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Antibiotic susceptibility evaluation of bacteria in meat slaughterhouses: A review

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ABSTRACT

The meat processing industry generates wastewater rich in organic matter, nutrients, pathogens, and antibiotic residues, posing significant environmental and public health risks. Of particular concern is the presence of multidrug-resistant (MDR) bacteria, notably ESKAPE pathogens and *Escherichia coli*, which can persist through treatment processes and enter the food chain. This review compiles current detection techniques for antibiotic resistance, ranging from conventional methods such as Kirby–Bauer disk diffusion and broth microdilution to advanced molecular approaches including PCR, DNA microarrays, microfluidics, and biosensors. Recent surveillance studies from global and Indian slaughterhouses reveal high resistance rates to multiple antibiotic classes, often linked to overuse in livestock and poor hygiene practices. The emergence of resistance complicates treatment strategies, necessitating novel interventions. Phytochemicals—such as essential oils, flavonoids, tannins, and alkaloids—demonstrate promising antimicrobial activity, often enhancing antibiotic efficacy through synergistic effects. Plant-derived compounds like carvacrol, eugenol, thymol, and caffeine disrupt bacterial membranes, inhibit virulence factors, and target quorum sensing pathways. Addressing MDR pathogens in the meat industry requires integrated measures: stringent antibiotic stewardship, improved waste treatment, regular resistance monitoring, and exploration of natural antimicrobials as sustainable alternatives. This combined approach is essential to safeguard public health, food safety, and the long-term efficacy of antimicrobial therapies.

Keywords: Multi-drug Resistant, Biosensors, Quorum Sensing

1. INTRODUCTION

The meat processing industry is a significant contributor to global food production, but it also generates substantial volumes of wastewater with complex compositions. Meat industry wastewater is characterized by high concentrations of organic matter, including proteins, fats, oils, and grease, as well as elevated levels of nitrogen, phosphorus, and other nutrients. Additionally, it may contain pathogens, antibiotic residues, heavy metals, and chemical contaminants originating from processing, cleaning, and sanitization activities. If inadequately treated, the discharge of such wastewater can have severe environmental consequences, including water pollution, eutrophication [1], and ecological degradation. The bacterial composition of meat industry wastewater is highly diverse, consisting of both beneficial and pathogenic species. Certain bacterial communities play a vital role in the natural decomposition of organic waste by facilitating biodegradation and nitrogen cycling, while others can present considerable risks to both the environment and public health. The wastewater from the meat industry contains various bacteria, including *Escherichia coli*, *Proteus vulgaris*, and *Staphylococcus aureus*. that can lead to contamination of water bodies if wastewater is not adequately treated [2].The presence of ESKAPE bacteria (*Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Enterobacter spp.*) in slaughterhouse and meat industry effluents [3] poses significant risks, as these bacteria can persist in wastewater treatment systems, contribute to the spread of antimicrobial resistance (AMR). Bacterial isolates, including *Bacillus sp.*, *Citrobacter sp.*, *Lactobacillus sp.*, *Micrococcus sp.*, *Salmonella sp.*, *Serratia sp.*, and *Streptococcus sp.*, were also identified [4]. ESKAPE bacteria shows to be the root cause for various nosocomial infections [5]. They are known to be the causative agents for various diseases such as endocarditis, urinary tract infections, pneumonia, bloodstream infections, meningitis, dermatitis, wound infection and respiratory tract infection [6,7]. *Escherichia coli* and *Staphylococcus aureus* are responsible for diseases like urinary tract infections,

gastrointestinal infections, sepsis, pneumonia and toxic shock syndrome [8-9]. At present, the emergence of antimicrobial resistance (AMR) in the meat industry bacteria, often exacerbated by the overuse of antibiotics in meat production, underscores the need for more efficient treatment strategies.

Currently, effective wastewater treatment is executed to meet environmental regulations and reduce the spread of contamination both in India and globally. In India, activated sludge processes are done [10]. Other than this, effluent treatment is done in three stages which includes screening, air floatation, breakdown using anaerobic and aerobic bacteria, and removal of pollutants [11]. Globally, it can be seen for the treatment of wastewater from the meat industry, preliminary treatment is done for removing solids and large particles using methods such as screeners, strainers, and sieves, which can achieve up to 60% solid removal and over 30% BOD reduction. Physicochemical treatments, including dissolved air flotation (DAF), coagulation, flocculation, and electrocoagulation, are applied for solid-liquid separation and nutrient removal. Advanced treatment technologies, such as membrane filtration, biological processes, anaerobic and aerobic digestion, constructed wetlands, and advanced oxidation processes (AOPs), are planned for the removal of organic matter, nutrients, and pathogens, offering comprehensive solutions for wastewater treatment in the meat industry [12]. In this review, we will provide insights for detection techniques, antibiotic resistance and susceptibility patterns, and also natural alternatives for the anti-bacterials.

2. DETECTION TECHNIQUES

Antibiotic-resistant bacteria are prevalent within the community and may also be contracted through nosocomial infections, postoperative complications, and ingestion of contaminated food. Given the critical nature of septic patients and the escalating incidence of antibiotic-resistant bacteria, there is a pressing need for expedited antibiotic susceptibility testing (AST). AST is extensively employed in clinical settings to ascertain the antibiotic resistance profiles of bacterial isolates, to inform antibiotic treatment strategies, and to forecast therapeutic outcomes.

2.1. Current technologies:

2.1.2. Disk diffusion method:

The antibiotic susceptibility of bacterial isolates was assessed using the Kirby-Bauer disk diffusion method. Bacterial inoculums were prepared by suspending freshly cultured bacteria in normal saline and adjusting the turbidity to a 0.5 McFarland standard. The suspension was then evenly spread onto Mueller-Hinton agar using a cotton swab to ensure uniform bacterial growth. Antibiotic-impregnated paper disks were placed on the agar surface, and the plates were incubated aerobically at $35 \pm 1^\circ\text{C}$ for 18–24 hours. After incubation, the zone of inhibition around each disk, indicating bacterial susceptibility, was measured [13,14].

2.1.2. Broth dilution method:

Antibiotic susceptibility testing was conducted using a 96-well microtiter plate, where bacteria in the logarithmic phase were inoculated into Mueller Hinton broth containing varying tetracycline concentrations. Each concentration was tested in triplicate, and plates were incubated at 37°C for 18 hours. Absorbance at 600 nm was measured using a microplate reader. The minimum inhibitory concentration (MIC) was defined as the lowest antibiotic concentration at which bacterial survival was $\leq 1\%$ [15-16].

2.1.3. Polymerase chain reaction:

PCR analysis identified various antibiotic resistance genes in *Escherichia coli* O157:H7 isolates, including tetracycline efflux pumps (tetA, tetB, tetC, tetD, tetE, and tetG), streptomycin phosphotransferases (strA and strB), and aminoglycoside adenylyltransferase (aadA). Additionally, genes encoding resistance to chloramphenicol (cmlA), florfenicol (floR), and sulfonamides (sulI and sulIII) were detected. Beta-lactamase-mediated ampicillin resistance (ampC) was also confirmed, highlighting the presence of multidrug resistance mechanisms in these bacterial strains [17-18].

2.1.4. DNA microarray:

Another molecular based detection technique is DNA microarray analysis. In a study it shows that to detect virulence and antimicrobial resistance genes in *Escherichia coli* isolates from Great Lakes recreational waters DNA microarray is applied. The microarray was designed with **312** oligonucleotide probes, targeting 189 virulence genes and 30 antimicrobial resistance genes, enabling a comprehensive genetic profile of the isolates. The procedure involved genomic DNA extraction, fluorescent labeling with Cy5-dCTP, hybridization, and fluorescence scanning using the ScanArray Lite system, with a signal-to-noise ratio greater than 2.0 considered a positive detection. This high-throughput approach allowed the identification of key virulence genes, including *hlyA* (hemolysin), *sfa* (S-fimbriae adhesion), *iroN* (siderophore receptor), and *pap* (*P. fimbriae*), which are associated with pathogenic *Escherichia coli* strains. Additionally, the study detected antimicrobial resistance genes, such as tet(A), tet(B), blaTEM, aadA1, and sulIII, conferring resistance to tetracyclines, beta-lactams, aminoglycosides, and sulfonamides. The results highlighted the prevalence of multidrug-resistant and potentially pathogenic *Escherichia coli* in recreational waters, emphasizing the need for continuous surveillance and effective wastewater treatment strategies [14]

2.2. Emerging technologies:

2.2.1. Microfluidics based detection:

Microfluidic-based antimicrobial susceptibility testing (AST) using asynchronous magnetic bead rotation (AMBR) detects bacterial growth by monitoring changes in magnetic bead movement within nanoliter droplets. Pathogenic bacteria such as *Escherichia coli* bind to antibody-coated beads, and as they proliferate, changes in viscosity and size alter bead rotation under a magnetic field. When exposed to antibiotics like β -lactams or aminoglycosides, susceptible strains show inhibited growth, maintaining bead movement, while resistant strains slow rotation due to continued proliferation [20-21].

2.2.2. Biochemical based detection:

Various biosensors can monitor bacterial growth by detecting biochemical markers produced by cells. These include quantitative variations in 16S rRNA, fluctuations in NADH and FADH levels, pH shifts, and bioluminescence triggered by genetic modifications (Maugeri et al., 2018). A 16-sensor array, consisting of gold electrodes, was designed with capture probes specific to uropathogens like *Escherichia coli*, *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Enterococcus spp.*, and the *Klebsiella-Enterobacter* group. Bacterial 16S rRNA was hybridized with biotin- and fluorescein-modified probes, and detection was achieved through horseradish peroxidase (HRP)-catalyzed amperometric measurement [22].

Current detection techniques for antibiotic susceptibility tests face several challenges, particularly in terms of time, sensitivity, and cost.

- Time is a significant factor; traditional methods can take 24 to 48 hours to yield results, delaying appropriate treatment decisions and potentially allowing infections to worsen. Rapid testing methods have been developed, but they often require sophisticated technology that may not be widely available.
- Sensitivity is another critical challenge. Many conventional assays may fail to detect low-level resistance or are unable to differentiate between closely related bacterial strains, leading to potential misinterpretation of results and inappropriate treatment choices.
- Cost also plays a crucial role, as advanced testing methods can be prohibitively expensive for many healthcare facilities, particularly in resource-limited settings, thereby limiting their accessibility and implementation.

Improving access to rapid and accurate diagnostic tools is essential for enhancing patient outcomes, as it enables healthcare providers to make informed treatment decisions swiftly and effectively.

In upcoming days, microcantilevers offer a novel approach for antibiotic susceptibility testing by providing high sensitivity and real-time monitoring of bacterial growth and response to various antimicrobial agents. Microcantilevers integrated with an atomic force microscopy (AFM) tip were utilized to investigate the micro-motions exhibited by cellular structures of *E. coli* and *Staphylococcus aureus* (Longo et al., 2013) (Behera et al., 2019). Another detection technique used is surface plasmon resonance (SPR), for the rapid and label free detection of methicillin-resistant *Staphylococcus aureus* (MRSA), methicillin-sensitive *Staphylococcus aureus* (MSSA), and borderline oxacillin-resistant *Staphylococcus aureus* (BORSAs); This methodology involves immobilization of anti-PBP2a antibodies onto a gold-coated sensor surface to specifically capture the penicillin-binding protein 2a (PBP2a), a marker of methicillin

resistance. Upon bacterial binding, SPR was used to detect changes in the refractive index, allowing real-time differentiation between MRSA, MSSA, and BORSA [23].

3. ANTIBIOTIC SUSCEPTIBILITY AND RESISTANCE PATTERN

Antibiotic susceptibility and resistance are critical aspects of bacterial infections, particularly concerning ESKAPE pathogens (*Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Enterobacter spp.*) and *Escherichia coli*. These bacteria are responsible for a significant proportion of hospital-acquired infections and have developed diverse mechanisms to evade antibiotic treatment. *Escherichia coli*, a common Gram-negative pathogen, contributes to both community-acquired and nosocomial infections, including urinary tract infections (UTIs), sepsis, and gastroenteritis. The increasing prevalence of antimicrobial resistance (AMR) among these pathogens poses a serious global health challenge. Resistance mechanisms such as efflux pumps, enzymatic degradation (e.g., β -lactamases), target modification, and biofilm formation reduce the efficacy of antibiotics, limiting treatment options [24]. The ESKAPE group, in particular, is notorious for its ability to "escape" standard antimicrobial therapies, leading to high morbidity and mortality rates.

Antibiotics target bacterial pathogens by inhibiting cell wall synthesis, protein synthesis, nucleic acid function, membrane integrity, or metabolic pathways. Glycopeptides like vancomycin are used against MRSA and *E. faecium* by preventing peptidoglycan cross-linking, weakening the bacterial cell wall. Alternatives include linezolid, which blocks protein synthesis, and daptomycin, which disrupts cell membranes, making them effective against vancomycin-resistant *E. faecium* (VRE). Carbapenem, such as imipenem and meropenem, inhibit cell wall synthesis and treat infections caused by *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*, colistin, a last-resort polymyxin, disrupts Gram-negative outer membranes, treating carbapenem-resistant *A. baumannii* and *Pseudomonas aeruginosa*. Tigecycline is another alternative for carbapenem-resistant *Klebsiella pneumoniae*, though resistance is emerging. Cephalosporins, a subclass of beta-lactams, also weaken bacterial cell walls, while fluoroquinolones inhibit DNA replication by targeting topoisomerase II and IV, preventing bacterial growth [24]. Antibiotics suitable for *Escherichia coli* infections include a variety of classes, each with specific applications based on the strain and resistance patterns observed in local populations. Beta-lactam antibiotics are commonly utilized, encompassing cephalosporins such as cefepime and ceftazidime, which are effective against many strains of *Escherichia coli*. Carbapenems like imipenem, ertapenem, and meropenem offer broad-spectrum coverage, particularly for resistant infections. For uncomplicated urinary tract infections, nitrofurantoin and trimethoprim-sulfamethoxazole are often preferred due to their efficacy against common *Escherichia coli* strains while minimizing the risk of resistance development.

Several novel antibiotics have been developed to combat multidrug-resistant (MDR) ESKAPE pathogens and *Escherichia coli*, targeting different bacterial mechanisms. Plazomicin (Zemdri), an aminoglycoside that binds to the 30S ribosomal subunit, was approved in 2018 for treating complicated urinary tract infections (cUTI) caused by *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *E. cloacae*, and carbapenem-resistant Enterobacterales (CRE). Cefiderocol (Fetroja), a siderophore-cephalosporin targeting penicillin-binding proteins (PBPs), received FDA approval in 2019 for infections caused by CRE, carbapenem-resistant *Acinetobacter baumannii* (CRAB), and carbapenem-resistant *Pseudomonas aeruginosa* (CRPA), particularly in cUTI cases. Imipenem with cilastatin and relebactam (Recarbrio) is a carbapenem- β -lactamase inhibitor combination approved in 2019 for treating infections caused by CRE and CRPA, including cUTI, complicated intra-abdominal infections (cIAI), and hospital-acquired pneumonia (HAP). Another carbapenem- β -lactamase inhibitor combination, meropenem-vaborbactam (Vabomere), was approved in 2017 and is used for CRE infections, especially cUTI and pyelonephritis.

Cefepime-zidebactam (WCK 5222), currently in Phase I trials, has demonstrated broad-spectrum activity against ESKAPE pathogens and pneumococci. Similarly, sulbactam-durlobactam (ETX2514SUL) is in Phase III trials, developed specifically for treating carbapenem-resistant *A. baumannii* infections. Tebipenem-pivoxil (SPR994), an oral carbapenem in Phase III trials, has shown effectiveness against extended-spectrum β -lactamase (ESBL)-producing *E. coli* and *Klebsiella pneumoniae* for home-based treatment of cUTI and pneumonia. These next-generation antibiotics

aim to address the growing challenge of antimicrobial resistance while improving treatment efficacy and accessibility for MDR infections [25].

A study in Germany analyzed 41 samples from two slaughterhouses (S1 and S2) across seven sampling points, including transport areas, stunning facilities, scalding water, eviscerators, production sites, and WWTP influent/effluent. Most *Escherichia coli* isolates were resistant to piperacillin and third-generation cephalosporins, with 3MDRO detected in 50.5% of S1 and 40.9% of S2 samples. Colistin resistance was found in 10.8% (S1) and 8.6% (S2), while all isolates remained susceptible to meropenem, imipenem, and amikacin. Similar resistance patterns were observed in *Klebsiella spp.*, *E. cloacae* complex, and *Citrobacter spp.*, with higher 3MDRO rates in S2 (67.9%). All *A. calcoaceticus*-*A. baumannii* isolates were resistant to cefotaxime, with lower resistance to piperacillin and ceftazidime. Among 58 MRSA isolates, all were resistant to oxacillin, ampicillin, and ceftoxitin, with erythromycin resistance higher in S2 (90.9%). The single VRE isolate from S1 was resistant to multiple antibiotics, including vancomycin and daptomycin (Savin et al., 2020b). In another study based on Germany, high resistance rates were observed in *Enterococcus spp.* against tetracycline (72.2%) and erythromycin (56.6%), while *Escherichia coli* showed significant resistance to ampicillin (42.2%) and sulfonamides (40.2%). Multidrug resistance was detected in both bacterial groups, with some *Escherichia coli* strains exhibiting resistance to three or more antibiotic classes. However, both species remained largely susceptible to ciprofloxacin and gentamicin [26]. A study tested the antibiotic susceptibility of *Salmonella* strains isolated from pork and packaged products in a Romanian slaughterhouse, revealing significant resistance to tetracycline (61.5%), ampicillin (50%), and other antibiotics, highlighting concerns for human health and treatment options [27].

A study examined antibiotic resistance in *Staphylococcus aureus* isolates from meat carcasses and raw milk in South African slaughterhouses and dairy farms. Resistance was highest to penicillin G (71.6%), oxacillin (66.7%), clindamycin (52.9%), erythromycin (48%), and tetracycline (39.2%). Multidrug resistance was common, while susceptibility was observed for ciprofloxacin, chloramphenicol, linezolid, and trimethoprim-sulfamethoxazole. The presence of resistance genes, such as *bla_Z* and *msrA*, indicates potential for resistance spread, underscoring the need for stringent antimicrobial monitoring in food production environments [28]. Another study in Dhaka city tested 35 *Staphylococcus aureus* isolates from meat samples for antibiotic susceptibility. Resistance was found to penicillin (85.71%), ampicillin (71.42%), streptomycin (100%), tetracycline (71.42%), amoxicillin (100%), and neomycin (85.71%), with no resistance to vancomycin or ciprofloxacin [29]. In Ibadan abattoir, Nigeria a study conducted antibiotic susceptibility tests on 30 isolates from retail meat tables, revealing high resistance rates, particularly to tetracycline, with *Escherichia coli* O157:H7 and *Salmonella typhi* showing 60% resistance, highlighting public health concerns [30].

A study analyzed antimicrobial resistance in *Escherichia coli* isolated from retail meats in Washington, DC. High resistance was observed to tetracycline (59%), sulfamethoxazole (45%), streptomycin (44%), cephalothin (38%), and ampicillin (35%), while lower resistance rates were found for gentamicin (12%), nalidixic acid (8%), chloramphenicol (6%), ceftiofur (4%), and ceftriaxone (1%). Multidrug resistance was prevalent, with some isolates resistant to up to eight antibiotics. Three shiga toxin-producing *Escherichia coli* (STEC) isolates were identified, with varying resistance patterns [31]. Antimicrobial susceptibility testing in Germany revealed that *E. coli* isolates from meat samples at slaughter exhibited resistance to 19 out of 26 tested antibiotics, with notable resistance patterns including beta-lactams and multi-drug resistance [32]. Antibiotic susceptibility testing of bacteria in meat samples from slaughterhouses in Faisalabad was performed using the disc diffusion method. Among 80 isolates, 28% were resistant to Cefixime, 20% to Ceftriaxone, 24% to Clavulanic acid, and 30% to Cefotaxime [33].

In India, a study revealed the highest resistance of *Acinetobacter baumannii* prevalence against gentamicin (87.17%), followed by tetracycline (79%), erythromycin (74%), azithromycin (67%), ciprofloxacin (59%), trimethoprim/sulfamethoxazole (56%), and rifampin (51%). However, in this investigation, only one isolate exhibited resistance to tetracycline, while another was resistant to erythromycin [34].

In another study, based on Delhi, India revealed high antibiotic resistance in non-typhoidal *Salmonella enterica* isolates from antibiotic-free (AF) chicken meat, with resistance rates of 87.0% to tetracycline, 53.0% to nalidixic acid, and 41.0% to ampicillin. Moderate resistance was observed to imipenem (28.0%), co-trimoxazole (27.0%), cefazolin

(23.0%), and ciprofloxacin (15.0%). Horizontally transmissible antibiotic resistance genes (ARGs), including tetA (77.0%), qnrS (53.0%), ampC (41.0%), and sul3 (21.0%), were frequently detected. Overall, 74.0% of *S. enterica* isolates were multidrug-resistant (MDR), with 45.0% carrying three or more ARGs, highlighting significant public health concerns [35].

While a study based on Madhya Pradesh showed that, 15 *Escherichia coli* isolates were tested for antibiotic resistance, where all (100%) exhibited resistance to erythromycin and streptomycin. High resistance was also observed against sulphadiazine (95.84%) and cephaloridine (87.50%). Moderate resistance was noted for cephalexin (41.69%), penicillin G (37.60%), ceftiofur and norfloxacin (33.36% each), enrofloxacin (27.40%), and carbenicillin (25.30%). The lowest resistance rates were recorded for amoxicillin and oxytetracycline, with only 16.70% of the isolates showing resistance [36].

4. PHYTOCHEMICAL APPROACH AS ANTIMICROBIALS

Natural antimicrobial approaches have gained increasing attention as promising alternatives to conventional antibiotics, particularly in the wake of rising antibiotic resistance. These strategies involve the use of bioactive compounds derived from plants, animals, and microorganisms that possess inherent antimicrobial properties. Phytochemicals such as essential oils, flavonoids, tannins, and alkaloids have demonstrated broad-spectrum activity against various bacterial strains by disrupting cell membranes, inhibiting enzyme function, or interfering with nucleic acid synthesis. For instance, catechins, a type of flavonoid, have been shown to alter membrane permeability and cause oxidative damage in *Escherichia coli* and *Bacillus subtilis*, leading to cell membrane disruption [37]. Additionally, tannins can inactivate microbial adhesins, enzymes, and cell envelope transport proteins by forming complexes with proteins, thereby inhibiting bacterial growth [38]. Alkaloids, another class of phytochemicals, exhibit antimicrobial properties through various mechanisms, including the inhibition of nucleic acid synthesis [39]. The essential oil of *Origanum minutiflorum*, predominantly composed of carvacrol (73.9%), exhibited potent antimicrobial activity against ciprofloxacin-resistant *Campylobacter* strains. The oil produced inhibition zones ranging from 10 to 28 mm and showed minimum inhibitory concentrations (MICs) as low as 7.8 µg/ml, particularly against strains like *C. lari*. *Salmonella Typhimurium* showed notable sensitivity to combinations of thymol, carvacrol, eugenol, cinnamaldehyde, and allyl isothiocyanate (AIT). In the case of *Escherichia coli*, eugenol was synergistic with tetracycline, while higher concentrations were required for other antibiotics. Thymol and cinnamaldehyde again showed broad-spectrum synergy comparable to their effects on *Salmonella*. *Staphylococcus aureus*, resistant to ampicillin, penicillin, and bacitracin, responded to combinations with essential oils. Eugenol was synergistic with penicillin and ampicillin respectively. Thymol and cinnamaldehyde were effective with ampicillin and bacitracin, with additional synergy observed with penicillin. Carvacrol showed synergistic activity with all three antibiotics, while AIT was effective only with bacitracin. For *Streptococcus pyogenes*, which was resistant to erythromycin, thymol, carvacrol, and AIT restored antibiotic efficacy through synergistic interaction [41]. Primary metabolites like myristic acid, a naturally occurring saturated fatty acid, have demonstrated antimicrobial activity against *Staphylococcus aureus*. Its effectiveness is believed to stem from its ability to disrupt the bacterial cell membrane, impairing vital cellular functions and ultimately inhibiting bacterial growth. *Skimmia anquetilia* exhibited notable antibacterial activity against *Pseudomonas aeruginosa*. The plant's bioactive compounds likely interfered with the bacterial cell membrane integrity or metabolic functions, leading to inhibited growth [41]. Caffeine, a bioactive compound extracted from *Coffea arabica*, has been shown to exhibit antimicrobial effects against *Pseudomonas aeruginosa*, particularly through interference with quorum sensing (QS) pathways. *Pseudomonas aeruginosa* relies on QS systems—such as Las, Rhl, and Pqs—for regulating virulence factor production, biofilm formation, and resistance mechanisms. Caffeine has been found to interact with QS regulatory proteins like LasR and RhlR, either by binding to their active sites or modulating their expression. This interference can suppress the synthesis of virulence factors like elastase, pyocyanin, and rhamnolipids, thereby weakening the pathogen's ability to establish infection and form resilient biofilms [42]. The extracts of *Hibiscus sabdariffa* demonstrated broad-spectrum antibacterial activity against

a diverse group of both Gram-positive and Gram-negative bacteria, including *Staphylococcus aureus*, *Bacillus stearothermophilus*, *Micrococcus luteus*, *Serratia marcescens*, *Clostridium sporogenes*, *Escherichia coli*, *Klebsiella pneumoniae*, *Bacillus cereus*, and *Pseudomonas fluorescens*. The effectiveness of *H. sabdariffa* against such a wide range of clinically relevant bacterial strains suggests the presence of potent phytochemicals capable of disrupting bacterial growth through mechanisms like cell wall interference, enzyme inhibition, or DNA damage [43].

5. CONCLUSION

To conclude, the emergence and dissemination of antibiotic-resistant bacteria in meat sourced from slaughterhouses represent a critical concern for public health, food safety, and the sustainability of antimicrobial therapies. This review underscores the significance of regularly assessing bacterial isolates for their antibiotic susceptibility profiles, as well as understanding the factors contributing to resistance, including excessive antibiotic use in livestock, poor sanitary practices, and inadequate regulatory enforcement. The presence of multidrug-resistant pathogens not only complicates treatment options but also increases the risk of transmission to humans through the food chain. Addressing this issue requires an integrated approach involving stricter antibiotic stewardship in animal husbandry, implementation of robust hygiene and monitoring systems in slaughterhouses, and intensified research into alternative treatments such as natural antimicrobials and bacteriophage therapy. Furthermore, raising consumer awareness and enforcing policy reforms can collectively contribute to controlling the spread of resistance. Ultimately, safeguarding the efficacy of antibiotics necessitates a global, multidisciplinary effort to monitor, understand, and mitigate the drivers of resistance in the meat production sector.

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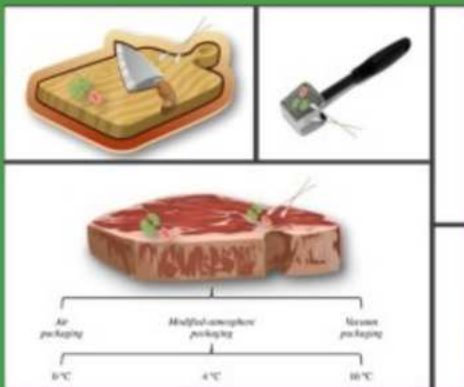
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Slaughterhouse microbes and their rising antibiotic resistance



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