

Article

Prevalence and co-occurrence of fluoroquinolone and tetracycline residues in broiler liver from retail markets in Mymensingh, Bangladesh

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Abstract: Antimicrobial drug residues in foods of animal origin pose a significant food safety and public health concern, contributing to antimicrobial resistance (AMR) in the food chain. In Bangladesh, the poultry industry is rapidly growing, yet there is minimal monitoring of antibiotic residues in retail products, coupled with low consumer awareness. This study aimed to assess the extent of fluoroquinolone and tetracycline residues in retail broiler liver samples and to evaluate the knowledge, attitudes, and practices (KAP) of consumers and vendors in Mymensingh, Bangladesh. The research was a cross-sectional study conducted between March and September 2018. Fifty broiler livers were purchased from four markets and screened for residues using thin-layer chromatography (TLC) with certified standards and predefined acceptance windows. Concurrent KAP interviews were conducted with 216 respondents, including 120 consumers and 96 vendors/handlers. The residue screening revealed that 45 out of 50 livers (90.0%; 95% CI 78.6–95.7) tested positive for at least one target class, with market-wise positivity ranging from 84.6% to 92.3%. Among the positive samples, bands consistent with ciprofloxacin were the most frequent, followed by oxytetracycline, doxycycline, and enrofloxacin. In the KAP survey, 39.8% of respondents demonstrated adequate knowledge ($\geq 75\%$ correct), 56.0% had favorable attitudes, and 36.6% reported appropriate practices. Vendors exhibited lower rates of appropriate practices compared to consumers (26.0% vs. 45.0%). Adequate knowledge (adjusted OR 3.72; 95% CI 2.02–6.86) and favorable attitudes (adjusted OR 2.41; 95% CI 1.29–4.52) were independently associated with appropriate practices. Additionally, markets with a higher mean vendor KAP index tended to have lower TLC positivity (Spearman $\rho = -0.95$; $P = 0.051$). Although TLC is qualitative and does not assess compliance with maximum residue limits (MRL), the high positivity rates indicate a substantial risk of consumer exposure at the point of sale. Targeted communication focused on vendor compliance with withdrawal periods, documentation/traceability, and standardized consumer advisories, alongside routine low-cost screening and confirmatory testing, could enhance stewardship and reduce downstream residues in this context.

Keywords: antimicrobial resistance; thin-layer chromatography; KAP; withdrawal-period; food safety

1. Introduction

Antimicrobial residues in foods of animal origin continue to pose a persistent threat to food safety, consumer health, and the sustainability of livestock production systems worldwide. These residues, when present beyond permissible limits, can trigger allergic reactions, disrupt gut microbiota, and, more critically, accelerate the

emergence and dissemination of antimicrobial resistance (AMR) along the food chain (Elbehiry *et al.*, 2025; Izah *et al.*, 2025). The World Health Organization (WHO) and the Food and Agriculture Organization (FAO) have emphasized that AMR represents one of the most pressing global health challenges of the 21st century, linking the misuse of antibiotics in animal agriculture to the increasing prevalence of resistant pathogens in humans. In this context, monitoring and managing antimicrobial residues in foods—particularly in animal-derived products—are vital components of a One Health strategy to safeguard both human and animal well-being (Coque *et al.*, 2023; Salam *et al.*, 2023; Aslam *et al.*, 2024).

The Codex Alimentarius Commission provides an international framework for managing veterinary drug residues through the establishment of maximum residue limits (MRLs) and recommended analytical approaches for screening and confirmation (Huang and Li, 2025). According to Codex principles, screening tests such as thin-layer chromatography (TLC) and enzyme-linked immunosorbent assays (ELISA) are designed for rapid detection of suspect samples but do not establish compliance with regulatory MRLs without quantitative confirmation by validated methods such as high-performance liquid chromatography (HPLC) or liquid chromatography–mass spectrometry (LC–MS/MS) (Brera *et al.*, 2011; Cammilleri *et al.*, 2019; Nugroho *et al.*, 2022; Getahun *et al.*, 2023; Khalifa *et al.*, 2024). Nevertheless, screening methods remain indispensable in resource-limited settings, where they can serve as early-warning tools to detect the presence of antibiotic residues in food products available at local markets (Pikkemaat, 2009).

Bangladesh's poultry industry has undergone rapid expansion over the past two decades, providing affordable animal protein to a growing population and contributing significantly to national food security (Rahman *et al.*, 2017; Masud *et al.*, 2020; Islam, 2025). However, this expansion has been accompanied by widespread antibiotic use, often without adequate veterinary oversight or adherence to withdrawal periods prior to slaughter (Salam *et al.*, 2023; Zhannara *et al.*, 2025). Previous studies have identified residues of fluoroquinolones, tetracyclines, sulfonamides, and macrolides in poultry meat, liver, and eggs, with liver tissues frequently showing higher residue concentrations due to their role in drug metabolism and accumulation. Such findings raise concerns about consumer exposure to residual antimicrobials through commonly consumed poultry products, particularly in informal market systems where traceability and regulatory monitoring are limited (Ulomi *et al.*, 2022; Wang *et al.*, 2022; Izah *et al.*, 2025).

Among the major antibiotic classes, fluoroquinolones and tetracyclines are widely used in poultry farming for therapeutic, prophylactic, and sometimes growth-promoting purposes (Abou-Jaoudeh *et al.*, 2024). Fluoroquinolones, such as ciprofloxacin and enrofloxacin, are broad-spectrum antimicrobials that inhibit bacterial DNA gyrase, while tetracyclines, including oxytetracycline and doxycycline, interfere with bacterial protein synthesis (Shariati *et al.*, 2022; Bacanlı, 2024). Their extensive and sometimes irrational use contributes to selection pressure on microbial communities, facilitating the emergence of resistant bacteria that can be transmitted to humans via the food chain, direct contact, or the environment. In Bangladesh, several studies have documented the persistence of these residues in retail poultry products, yet systematic surveillance at the market level remains sporadic and largely confined to academic or pilot initiatives (Sarker *et al.*, 2018; Sani *et al.*, 2023).

While analytical detection of residues provides a quantitative picture of contamination, understanding the behavioral and structural factors that contribute to residue occurrence is equally essential for effective intervention. The knowledge, attitudes, and practices (KAP) of market actors—particularly vendors and consumers—shape how antibiotics are sourced, handled, and perceived (Hassan *et al.*, 2021; Sawadogo *et al.*, 2023). Inadequate knowledge about withdrawal periods, lack of consumer demand for residue-safe products, and limited vendor accountability contribute to the persistence of residues in the retail supply chain. Therefore, coupling residue monitoring with behavioral assessment provides a more holistic understanding of how risk is generated and potentially mitigated at the point of sale (Mithuna *et al.*, 2024).

This study was based on the hypothesis that poor antibiotic stewardship and non-compliance with withdrawal periods among poultry vendors contribute to fluoroquinolone and tetracycline residues in retail broiler livers in Mymensingh markets. The study asked research question, are the knowledge, attitudes, and practices of poultry vendors and consumers associated with the occurrence of fluoroquinolone and tetracycline residues in retail broiler livers sold in Mymensingh markets? Considering the hypothesis, and research question the study was conducted to determine the prevalence of fluoroquinolone and tetracycline residues, assess KAP among consumers and vendors, and explore the link between vendor behavior and residue detection in the markets. The findings highlight the need for market-level interventions, including vendor training, consumer awareness, and routine screening. Strengthening stewardship and integrating low-cost monitoring tools like TLC could reduce antimicrobial residues and help prevent the spread of antimicrobial resistance through the food chain.

2. Materials and Methods

2.1. Ethical approval and informed consent

This study involved no live-animal procedures; retail food items were purchased from open markets and therefore did not require animal ethics approval under departmental policy. Verbal informed consent was obtained and documented by the interviewer before each anonymous interview with adults (≥ 18 years). Participation was voluntary, with the right to refuse or withdraw at any time.

2.2. Study area and sampling period

The study was carried out in Mymensingh city, Bangladesh, between March and September 2018, covering four major retail markets with high customer activity, including Sesh More (M1), K.R. Market (M2), Fossil's Square (M3), and Zabberer More (M4) (Figure 1).

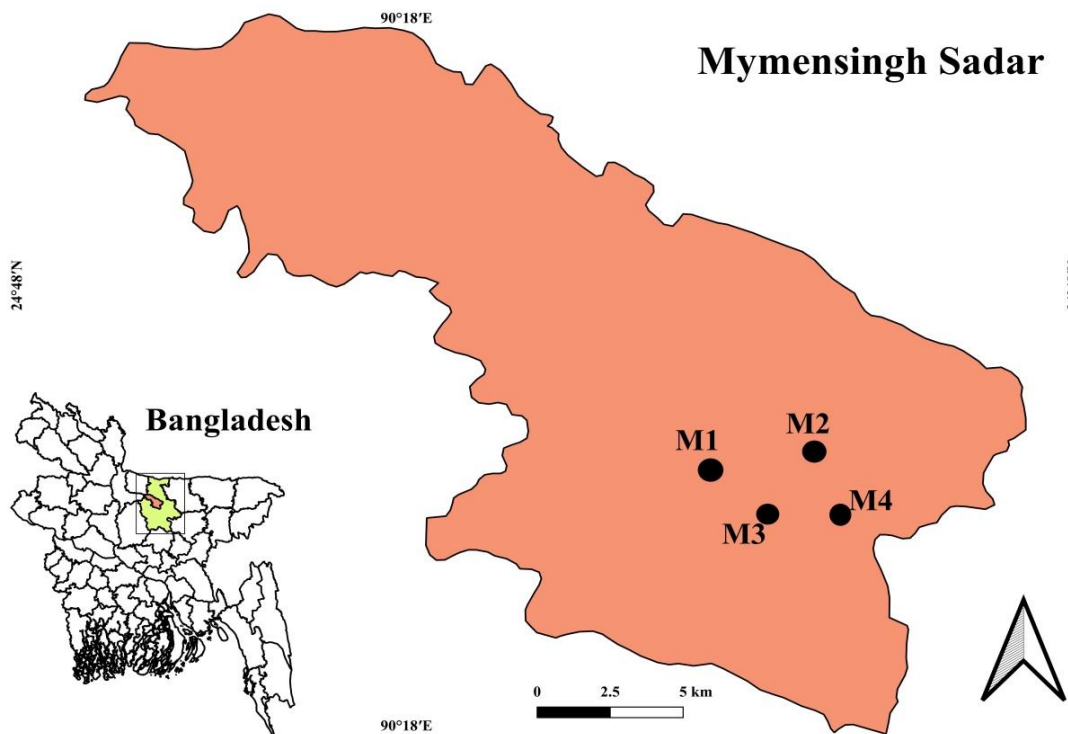


Figure 1. The study was conducted in Mymensingh city, Bangladesh.

2.3. Study design

A cross-sectional study was conducted to assess antimicrobial residues in broiler livers and to evaluate the KAP of market actors, including both consumers and vendors or handlers. Residue screening and KAP surveys were carried out simultaneously in the same markets during the study period. The survey targeted two respondent groups: consumers purchasing broiler meat and vendors or handlers engaged in selling or processing broilers. A total of 216 respondents were included, comprising 120 consumers and 96 vendors, selected through systematic intercept sampling across different markets and time blocks. Data were collected using a structured questionnaire containing 25 to 30 items that covered demographic information, as well as specific sections on knowledge, attitudes, and practices related to antibiotic use and food safety (Table 1). The questionnaire was carefully translated, back-translated, pre-tested for clarity, and administered anonymously, with each interview taking approximately eight to ten minutes to complete.

Table 1. Study design components with sample types, respondent categories, market locations, and sample sizes.

Component	Sample/respondent type	Market (s)	Sample size (n)
Residue screening	Broiler livers	M1-M4	50
KAP survey	Consumers	M1-M4	120
KAP survey	Vendors/handlers	M1-M4	96

2.4. Sample size and selection (liver residues)

A total of 50 individual broiler liver samples, each weighing approximately 100–150 g, were collected from the four selected markets (M1–M4) to represent typical consumer exposure patterns. On each sampling day, purchases were made from different vendors to reduce potential clustering and ensure sample diversity. The collected samples were immediately sealed in sterile zipper bags, transported on ice to the laboratory within two to three hours, and stored at -20°C until analysis. Each liver sample was subjected to a single freeze–thaw cycle prior to extraction to maintain sample integrity.

2.5. Chemicals and reference standards

Certified reference standards ($\geq 99\%$ purity) for ciprofloxacin (CIP), enrofloxacin (ENR), oxytetracycline (OTC), and doxycycline (DOX) were procured from Sigma-Aldrich through Renata Limited, Bangladesh. The solvents and reagents used included phosphate-buffered saline (PBS, pH 6.5), trichloroacetic acid (TCA, 30%), diethyl ether (DEE), acetone (RCI Labscan, Thailand), and methanol (Merck, Germany). Ultrapure water was used for all aqueous preparations to avoid contamination and maintain analytical precision.

2.6. Sample processing and extraction

Liver extraction was performed following previously described protocols with minor modifications (Sarker *et al.*, 2018). Approximately 2.0 g of thawed liver tissue was homogenized with 8 mL of PBS (pH 6.5) in a sterile 50 mL tube. Proteins were precipitated by adding 2 mL of 30% TCA, followed by vortexing (Vortex-XHC, Wincom, China) for 30 seconds and centrifugation (Hettich D-78532, Germany) at $4,000\times g$ for 10 minutes at room temperature. The resulting supernatant was transferred to a separatory funnel and extracted twice with 10 mL of diethyl ether, each time mixed vigorously for approximately one minute. The combined organic layers were dried over anhydrous sodium sulfate, filtered, and evaporated under gentle air or nitrogen at approximately 40°C to dryness. The residues were finally reconstituted in 1.0 mL of methanol for subsequent TLC analysis.

2.7. Thin-layer chromatography (TLC) screening

TLC was conducted following the method described by Sarker *et al.* (2018). Analyses were carried out on aluminum-backed silica gel 60 F254 plates (MN-Germany). For each plate, 5–10 μL of the sample extract and 2–5 μL of the working standards were applied as separate tracks, maintaining a minimum spacing of 1.0 cm between tracks and 1.5 cm from the plate edge. The developing chamber was pre-saturated for 15 minutes with a 1:1 (v/v) mixture of acetone and methanol as the mobile phase. Plates were then developed to an 8.0 cm solvent front, air-dried, and visualized under UV light at 254–256 nm (F18W, Germany). A sample was considered positive when a discrete band matched the retention factor (R_f) of the corresponding standard within ± 0.05 and exhibited intensity above the negative control without artifacts. Instances where multiple qualifying bands appeared on a single track were recorded as co-detections.

2.8. Quality assurance and quality control

Each analytical batch included a solvent blank, a negative matrix control, a positive control prepared by spiking the liver matrix at approximately one to two times the TLC limit of detection for each antibiotic, and a standard ladder. Any plates that showed distorted solvent fronts, control failures, or ambiguous bands were repeated to maintain analytical accuracy. The acceptance criteria for band identification and R_f values were defined in advance, prior to unblinding the market identities of the samples.

2.9. Scoring and data management

Knowledge was scored from 0 to 12, with each correct response assigned a value of 1, and a score of ≥ 9 defined as adequate. Attitudes were scored from 12 to 60, with a score of ≥ 48 considered favorable. Practices were scored from 0 to 12, with “Always” on good behaviors coded as 1, and a score of ≥ 9 defined as appropriate. A composite KAP index ranging from 0 to 1 was calculated by averaging the normalized domain scores. For missing items, values were imputed within a domain if $\geq 75\%$ of items were answered. Data were managed in IBM SPSS Statistics v26. Cronbach’s α was used to assess the internal reliability of the attitude scale.

2.10. Statistical analysis

Residue prevalence was expressed as proportions with 95% Wilson confidence intervals (CI), and group comparisons were performed using χ^2 or Fisher’s exact tests as appropriate. KAP data were summarized descriptively, and odds ratios (OR) with 95% CI were calculated. Predictors of appropriate practice were analyzed using multivariable binary logistic regression, adjusting for role, age, sex, education, market,

knowledge, and attitude, with model fit assessed by the Hosmer–Lemeshow test. The relationship between market-level vendor KAP indices and TLC residue positivity was examined using Spearman's ρ . Statistical significance was set at two-sided $\alpha = 0.05$. All statistical analyses were conducted using IBM SPSS Statistics version 26.0, and the study area map was prepared using QGIS version 3.42.

3. Results

3.1. Liver residue screening

Overall, 45 out of 50 broiler liver samples screened positive for at least one target antibiotic class (fluoroquinolones or tetracyclines) using TLC, corresponding to an overall positivity of 90.0% (95% CI 78.60–95.70). Market-level positivity of fluoroquinolone and tetracycline residues ranged from the highest in Zabberer More at 92.3%, followed by Sesh More and K.R. Market, both at 91.7%, and was lowest in Fossil's Square at 84.6%. Market-wise positivity ranged from 84.6% to 92.3%, and no significant difference was observed across the four markets ($\chi^2 = 0.57$, $P = 0.90$) (Table 2).

Table 2. Positivity of fluoroquinolone and tetracycline residues in broiler liver samples by retail market.

Market (code)	Tested (n)	Positive (n)	Positive (%)	95% CI
Sesh More (M1)	12	11	91.70	64.60-98.50
K.R. Market (M2)	12	11	91.70	64.60-98.50
Fossil's Square (M3)	13	11	84.60	57.80-95.70
Zabberer More (M4)	13	12	92.30	66.70-98.60
Total	50	45	90.00	78.60-95.70

Among the 45 TLC-positive samples, ciprofloxacin-consistent bands were the most frequently detected, appearing in 24 instances (38.10%), followed by oxytetracycline in 16 instances (25.40%), doxycycline in 14 instances (22.20%), and enrofloxacin in 9 instances (14.30%) (Table 3). Co-detections were common, with some samples exhibiting multiple antibiotic-consistent bands on the same track, resulting in a total of 63 detections across all samples.

Table 3. Detection frequency of specific antibiotics in TLC-positive broiler liver samples (n = 45).

Antibiotic	Detections (count)	Share of detections (%)
Ciprofloxacin	24	38.10
Oxytetracycline	16	25.40
Doxycycline	14	22.20
Enrofloxacin	9	14.30
Total detections	63	100

3.2. KAP survey

A total of 216 respondents were interviewed across the four markets, including 120 consumers and 96 vendors or handlers. The overall mean age was 35.20 ± 11.40 years, with consumers averaging 33.80 ± 10.70 years and vendors 37.10 ± 11.80 years. Males comprised 58.30% of the total sample, representing 52.50% of consumers and 66.70% of vendors. More than half of the participants (57.40%) had attained education at the secondary level or above. The Attitude scale demonstrated good internal consistency with a Cronbach's α of 0.82 (Table 4).

Table 4. Demographic characteristics of respondents by role (n = 216).

Characteristic	Consumers (n = 120)	Vendors (n = 96)	Total (n = 216)
Age, years (mean $\pm$ SD)	33.80 ± 10.70	37.10 ± 11.8	35.20 ± 11.40
Male [n (%)]	63 (52.50)	64 (66.70)	126 (58.30)
Education $\geq$ secondary, n (%)	78 (65.0)	46 (47.90)	124 (57.40)

SD=standard deviation.

Overall, 39.8% of respondents demonstrated adequate knowledge, defined as correctly answering at least 75% of knowledge items. Favorable attitudes were observed in 56.0% of participants, with the attitude scale showing good internal consistency (Cronbach's $\alpha = 0.82$). Appropriate practices, defined as reporting “always” for at

least 9 of 12 practice items, were reported by 36.6% of respondents. Consumers exhibited higher rates of appropriate practice than vendors (45.0% vs 26.0%), with vendors having lower odds of appropriate practice compared to consumers (odds ratio = 0.43; 95% CI 0.24–0.77; $P = 0.0045$). Both adequate knowledge and favorable attitudes were positively associated with appropriate practices in bivariate analyses and remained significant predictors in multivariable models (Table 5).

Table 5. Knowledge, attitude, and practice responses among respondents (n = 216).

Items	Values
Knowledge	Correct [n (%)]
K1. Antibiotics treat bacterial infections	182 (84.30)
K2. Antibiotics sometimes used to promote growth in broilers	146 (67.60)
K3. Using antibiotics without veterinary advice can be harmful	176 (81.50)
K4. A withdrawal period is required after antibiotics before slaughter	154 (71.30)
K5. Early slaughter can leave residues in meat/offal	162 (75.0)
K6. Liver may contain higher residues than muscle	132 (61.10)
K7. Usual cooking always destroys residues (False)	91 (42.10)
K8. Residues may contribute to AMR	168 (77.80)
K9. Antibiotics do not treat viral infections	170 (78.70)
K10. Buying from rule-compliant vendors reduces residue risk	144 (66.70)
K11. Only a veterinarian should prescribe antibiotics for poultry	158 (73.10)
K12. Checking labels/invoices helps identify recent antibiotic use	118 (54.60)
Adequate knowledge ($\geq 9/12$), n (%)	86/216 (39.80)
Attitude	Mean $\pm$ SD
A1. “Antibiotics are necessary even without disease” (reverse)	2.40 $\pm$ 1.20
A2. Withdrawing antibiotics before slaughter is essential	4.20 $\pm$ 0.90
A3. Authorities should enforce penalties for violations	4.10 $\pm$ 0.90
A4. Vendors should display compliance/source information	3.80 $\pm$ 1.0
A5. Willing to pay/accept small margin for residue-safe products	3.50 $\pm$ 1.10
A6. Residues in liver are a bigger concern than in muscle	3.70 $\pm$ 1.0
A7. Advising customers about residues is my responsibility	3.60 $\pm$ 1.10
A8. Antibiotics should not be sold without prescription	4.0 $\pm$ 1.0
A9. Training for vendors on stewardship is useful	4.10 $\pm$ 0.90
A10. I trust certified/inspected suppliers more	3.90 $\pm$ 1.0
A11. If residues are common, I would avoid liver for family	3.40 $\pm$ 1.20
A12. Awareness campaigns change behavior	3.80 $\pm$ 1.0
Total Attitude score (12–60)	46.10 $\pm$ 7.80
Favorable attitude (≥ 48)	121/216 (56.0)
Practice	Good [n (%)]
P1. Ask about recent antibiotic use	78 (36.10)
P2. Avoid buying/selling birds that appear recently medicated	96 (44.40)
P3. Prefer sources with written records/receipts	88 (40.70)
P4. Wash hands/tools after handling raw liver/meat	156 (72.20)
P5. Keep raw liver separate from ready-to-eat foods	142 (65.70)
P6. Cook liver thoroughly	168 (77.80)
P7. Inform customers/family about safe handling/cooking	114 (52.80)
P8. Buy/sell from suppliers with better hygiene	132 (61.10)
P9. Avoid using leftover antibiotics without advice	139 (64.40)
P10. Support and comply with inspections	128 (59.30)
P11. Discard stock if advised it violates withdrawal period	104 (48.10)
P12. Follow news/education on residues	92 (42.60)
Appropriate practice ($\geq 9/12$), n (%)	79/216 (36.60)

3.3. Linkage between vendor KAP and residue positivity

Fossil’s Square (M3), with the highest average vendor KAP index of 0.66, had the lowest residue positivity at 84.6%, whereas Zabberer More (M4), with the lowest vendor KAP index of 0.48, exhibited the highest residue positivity of 92.3%. Vendor KAP indices, calculated on a 0–1 scale, were averaged for each market and compared with market-level TLC positivity. Markets with higher mean vendor KAP tended to have lower residue positivity, indicating a potential inverse relationship between vendor stewardship and the presence of

antimicrobial residues. The Spearman correlation coefficient was -0.95 , approaching statistical significance ($P = 0.051$; $n = 4$), suggesting a strong negative association despite the small number of markets (Table 6).

Table 6. Vendor KAP index versus TLC positivity by market.

Market	Vendor KAP index (mean)	TLC positivity (%)
Sesh More (M1)	0.52	91.70
K.R. Market (M2)	0.50	91.70
Fossil’s Square (M3)	0.66	84.60
Zabberer More (M4)	0.48	92.30

4. Discussion

The study demonstrated an alarmingly high prevalence of antimicrobial residues in broiler liver samples, with 90.0% of the tested samples positive for at least one antibiotic class, indicating extensive and potentially indiscriminate use of antibiotics in poultry production. Such a high positivity rate suggests that withdrawal periods before slaughter are either inadequately enforced or poorly understood by poultry producers and vendors (Rware *et al.*, 2024; Adhikari *et al.*, 2025). This finding aligns with earlier studies in Bangladesh and other low- and middle-income countries, where weak regulatory frameworks and limited awareness among stakeholders contribute to persistent residue contamination in animal-derived foods (Banna *et al.*, 2021). Although residue positivity varied slightly among markets—from 84.6% at Fossil’s Square to 92.3% at Zabberer More—the absence of a statistically significant difference ($\chi^2 = 0.57$, $P = 0.90$) indicates that antimicrobial misuse is widespread and not confined to specific locations. This uniformity across markets reflects systemic issues along the poultry production and distribution chain rather than isolated market-level practices. Factors such as over-the-counter antibiotic availability, lack of veterinary supervision, and pressure for rapid flock turnover likely contribute to the pervasive presence of residues (Masud *et al.*, 2020; Adhikari *et al.*, 2025).

The detection of both fluoroquinolones and tetracyclines is particularly concerning, given their critical importance in both human and veterinary medicine. Continuous exposure to low-dose residues through food can facilitate antimicrobial resistance development in gut microbiota, posing a major public health risk. Moreover, chronic ingestion of such residues may lead to allergic reactions, dysbiosis, and other long-term health implications (Granados-Chinchilla and Rodríguez, 2017; Abou-Jaoudeh *et al.*, 2024).

The residue profiling of TLC-positive samples revealed ciprofloxacin as the most frequently detected antibiotic, accounting for 38.10% of all detections, followed by oxytetracycline, doxycycline, and enrofloxacin. The predominance of ciprofloxacin, a widely used fluoroquinolone, suggests its extensive and perhaps uncontrolled application in broiler farming, possibly for disease prevention and growth promotion (Sarker *et al.*, 2018; Sani *et al.*, 2023). Liver is a reasonable sentinel tissue for screening because of its central role in drug biotransformation and potential for higher residue burdens than muscle (Tufa, 2016). The relatively high detection of tetracyclines, particularly oxytetracycline and doxycycline, further indicates their continued use as broad-spectrum agents despite existing regulatory restrictions. The occurrence of multiple antibiotic residues within the same sample points to concurrent or sequential administration of different drug classes, reflecting a lack of veterinary guidance and poor withdrawal management practices (Sarker *et al.*, 2018; Sani *et al.*, 2023). The presence of multiple residues is particularly concerning as it increases the risk of antimicrobial resistance transfer through the food chain, undermines the efficacy of essential antibiotics, and may lead to cumulative toxicological effects in consumers (Farkas *et al.*, 2025; Izah *et al.*, 2025).

From a risk-management standpoint, our findings should be interpreted through the Codex framework: TLC is an appropriate, low-cost screening tool to detect suspect samples, but it does not establish regulatory non-compliance relative to MRLs; confirmation requires a validated quantitative method and residue definitions specific to each marker analyte (Chakma *et al.*, 2023; Sayari *et al.*, 2025). Nevertheless, a 90% screening positivity in retail liver is epidemiologically meaningful. Even when final concentrations are below MRLs, recurrent exposure—combined with improper handling and cooking beliefs—can contribute indirectly to selection pressure for AMR along the food chain (Landers *et al.*, 2012; McEwen and Collignon, 2018). The KAP profile observed here underscores this interface, fewer than half of respondents recognized that routine cooking does not reliably eliminate antibiotic residues, and documentation-seeking behaviors were weak. These deficits are actionable targets for market-level stewardship.

The demographic profile and KAP outcomes of respondents highlight key behavioral and cognitive gaps influencing antimicrobial residue risk in the poultry market chain. Most participants were young to middle-aged

adults with a moderate educational background, suggesting reasonable potential for behavioral change through awareness interventions. Although over half of the respondents exhibited favorable attitudes toward antibiotic use and food safety, less than half demonstrated adequate knowledge, and only about one-third practiced appropriate handling and usage behaviors. This discrepancy between attitude and practice indicates that positive perceptions do not consistently translate into responsible actions, possibly due to weak regulatory oversight, lack of practical training, or economic incentives that favor short-term profit over compliance (Hassan *et al.*, 2021; Azim *et al.*, 2023). The finding that consumers reported significantly higher appropriate practices than vendors underscores the critical influence of vendor-side behaviors in residue control. Vendors' lower adherence to best practices may stem from limited technical understanding of withdrawal periods, insufficient enforcement of market-level regulations, and inadequate communication between producers and retailers (Wallace *et al.*, 2022; Khalifa *et al.*, 2024).

Evidence from behavior-change programs in food markets suggests that combining on-site training, visible compliance cues (e.g., stall placards indicating last inspection date), and modest economic incentives can shift both attitudes and practices (Azim *et al.*, 2023). Third, strengthening veterinary gatekeeping restricting non-prescription access to medically important antibiotics remains a cornerstone for reducing misuse upstream, thereby reducing downstream residues and AMR risk (Landers *et al.*, 2012; McEwen and Collignon, 2018). The analysis revealed a strong inverse association between vendor KAP scores and the prevalence of antimicrobial residues across markets, underscoring the potential role of human behavior in influencing food safety outcomes. Fossil's Square, which demonstrated the highest average vendor KAP index (0.66), also showed the lowest residue positivity (84.6%), whereas Zabberer More, with the lowest KAP index (0.48), exhibited the highest residue positivity (92.3%). This pattern suggests that markets with better-informed and more responsible vendors tend to have lower levels of contamination in broiler liver samples. The strong negative Spearman correlation ($\rho = -0.95$, $P = 0.051$) reinforces the notion that improved vendor awareness and compliance could meaningfully reduce antimicrobial residue risks at the retail level. Although the result narrowly missed conventional statistical significance, likely due to the small number of markets, the magnitude and direction of the correlation are biologically and behaviorally plausible. Vendors with stronger stewardship—reflected in greater adherence to withdrawal periods, cleaner handling practices, and awareness of antibiotic misuse—are less likely to sell contaminated products (Pearson and Chandler, 2019).

This study has several limitations. The TLC method is qualitative and cannot confirm MRL exceedance or differentiate overlapping antibiotic compounds. Farm-level drug histories were not collected, limiting interpretation of non-compliance versus legitimate use. The KAP survey relied on self-reported data and a limited market sample, introducing possible bias. The correlation between vendor KAP and residue positivity is based on few clusters and should be interpreted cautiously. Despite these constraints, the study provides valuable insights by linking residue screening with behavioral data and highlighting vendor-focused, low-cost interventions to improve antibiotic stewardship and food safety in local markets.

5. Conclusions

This study evaluated antimicrobial drug residues in retail broiler livers and examined how vendor practices may influence residue occurrence. Widespread fluoroquinolone and tetracycline residues were detected, and vendors generally showed weaker practices than consumers. Better knowledge and responsible attitudes were associated with safer behavior, while markets with stronger stewardship exhibited fewer residues, suggesting that vendor practices at the retail level can affect food safety outcomes. Low-cost screening like TLC can serve as an early warning system if supported by confirmatory testing. Targeted training, monitoring of withdrawal-period compliance, and improved consumer guidance are crucial for reducing risks. Although limited by the qualitative nature of TLC and the small sample scope, future studies should expand geographically, include quantitative residue analysis, and assess whether vendor-focused interventions effectively reduce contamination levels in retail broiler products.

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Data availability

Detail information on the datasets and materials used in this study are available from the corresponding author on reasonable request.

Conflict of interest

None to declare.

Authors' contribution

Mohammad Asaduzzaman: sample collection, study design, writing the first draft of the manuscript; Md. Zakaria Al Noman: laboratory screening, quality control, data curation, statistical analyses; Tanjina Afrin: data curation, manuscript editing; Sharmin Aktar: field interviews; Jannatul Ferdous: field interviews; Sabbya Sachi: sample collection, field interviews; Md. Rakib Hasan: manuscript writing, quality control, statistical analyses; Md. Sajanul Haque: manuscript writing, quality control, figure and tables preparation; Mahmudul Hasan Sikder: supervision, study design, project administration, funding. All authors have read and approved the final manuscript.

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