971
https://doi.org/10.55002/mr.5.2.114
http://www.bmsa.info/meatresearch

[1]Department of Animal Science,
Bangladesh Agricultural University,
Mymensingh-2202, Bangladesh;

***Corresponding Author:**
**MA Hashem**
E-mail: hashem_as@bau.edu.bd

**Keywords:**
Antibiotics
Beef Cattle
Bangladesh
Organ
Residues

**Article Info:**
Received: 11 March 2025
Accepted: 13 April 2025
Published online: 30 April 2025

**Research Article**

# Assessment of antibiotic residues in beef cattle slaughtered in bangladesh: Implications for food safety and public health

MT Kamal[1], RA Deen[1], B Ahmed[1], MA Hashem[1]*

## Abstract

This study investigates the concentrations of tetracycline, ciprofloxacin, and enrofloxacin residues in various tissues of beef cattle post-slaughter in Bangladesh, emphasizing their implications for food safety and public health. Residue levels were analyzed across heart, kidney, liver, lung, and muscle tissues, utilizing high-sensitivity detection methods. The findings reveal that Tetracycline is present in all tested tissues, with the highest concentration observed in heart tissue (109.68 ppb) and the lowest in the liver (55.19 ppb), all remaining below the international maximum residue limits (MRLs). Ciprofloxacin was detected primarily in kidney and liver tissues, with concentrations substantially below the detection limits in heart, lung, and muscle. Enrofloxacin showed the highest residue levels in muscle tissues (45.36 ppb) and was undetectable in kidney and liver. These results highlight effective antibiotic residue management in the evaluated tissues, with all concentrations safely below MRLs, ensuring compliance with global food safety standards. However, the observed presence of these antibiotics, particularly the variations in residue levels, underscores the need for stringent monitoring and regulation to prevent potential health risks associated with antibiotic resistance and hypersensitivity reactions. This study contributes to the ongoing efforts to safeguard public health by providing a comprehensive evaluation of antibiotic residues in beef, thereby reinforcing the importance of adherence to pharmacological guidelines and regulatory standards in livestock management.

## Introduction

Livestock plays a vital role in the economy of Bangladesh, contributing significantly not only to the agricultural sector but also to national income, employment, and food security. According to the Department of Livestock Services (DLS) in Bangladesh, the contribution of the livestock sector to overall GDP has been provisionally estimated at 1.85% and the growth rate of the livestock sector was 3.23% during 2022–23. This sector is an integral part of the rural economy and is pivotal in supporting the livelihoods of millions of smallholder farmers and landless laborers (Haque et al., 2023). As Bangladesh continues to experience robust economic growth, the livestock sector emerges as a critical component in sustaining and accelerating this progress. About 80 to 85% of the households keep livestock in rural areas and most of them are landless, marginal, and small farmers (Bhuiyan et al., 2016). Livestock may be considered as "cash income" to rural farmers that is instantly available for sale (Hossen, 2008). Beef cattle production has become an important business for small farmers. Growth performance in various indigenous and regional breeds, such as the Jamuna basin lambs, has demonstrated the economic potential of livestock if managed efficiently (Hashem et al., 2020). Now it is necessary to find out the limitations of the existing beef production system to make it more sustainable at the farmer's level.

Cattle fattening has become an increasingly vital component of the agricultural sector in Bangladesh, playing a crucial role in meeting the country's escalating meat demand. As the population grows and incomes rise, the appetite for meat, particularly beef, continues to surge. This has positioned cattle fattening as a key industry not only for food security but also for economic prosperity. Traditionally aligned with major cultural and religious festivities, where the demand for meat spikes dramatically, cattle fattening ensures a stable supply of beef to accommodate these peaks in consumption. Over the years, the practice has evolved from small-scale, traditional methods to more intensive fattening operations, highlighting its growing importance in the agricultural landscape of Bangladesh. The expansion of this sector is instrumental in stabilizing meat prices and providing livelihood opportunities for many, including smallholder farmers and agricultural entrepreneurs. Consumer expectations for meat quality and safety are also increasing, leading researchers and producers to explore novel natural additives like ginger and tulsi extracts for improving meat preservation and sensory acceptability (Hossain et al., 2021; Siddiqua et al., 2018). As such, cattle fattening is not only about satisfying the immediate meat needs of the population but also about fostering sustainable economic growth and food sovereignty in Bangladesh (Ferdush et al., 2023).

The application of antibiotics in cattle fattening in Bangladesh ranges from therapeutic to non-therapeutic uses, including disease prevention and growth promotion. While these practices can improve feed efficiency and growth rates, leading to increased meat production, they come with substantial risks (Kasimanickam et al., 2021).

The indiscriminate or unregulated use of antibiotics is rampant, primarily due to the lack of stringent enforcement of veterinary drug use regulations and a widespread lack of awareness among farmers about the consequences of antibiotic misuse. Many farmers, especially in rural or less developed areas, may not be fully aware of the required withdrawal periods for antibiotics—the time required to elapse between the last antibiotic administration and the slaughter of the animal, ensuring that no residual drugs remain in the meat. Non-compliance with these withdrawal periods can lead to antibiotic residues in the beef, posing health risks to consumers. Moreover, post-harvest processing and storage practices—including innovations like gamma irradiation—have shown potential in reducing microbial load and extending shelf life, but they must be aligned with broader food safety strategies (Haquea et al., 2017).

The assessment of these residues is crucial, for not only ensuring the safety and quality of meat consumed by the public but also for addressing broader issues related to food safety, antibiotic resistance, and public health (Arsene et al., 2022). However, this practice often leads to the presence of antibiotic residues in meat, which can persist beyond the slaughter and processing stages (Manyi-Loh et al., 2018). The consumption of such contaminated meat poses health risks including the potential development of antibiotic-resistant bacteria among consumers. This resistance can undermine the effectiveness of antibiotics, making infections harder to treat and increasing medical costs and morbidity. Moreover, the overuse and misuse of antibiotics in livestock can contribute to the development of antibiotic-resistant bacteria, a severe threat that undermines the efficacy of these vital medications (Salam et al., 2023).

The situation is exacerbated by limited access to veterinary services and reliable information on proper antibiotic usage. Often, farmers depend on advice from unqualified or poorly informed intermediaries for veterinary drugs, including antibiotics. This scenario not only leads to improper dosages and inappropriate antibiotic choices but also heightens the risk of drug resistance (Phillips et al., 2004). Given these issues, there is an urgent need for comprehensive strategies to improve antibiotic stewardship in cattle fattening practices in Bangladesh. This includes enhancing regulatory frameworks, providing education and training for farmers on responsible antibiotic use, and improving surveillance and monitoring systems to ensure compliance with drug withdrawal periods (Cole et al., 2024).

Addressing these challenges is crucial for ensuring public health safety, maintaining animal health and welfare, and securing the sustainability of the livestock sector in Bangladesh. Furthermore, advances in meat science—such as the integration of spectroscopy and artificial intelligence—offer promising tools for rapid, non-invasive meat quality assessment, which could help detect safety and quality issues like antibiotic residues more efficiently (Sarker et al., 2024a). Studies also suggest that factors like post-rigor processing (Khatun et al., 2025), water-holding capacity, and muscle histomorphology (Sarker et al., 2024b) significantly influence sensory attributes and tenderness, which are key quality markers for consumer acceptance. These insights are critical as Bangladesh seeks to modernize its meat production standards in line with global expectations.

This study is critically important as it investigates the presence of antibiotic residues—specifically tetracycline, ciprofloxacin, and enrofloxacin—in various tissues of beef cattle in Bangladesh, an issue of growing concern in public health due to its implications for antibiotic resistance, identified by the World Health Organization as a major global health threat. With the increasing consumption of meat in Bangladesh and lax regulatory oversight on antibiotic use in livestock, this study aims to quantify the residue levels of these antibiotics in edible tissues such as the heart, kidney, liver, lung, and muscle to assess the compliance with safe consumption levels and recommended withdrawal periods. Furthermore, the study evaluates the effectiveness of the High-Performance Liquid Chromatography (HPLC) techniques employed, using a Hewlett Packard (HP) 1050 multi-solvent system, ensuring precise and accurate measurements crucial for assessing risks associated with antibiotic residues. The objectives are to determine the concentrations of specific antibiotics in beef tissues, evaluate the efficacy of current extraction and detection methods, assess the risks these residues pose to consumers, particularly the risk of developing antibiotic-resistant infections, and provide robust data to inform and influence policymakers in Bangladesh to enhance food safety standards. By doing so, this study seeks to safeguard consumer health and guide improvements in livestock management practices.

## Materials and Methods

### Ethical approval

Ethical approval for conducting animal research was not necessary for this study, as it did not involve live animals. Instead, meat samples were purchased from government-approved slaughterhouses, where the animals had already been slaughtered.

### Experimental analysis location

In this study, we focused on beef cattle from three major slaughterhouses in Dhaka, Bangladesh. To ensure the integrity of our samples, we collected 100 grams of raw beef from various tissues – heart, kidney, liver, lung, and muscle – early in the morning and within 8 hours of slaughter. This timing was strategically chosen to reduce microbial contamination that could escalate due to environmental temperatures throughout the day. For the analysis, we precisely measured 10 grams of each meat sample and placed them into sterile flasks with 90 ml of distilled water. The samples were then thoroughly homogenized using a pestle and mortar under aseptic conditions to ensure cleanliness and prevent any external contamination. These prepared samples were subsequently analyzed at the National Food Safety Laboratory (NFSL), which operates under the Institute of Public Health in Dhaka, Bangladesh. The analysis was conducted using High-Performance Liquid Chromatography (HPLC), adhering to the methodologies established by (Anadon et al. 1995; García Ovando et al. 2004). This laboratory setting was chosen for its advanced facilities and expertise in handling food safety analyses, ensuring precise and reliable measurement of the antibiotic residues in the cattle tissues.

### Apparatus

For the HPLC analysis in our study, we employed a comprehensive setup consisting of a Hewlett Packard (HP) 1050 multisolvent delivery system. This system was outfitted with an HP 1050 fluorescence detector, which is integral for detecting and quantifying the fluorescence of our analytes under specific light wavelengths. The detection process was further refined using an HP

Integrator, which was utilized to record the peak ratios – a critical measurement in HPLC that helps in identifying and quantifying substances based on their chromatographic retention time and peak shape. Chromatographic separation, a core process in HPLC, was conducted using a reverse-phase Phenomenex aqua 5μ C18 column. The dimensions of this column are 150 x 4.60 mm with particle sizes of 5 micrometers, which is ideal for achieving excellent separation with high resolution. To enhance the durability and maintain the integrity of the analytical column, a Phenomenex security guard column C18 ODS (octadecyl silane) was incorporated. This guard column, measuring 4 mm in length and 30 mm in internal diameter, plays a vital role in protecting the main column from potential contaminants that could compromise its performance and shorten its usable life.

### Reagents and materials

(a) Antimicrobial Standards: The antibiotics tested included ciprofloxacin, enrofloxacin, and tetracycline, all of which were sourced from ASPO Lab in Bangladesh. These standards are essential for establishing the reference points needed to quantify the antibiotic residues in the sample tissues.

(b) Water: The water used throughout the experiments was distilled and deionized to remove any impurities or ions that could interfere with the results. Additionally, it was filtered through a 0.45 μm filter to ensure it was free from any particulate matter, providing a clean and controlled medium for the experiments.

(c) Buffer Solution: A phosphate buffer solution with a concentration of 0.1M and a pH of 7.2 was prepared. This buffer helps maintain a consistent pH during the analysis, which is crucial for the stability and solubility of the antibiotics under investigation.

(d) Working Standard Solutions: These solutions were meticulously prepared from the stock standards of the antibiotics. Dilutions were made to achieve concentrations of 0.00375, 0.0075, 0.015, 0.03, 0.06, and 0.1 μg/mL for each of ciprofloxacin, enrofloxacin, and tetracycline. These dilutions are used to create calibration curves that help in accurately determining the antibiotic concentrations in the sample extracts.

(e) Solvents: High-purity solvents including Dichloromethane, Acetonitrile, and Triethylamine – all of HPLC grade – were used. Dichloromethane acts as an organic solvent to help extract antibiotics from the tissue samples effectively. Acetonitrile is utilized in the mobile phase for its excellent elution properties, and Triethylamine is used to adjust the pH of the mobile phase, optimizing the separation of antibiotic compounds during HPLC analysis.

### Chromatography conditions

For the chromatographic analysis, the mobile phase consisted of a mixture of water, acetonitrile, and triethylamine, blended in an 80:19:1 ratio. This mixture was then adjusted to a pH of 3.0 using phosphoric acid to optimize the separation conditions. Prior to use in experiments, the mobile phase was meticulously filtered through a 0.45μm nylon membrane to remove any particulates that could interfere with the analysis. The flow rate of the mobile phase through the chromatography system was set at 1.2 mL per minute. Detection of the antimicrobials – ciprofloxacin, enrofloxacin, and balofloxacin – was conducted using a fluorescence detector. The settings for the fluorescence detection were carefully chosen to ensure precise measurement: the excitation wavelength was set at 295 nm and the emission wavelength was at 500 nm. Additional settings on the detector included using lamp 3, setting the PMT (photomultiplier tube) gain to 9, and a response time of 6. These specific conditions were critical as they allowed for the accurate and reliable detection of the three antimicrobials in the samples analyzed. This meticulous setup ensured that each compound was properly identified and quantified without interference from other substances in the samples.

### Extraction Procedure

The extraction process began with homogenizing 0.2 grams of muscle tissue in 2 mL of phosphate buffer. To this homogenate, 8 mL of dichloromethane was added, and the mixture was vortexed for one minute and then centrifuged at 4000 rpm for 20 minutes. Following centrifugation, the upper aqueous layer was removed and discarded, allowing the organic phase to be carefully transferred into a clean tube. The muscle tissue residue was then subjected to a second extraction with an additional 6 mL of dichloromethane to ensure comprehensive recovery of the analytes. After the second extraction, the organic layers from both extractions were combined. This combined organic phase was then evaporated under a stream of nitrogen at a temperature of 30°C to remove the dichloromethane, leaving behind the concentrated extract.

The dry residue was subsequently redissolved in 200 µL of the mobile phase appropriate for HPLC analysis. From this solution, 100 µL was taken and injected into the HPLC system for analysis. The quantification of the antimicrobial agents per gram of muscle tissue was based on the comparison of the peak areas obtained from the HPLC analysis with those of a pre- established standard curve. This standard curve relates known concentrations of the antimicrobial agents to their respective HPLC response, facilitating accurate measurement of the unknown samples.

### Standard Curve and Linearity

To establish a standard curve for assessing linearity, homogenized muscle tissue samples were spiked with antimicrobials at concentrations of 0.01, 0.03, 0.06, and 0.1 μg/g. Following the spiking, these samples underwent the extraction process as previously outlined. The analytical results were represented by plotting the peak areas of ciprofloxacin, enrofloxacin, and tetracycline against their corresponding concentrations to evaluate linearity.

The limit of quantification was defined as the lowest concentration of the antimicrobials that could be reliably quantified, with an acceptable signal-to- noise ratio of 10. This ratio was calculated as ten times the standard deviation of the blank samples divided by the slope of the standard curve. The limit of detection on the other hand, was established as the lowest concentration at which antimicrobials could be detected with a signal-to-noise ratio of three. This method ensures that even the smallest detectable quantities are measured with adequate confidence, reflecting the sensitivity and precision of the analytical method used.

### Recovery

To assess the recovery efficiency of the analytical method, working standard solutions of ciprofloxacin, enrofloxacin, and tetracycline were added to re- dissolved extracts of blank muscle samples that had been processed according to the previously described extraction procedure. The concentrations of the standards added were 0.03, 0.01, and 0.1 µg/mL, respectively. These concentrations were assumed to represent 100% of the antimicrobial concentration in the muscle samples. The recovery of each antimicrobial was then calculated by comparing the observed antimicrobial peak areas from the HPLC analysis to the theoretical peak areas (representing 100% recovery). The reduction in peak areas from these added standards in the muscle samples, relative to the expected concentrations, was quantified. The percentage of recovery was expressed as the ratio of the observed peak area to the theoretical (100% recovery) peak area, multiplied by 100 to convert this ratio into a percentage. This process measures the efficiency and accuracy with which the antimicrobial residues can be recovered from muscle tissues using the extraction and analysis method described.

### Statistical Analysis

The collected data were statistically analyzed using IBM SPSS Statistics software v.20 (Corp. 2020). For each set of samples, which were processed in triplicate, the mean values and standard deviations were calculated to assess central tendency and variability, respectively. To determine the statistical significance of the observed differences among the various sample groups, an ANOVA was performed. A significance level of $p < 0.05$ was adopted, meaning that differences with a p-value less than 0.05 were considered statistically significant, indicating a high likelihood that observed differences were not due to random variation. Furthermore, to delve deeper into the specific differences between group means, Duncan's multiple-range test was applied. This post- hoc comparison was utilized to identify which specific groups differed from each other after the initial ANOVA showed significant results. This approach allowed for a detailed comparison and provided a clear understanding of the distribution and relationships among the sample means.

## Results

### Tetracycline concentration in organs and muscles of beef

The current study demonstrated varying levels of tetracycline residue concentrations across different organs and muscles of cattle meat in Bangladesh (Table 1). The heart tissue displayed the highest average concentration of tetracycline at 109.68 ppb, with a relatively narrow range between the minimum value of 98.53 ppb and the maximum of 115.38 ppb. This suggests a consistent presence of tetracycline across the heart samples analyzed. Despite the high residue level compared to other tissues, there is no MRL currently set for heart tissue, making it difficult to assess the compliance of these levels with regulatory standards. For the kidney samples, the average tetracycline concentration was recorded at 78.20 ppb. The range of concentrations from 65.53 ppb to 87.39 ppb indicates variability but remains significantly below the MRL of 600 ppb set by Commission Regulation (EU) No. 37/2010. This suggests that kidney tissues while showing accumulation of tetracycline, are well within safe consumption limits. Liver tissues exhibited the lowest average concentration of tetracycline at 55.19 ppb, with the values spanning from 46.92 ppb to 63.02 ppb. Similar to the kidneys, the liver's tetracycline concentrations are considerably below the regulatory limit of 300 ppb. This low level of residue indicates effective management of antibiotic use in terms of liver safety thresholds. The lung samples showed more variation in tetracycline levels, averaging 100.72 ppb. The concentration ranged widely from 81.93 ppb to 128.98 ppb. This variation might reflect different levels of exposure or metabolism of tetracycline in lung tissues. No MRL is specified for lung tissue, which complicates the evaluation of these findings against safety benchmarks.

**Table 1.** Tetracycline concentration in organs and muscle of beef cattle

| Sample Name | No. of Samples | Mean ± SD (ppb) | Minimum (ppb) | Maximum (ppb) | MRL (ppb) |
|---|---|---|---|---|---|
| **Heart** | **3** | 109.68 ± 9.66 | 98.53 | 115.38 | - |
| **Kidney** | **3** | 78.20 ± 11.4 | 65.53 | 87.39 | 600* |
| **Liver** | **3** | 55.19 ± 8.06 | 46.92 | 63.02 | 300* |
| **Lung** | **3** | 100.72 ± 24.92 | 81.93 | 128.98 | - |
| **Muscle** | **3** | 69.55 ± 11.59 | 57.68 | 80.83 | 100* |
| ***p*-value** | | 0.059 | | | |

ppb, parts per billion; *, Commission Regulation (EU) No. 37/2010; MRL, maximum residue limit

Muscle tissue, often a critical point of inspection for meat consumption, showed an average tetracycline concentration of 69.55 ppb, with a range from 57.68 ppb to 80.83 ppb. This is under the set MRL of 100 ppb, indicating that the muscle tissues are safe for consumption with respect to tetracycline residues. The statistical analysis of the data across these various tissues resulted in a p- value of 0.059. This value, while close to the conventional threshold of 0.05 for statistical significance, suggests that the observed differences in tetracycline concentrations among the tissues are not statistically significant. These results highlight that all measured concentrations of tetracycline in the tissues of beef cattle remain well below the respective safety limits, affirming the effective regulation and management of antibiotic use in the beef industry. This provides reassurance about consumer safety and points towards adherence to pharmacological guidelines in livestock management.

### Ciprofloxacin Concentration in Organs and Muscles of Beef

The study conducted on beef cattle assessed the concentrations of ciprofloxacin across various tissues, revealing that this antibiotic was detectable in only the kidney and liver, while concentrations in the heart, lung, and muscle tissues remained below the detection limit (Table 2). Specifically, the ciprofloxacin concentration in the kidney tissue averaged 8.28 ppb, with a range extending up to 14.63 ppb. The variation among kidney samples, indicated by a standard deviation of 7.50 ppb, suggests some inconsistencies in antibiotic residue levels, potentially due to differences in drug metabolism or tissue deposition among the individual cattle tested. Liver tissues exhibited a notably higher concentration of ciprofloxacin, with an average of 34.81 ppb and a maximum concentration of 48.20 ppb, marked by a standard deviation of 15.03 ppb. This indicates a higher accumulation of the antibiotic in the liver compared to the kidney and shows significant variability among the samples. The higher levels in the liver could be attributed to the organ's role in metabolizing and clearing medications, including antibiotics.

The absence of detectable ciprofloxacin in the heart, lung, and muscle tissues indicates either very low penetration of the drug into these tissues or efficient clearance that keeps residue levels below measurable thresholds. This finding is significant as it suggests limited exposure to the drug through the consumption of these particular beef tissues. The statistical analysis yielded a p-value of 0.087, suggesting that the differences in ciprofloxacin concentrations between the detectable tissues (kidney and liver) are not statistically significant at the conventional alpha level of 0.05. However, the proximity of the p-value to the threshold indicates a trend that might warrant further investigation with a larger sample size to better understand the distribution patterns of ciprofloxacin in beef cattle.

**Table 2.** Ciprofloxacin concentration in organs and muscle of beef cattle

| Sample Name | No. of Sample | Mean ± SD (ppb) | Minimum (ppb) | Maximum (ppb) |
|---|---|---|---|---|
| **Heart** | **3** | BDL | BDL | BDL |
| **Kidney** | **3** | 8.28 ± 7.50 | BDL | 14.63 |
| **Liver** | **3** | 34.81 ± 15.03 | 18.56 | 48.20 |
| **Lung** | **3** | BDL | BDL | BDL |
| **Muscle** | **3** | BDL | BDL | BDL |
| ***p*-value** | | 0.087 | | |

ppb, parts per billion; BDL, below detection limit

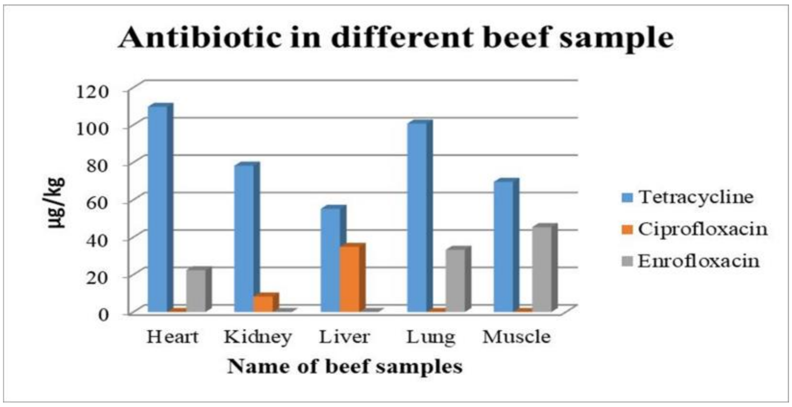


**Figure 1.** Antibiotics in different organs of beef cattle.

## Enrofloxacin Concentration in Organs and Muscle of Beef Cattle

The study detailed in Table 3 investigated the concentration of enrofloxacin in various organs and muscle tissues of beef cattle. Notably, enrofloxacin was detectable in the heart, lung, and muscle tissues but remained below detection limits in both kidney and liver samples. In the heart, the average concentration was 22.35 ppb, though the spread was considerable (standard deviation of 19.95 ppb), ranging from undetectable levels to a maximum of 38.37 ppb. This variation could suggest differences in how enrofloxacin accumulates or is metabolized in heart tissue among individual cattle. Lung tissues showed a more consistent presence of enrofloxacin, with a mean concentration of 33.23 ppb and a narrower standard deviation of 4.02 ppb. The values ranged from 28.70 ppb to 36.38 ppb, indicating a more stable distribution across the sampled lung tissues. Muscle tissues exhibited the highest concentrations of enrofloxacin, with an average of 45.36 ppb. The variability in muscle tissues, denoted by a standard deviation of 9.73 ppb, spanned from 36.92 ppb to 56.00 ppb, again highlighting potential differences in drug absorption or elimination.

The absence of detectable enrofloxacin in the kidney and liver is significant, suggesting either very effective clearance mechanisms in these organs or limited permeability of the drug to these tissues. The observed data generated a p-value of 0.070, which indicates that while there are observable differences in enrofloxacin concentrations among the tissues where it was detectable, these differences are not statistically significant at the conventional threshold of 0.05. However, the proximity of the p-value to this threshold suggests a potential trend that might benefit from further investigation, possibly with an increased sample size or additional testing methods to confirm consistency and ensure comprehensive safety assessments in line with pharmacological standards. This study underscores the importance of monitoring antibiotic residues in edible animal tissues, critical for food safety and regulatory compliance.

**Table 3.** Enrofloxacin concentrations in organs and muscle of beef cattle

| Sample Name | No. of Sample | Mean ± SD (ppb) | Minimum (ppb) | Maximum (ppb) |
|---|---|---|---|---|
| **Heart** | **3** | 22.35 ± 19.95 | BDL | 38.37 |
| **Kidney** | **3** | BDL | BDL | BDL |
| **Liver** | **3** | BDL | BDL | BDL |
| **Lung** | **3** | 33.23 ± 4.02 | 28.70 | 36.38 |
| **Muscle** | **3** | 45.36 ± 9.73 | 36.92 | 56.00 |
| ***p*-value** | | 0.070 | | |

ppb, parts per billion; BDL, below detection limit

The summarized data in Table 4 provides crucial insights into the residue levels of enrofloxacin and ciprofloxacin combined in various beef cattle tissues and their compliance with established MRLs as set by Commission regulation (EU) No. 37/2010. The total concentrations reported for these antibiotic residues were found to be significantly below the MRL in all tissues with set limits, underscoring compliance with regulatory standards aimed at ensuring food safety. Specifically, the heart and lung tissues, for which no specific MRLs are set, recorded residue levels of 22.35 ppb and 33.23 ppb respectively. These values, while unregulated, are lower compared to the highest MRLs set for other tissues, indirectly suggesting safety in consumption. The kidney tissues, which have a relatively high MRL of 200 ppb, showed a very low residue level of 8.28 ppb, well within safe limits. Similarly, liver tissues displayed a residue amount of 34.81 ppb, also safely below the MRL of 300 ppb, confirming the effective management of antibiotic use in these animals.

Most critically, muscle tissues, which are the primary concern for direct human consumption, recorded a residue level of 45.36 ppb. This level is also significantly lower than its MRL of 100 ppb, confirming that the muscle tissue of these cattle is safe for consumption for the residues of these specific antibiotics. These findings indicate that the current management practices regarding the use of enrofloxacin and ciprofloxacin in beef cattle are effective in keeping residue levels within the safety thresholds set by regulatory bodies. This is paramount not only for consumer health but also for maintaining public confidence in agricultural and food safety standards.

Figure 1 illustrates the distribution of antibiotics across various organs and muscle tissues in cattle, including the heart, kidneys, liver, lungs, and muscles. The data reveal that tetracycline is present in all sampled organs of beef cattle, exhibiting higher concentrations compared to those of enrofloxacin and ciprofloxacin. This suggests that tetracycline accumulates more extensively within these tissues, underscoring its prevalent use and potentially different pharmacokinetics compared to the other studied antibiotics.

## Discussion

The presence of antibiotic residues such as tetracycline, ciprofloxacin, and enrofloxacin in beef cattle post-slaughter is a matter of significant concern due to their potential impact on human health and food safety. Antibiotics are among the principal veterinary pharmaceuticals used extensively in the agricultural sector, posing a significant risk of food contamination. The residual presence of these drugs in meat products can lead to a spectrum of adverse health effects that underscore the critical need for rigorous food safety protocols. The residues of antibiotics in animal-derived food products, particularly meat, are associated with several health risks. These include the emergence and proliferation of multidrug- resistant bacteria, which are a growing public health crisis (Kjeldgaard et al. 2012; Chang et al. 2015). These resistant strains complicate the treatment of infections, often rendering standard antibiotics ineffective. Furthermore, antibiotic residues can trigger allergic and potentially life-threatening anaphylactic reactions in susceptible individuals (Baynes et al. 2016). Another significant health concern is the disruption of the gut microbiome, which plays a crucial role in digestion, immunity, and overall health (Cotter et al. 2012). Additionally, the risk of horizontal gene transfer, where resistance genes are transferred from animal microbiomes to human pathogens, poses a direct threat to human health (Marshall & Levy 2011). Tetracyclines, due to their broad-spectrum activity, are extensively used in livestock for disease prevention, treatment, and growth promoters (Scaria et al. 2021). These compounds are administered through various routes, including orally via feed or water, parenterally, or through intramammary infusion. Their pharmacokinetic properties, particularly enterohepatic recirculation, allow small amounts of these drugs to persist in animal tissues for extended periods post- administration, increasing the risk of human exposure through meat consumption (Fletouris 2000). Our study's findings align with and expand upon those from previous research. For example, (Zhang et al. 2021) observed relatively lower maximum concentrations of key antibiotics in cattle muscle compared to our data. Notably, the highest tetracycline concentration we detected exceeded findings from other regions such as Saudi Arabia (El- Ghareeb et al. 2019). Such comparisons are essential as they highlight regional variations in antibiotic usage and residue levels, which could be influenced by local regulatory environments, farming practices, and enforcement of withdrawal periods. The variability in residue levels found in international studies suggests inconsistencies in regulatory compliance and differences in veterinary practices across regions. For instance, studies from India (Verma et al. 2021) and Tanzania (Mgonja et al. 2017) reported a significant presence of tetracycline in meat products, contrasting with our findings where lower concentrations were observed. Such discrepancies necessitate stringent and uniformly enforced regulations to ensure public safety and trust in food systems (Mohammed et al. 2022).

Moreover, setting and adhering to strict maximum residue limits as mandated by entities such as the European Union (Commission Regulation (EU) No. 37/2010) is crucial. Our results, which show residue levels below these limits, indicate compliance but also point toward the need for continuous monitoring to preempt any public health risks. The diversity in the incidence rates of antibiotic residues, as noted in studies from countries like Nigeria (Ibrahim et al. 2010) and Indonesia (Sanz et al. 2015; Widiastuti et al. 2022) compared to Spain (Nchima et al. 2017), reflects varying levels of regulatory control and surveillance efficacy. These findings argue strongly for enhanced international cooperation in monitoring and standardization practices to manage antibiotic use in livestock more effectively. The presence of antibiotic residues in meat products represents a significant challenge to public health that requires coordinated global efforts. Enhanced surveillance, stricter regulatory frameworks, and increased public awareness are essential to mitigate the risks associated with antibiotic residues in meats. Furthermore, fostering a deeper understanding of the implications of antibiotic use in livestock through ongoing research will help refine strategies to tackle this complex issue effectively.

## Conclusions

This study on antibiotic residues in post-slaughter beef cattle in Bangladesh reveals significant insights into antibiotic management within the agricultural sector. We observed varying levels of tetracycline across different tissues, with the highest in the heart and the lowest in the liver, yet all levels remained well below international maximum residue limits, indicating effective regulation and compliance. Additionally, ciprofloxacin was primarily detected in the kidneys and liver, while enrofloxacin was more evenly distributed across various tissues. The absence of detectable levels in some tissues suggests efficient drug clearance, minimizing health risks from consumption. The study underscores the safety of Bangladeshi beef, aligning with stringent pharmacological guidelines and supporting the continuation of its international market presence. However, the absence of established residue limits tissues points to a need for more comprehensive regulatory standards and further research to fully understand antibiotic pharmacokinetics in cattle. Moving forward, maintaining low residue levels will require enhanced surveillance, ongoing education for those in the livestock industry, and further studies to refine management practices. Overall, this research provides a solid foundation for future efforts to ensure the safety and integrity of beef products in Bangladesh, addressing current gaps, and enhancing antibiotic use strategies to protect public health and support sustainable industry practices.

## Acknowledgments

Authors are grateful to IFEL, UGC and MOST for funding this research.

## Conflicts of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## References

Anadon A, Martinez-Larranaga MR, Diaz MJ, Bringas P, Martinez MA, Fernandez-Cruz ML, Fernandez MC. Fernandez R. 1995. Pharmacokinetics and residues of enrofloxacin in chickens. American Journal of Veterinary Research, 56(4):501-506. https://doi.org/10.2460/ajvr.1995.56.04.501

Arsene MMJ, Davares AKL, Viktorovna PI, Andreevna SL, Sarra S, Khelifi I, Sergueievna DM. 2022. The public health issue of antibiotic residues in food and feed: Causes, consequences, and potential solutions. Veterinary World, 15(3): 662-671. https://doi.org/10.14202/vetworld.2022.662-671

Baynes RE, Dedonder K, Kissell L, Mzyk D, Marmulak T, Smith G, Tell L, Gehring R, Davis J, Riviere JE. 2016. Health concerns and management of select veterinary drug residues. Food and Chemical Toxicology, 88(4): 112-122. https://doi.org/10.1016/j.fct.2015.12.020

Bhuiyan KJ, Hashem MA, Hossain MM, Hossain MD. 2016. Organic beef cattle production pattern at Shahjadpur upazilla of Sirajgonj district in Bangladesh. Bangladesh Journal of Animal Science, 45(1): 25-30. https://doi.org/10.3329/bjas.v45i1.27484

Chang Q, Wang W, Regev-Yochay G, Lipsitch M, Hanage WP. 2015. Antibiotics in agriculture and the risk to human health: how worried should we be? Evolutionary Application, 8(3): 240-247. https://doi.org/10.1111/eva.12185

Cole J, Mughal AN, Eltholth M, Thomas A, Holmes M. 2024. Transdisciplinary approaches to addressing factors that influence antimicrobial use in dairy cattle: A scoping review. Heliyon, 10(4):1-11. https://doi.org/10.1016/j.heliyon.2024.e25550

Corp. I. 2020. IBM SPSS Statistics for Windows (Version 27.0) [Computer software]. IBM Corp.

Cotter PD, Stanton C, Ross RP, Hill C. 2012. The impact of antibiotics on the gut microbiota as revealed by high throughput DNA sequencing. Discovery Medicine, 13(70):193-199.

El-Ghareeb WR, Mulla ZS, Meligy AMA, Darwish WS, Edris AM. 2019. Antibiotic Residue Levels in Camel, Cattle and Sheep Tissues Using Lc-Ms/Ms Method. Journal of Animal and Plant Sciences-Japs, 29(4): 943-952.

Ferdush J, Hossain M, Hasan M, Khatun R, Sadrul S, Rahman M, Touhid M, Islam S. 2023. Economics of Cattle Fattening - A Case of Bangladesh. Qeios, pp: 1-22. https://doi.org/10.32388/whwlts.2

Fletouris DJ. 2000. Drug Residues in Foods: Pharmacology: Food Safety, and Analysis.

Ghimpețeanu OM, Pogurschi EN, Popa DC, Dragomir N, Drăgotoiu T, Mihai OD, Petcu CD, 2022. Antibiotic use in livestock and residues in food-A public health threat: A review, Foods. 11(10): p1430. https://doi.org/10.3390/foods11101430

Haque MA, Alam MZ, Iqbal A, Lee YM, Dang CG, Kim JJ. 2023. Genome-wide Association studies for body conformation traits in Korean Holstein Population. Animals, 13(18): p.2964. https://doi.org/10.3390/ani13182964

Haquea MA, Hashema MA, Mujaffar MM, Rimaa FJ, Hossainb A. 2017. Effect of gamma irradiation on shelf life and quality of beef. Journal of Meat Science and Technology, 5(2): 20–28.

Hashem MA, Islam T, Hossain MA, Kamal MT, Sun MA, Rahman MM. 2020. Production performance of Jamuna basin lamb under semi-intensive management system in Bangladesh. Journal of Animal and Veterinary Advances, 19(11): 150–158.

Hossain MS, Rokib M, Habib M, Kabir MH, Hashem MA, Azaz MA, Rahman MM, Ali MS. 2021. Quality of spent hen sausages incorporated with fresh ginger extract. Meat Research, 1(1). Article No. 04. DOI: https://doi.org/10.55002/mr.1.1.4

Hossen MJ, Hossain MS, Abedin MJ, Karim MR, Rume FI, 2008. Animal production strategies in southern region of Bangladesh. The Agriculturists, 6(1-2):77-83. https://doi.org/10.3329/agric.v6i1.5217

Ibrahim A, Junaidu A, Garba MK, 2010. Multiple antibiotic residues in meat from slaughtered cattle in Nigeria. The Internet Journal of Veterinary Medicine, 8(1):354.

Kasimanickam V, Kasimanickam M, Kasimanickam R. 2021. Antibiotics Use in Food Animal Production: Escalation of Antimicrobial Resistance: Where Are We Now in Combating AMR? Medical Sciences, 9(1):14. https://doi.org/10.3390/medsci9010014

Khatun R, Akter I, Deen RA, Ghosh D, Hashem MA. 2025. Are there any differences on sensory and physicochemical attributes of pre-and post-rigor broiler meat? Meat Research, 5(1). Article No. 106. DOI: https://doi.org/10.55002/mr.5.1.106

Kjeldgaard J, Cohn MT, Casey PG, Hill C, Ingmer H. 2012. Residual antibiotics disrupt meat fermentation and increase risk of infection. MBio, 3(5):10-128. https://doi.org/10.1128/mbio.00190-12

Manyi-Loh C, Mamphweli S, Meyer E, Okoh A. 2018. Antibiotic Use in Agriculture and Its Consequential Resistance in Environmental Sources: Potential Public Health Implications. Molecules, 23(4): p795. https://doi.org/10.3390/molecules23040795

Marshall B.M, Levy S.B. 2011. Food animals and antimicrobials: impacts on human health. Clinical Microbiology Reviews: 24(4):718-33. https://doi.org/10.1128/cmr.00002-11

Mgonja F, Mosha R, Mabiki F, Choongo K. 2017. Oxytetracycline residue levels in beef in Dodoma region, Tanzania. African Journal of Food Science, 11(2):1-4. https://doi.org/10.5897/AJFS2016.1532

Mohammed N, Adare Mengistu D, Abdurehman A, Belina D, Mengistu S. 2022. Determination of Tetracycline Residues in Kidney and Muscle of Beef Cattle Slaughtered in Dire Dawa and Harar Municipal Abattoirs, Eastern Ethiopia. Environ Health Insights, 16(1):1-11. https://doi.org/10.1177/11786302221109720

Nchima G, Choongo K, Muzandu K, Nalubamba K, Muma J, Bumbangi F, Monga G, Kangwa HJAJRC. 2017. Determination of oxytetracycline and sulphamethazine residues in marketed beef from selected parts of Zambia to assess compliance with maximum residual limits. American Journal of Research Communication, 5: 42-64.

Ovando HG, Gorla N, Weyers A, Ugnia L, Magnoli A. 2004. Simultaneous quantification of ciprofloxacin, enrofloxacin and balofloxacin in broiler chicken muscle. Archivos de Medicina Veterinaria, 36(1): 93-8.

Phillips I, Casewell M, Cox T, De Groot B, Friis C, Jones R, Nightingale C, Preston R, Waddell J. 2004. Does the use of antibiotics in food animals pose a risk to human health? A critical review of published data. Journal of Antimicrobial Chemotherapy, 53(1):28-52.

Salam MA, Al-Amin MY, Salam MT, Pawar JS, Akhter N, Rabaan AA, Alqumber MAA. 2023, Antimicrobial Resistance: A Growing Serious Threat for Global Public Health. Healthcare, 11(13):1-20. https://doi.org/10.3390/healthcare11131946

Sanz D, Razquin P, Condón S, Juan T, Juan T, Herraiz B, Mata L. 2015, Incidence of Antimicrobial Residues in Meat Using a Broad Spectrum Screening Strategy. European Journal of Nutrition & Food Safety, 5(3):156-165.

Sarkar R, Jahan I, Sarker T, Deen RA, Ghosh D, Akther S, Hashem MA. 2024. Drip loss assessment by standard bag and filter paper wetness method and their relationships with broiler chicken meat quality classification. Meat Research, 4(4). Article No. 100. DOI: https://doi.org/10.55002/mr.4.4.100

Sarker T, Deen RA, Ghosh D, Mia N, Rahman MM, Hashem MA. 2024a. AI-driven approach and NIRS: A review on meat quality and safety. Meat Research, 4(6). Article No. 105. DOI: https://doi.org/10.55002/mr.4.6.105

Sarker T, Jahan R, Deen RA, Ghosh D, Hashem MA. 2024b. Tenderness, histomorphology of muscle fiber and gene expression in primal cuts of cattle – A Review. Meat Research, 4(5). Article No. 101. DOI: https://doi.org/10.55002/mr.4.5.101

Scaria J, Anupama KV, Nidheesh PV. 2021. Tetracyclines in the environment: An overview on the occurrence, fate, toxicity, detection, removal methods, and sludge management. Science of the Total Environment, 1(771):145291. https://doi.org/10.1016/j.scitotenv.2021.145291

Siddiqua T, Hossain MA, Khan M, Hashem MA. 2018. Formulation of value-added beef meatball using tulsi (Ocimum sanctum) leaf extract as a source of natural antioxidant. Journal of the Bangladesh Agricultural University, 16(2): 260–265. DOI: https://doi.org/10.3329/jbau.v16i2.37980

Verma MK, Ahmad AH, Pant D, Patwal PC. 2021. Evaluation of Oxytetracycline residues in Chicken Meat Samples by HPLC. The Pharma Innovation Journal, 10(4S):155-157.

Widiastuti R, Martindah E, Anastasia Y. 2022. Detection and Dietary Exposure Assessment of Fluoroquinolones Residues in Chicken Meat from the Districts of Malang and Blitar, Indonesia. Tropical Animal Science Journal, 45(1):98-103. https://doi.org/10.5398/tasj.2022.45.1.98

Zhang Y, Lu J, Yan Y, Liu J, Wang M. 2021. Antibiotic residues in cattle and sheep meat and human exposure assessment in southern Xinjiang, China. Food Science & Nutrition, 9(11):6152-6161. https://doi.org/10.1002/fsn3.2568