



RESEARCH ARTICLE

Detection of Drug Residues in Beef Samples Collected from Different Slaughterhouses of Lahore, Pakistan

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ABSTRACT

Antibiotics and their metabolites detected in food are hazardous to human health being a causative agent of allergic reactions as well as are responsible for antibiotic resistance in pathogenic microorganisms. The present study was conducted to determine the antibiotic residues in raw beef samples collected from different slaughterhouses of Lahore, i.e., Baker Mandi, PAMCO, Zenith and M. Yousaf & Sons. A total of 150 samples were collected from these slaughterhouses and examined in connection with antibiotic residues. Antibiotic residues demonstrated higher occurrence in beef samples collected from M. Yousaf & Sons (36%) followed by Baker Mandi (32%), Zenith (18%) and PAMCO (14%). Analysis of all the beef samples collected from slaughterhouses showed that 70% were contaminated with antibiotic residues. The results of the present study suggest a serious threat of antibiotic residues in humans due to consumption of contaminated beef sold in these slaughterhouses.

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INTRODUCTION

Antimicrobial drugs are regularly used to treat the animals against various diseases for the survival of animals. Unfortunately, residues of these antibiotics persist in meat due to the lack of knowledge about proper dose, quantity, side effects and withdrawal time from the body system of the animals. The consumption of meat containing antibiotic residues has severe negative consequences on human health and quality of life (Beyene, 2016). Antibiotics are used at vast scale in all types of meat industry, livestock, dairy and poultry due to their easy availability at cheaper rates and effectiveness (Novais *et al.*, 2010).

The antibiotics are administered to animals through various methods (intramuscularly, orally and dietary via food) and thus retain in different body sites of animals accordingly (Berends *et al.*, 2001). Presence of antibiotic residues in the meat of animals sold for human consumption has become a matter of public concern due to their potential hazardous effect on human health when antibiotic residues gain entry into the human food chain. The illegal and frequent use of veterinary drugs causes buildup of antibiotic residues in meat and other animal driven products that pose serious health hazards to

consumers (Prajwal *et al.*, 2017). Recently, the safety of antibiotic residues in the environment has become a matter of increasing public scrutiny and legal requirements (Dahshan *et al.*, 2015).

Microbial inhibition tests are used to detect the antibiotic residues in meat and other products of animals as a screening method, i.e. STAR (Screening Test for Antibiotic Residues) (McGlinchey *et al.*, 2008; Cristofani *et al.*, 2009). The other screening methods are carried out by employing various approaches, i.e., chromatic and immunological (Wang *et al.*, 2017; Sophila *et al.*, 2018).

Microbial screening methods are considered suitable for the determination of antimicrobial drug residues because they are cost effective and less expensive than chromatographic and immunochemical methods. The objective of the present study was to detect the antibiotic residues in the meat samples collected from different slaughterhouses of Lahore.

MATERIALS AND METHODS

The beef samples for the present study were collected from commercially important slaughterhouses of Lahore (M. Yousaf & Sons, PAMCO, Baker Mandi slaughterhouse and Zenith slaughterhouse) and beef

samples were collected from November, 2016 to April, 2017. A total of 140 samples were collected from selected slaughterhouses. The 35 meat samples were collected from each sampling site. Samples were brought to the Department of Zoology, Lahore College for Women University, for further processing.

Procedure for detection of antibiotic residues in beef samples: Microbial inhibition test, a qualitative field disc assay was employed by using Nutrient agar by following the method as described by Hakem *et al.* (2013). A small amount of bacterial strain, *Bacillus subtilis* (Fig. 1) was inserted into the nutrient broth. After 24 hours, the material was spread on the nutrient agar plates and the small disc shaped samples were evaluated for the detection of drug residues by estimating the diameter of zone of inhibition (Malki *et al.*, 2013).

The presence or absence of a transparent zone (Fig. 2) around beef samples was used as an indicator of antibiotic residues in examined samples. Beef samples were considered positive if zone of inhibition around the samples was detected after incubation of 24 hours, while without transparent zone, samples were considered negative for presence of antibiotic drug residues.

RESULTS

During the current study, 70% of beef samples (n=98) sold at different slaughterhouses for human consumption showed the presence of antibiotic residues. The results showed considerable contamination of beef with antibiotics and after the administration of antibiotics, the animals were slaughtered earlier before their withdrawal time for the particular drug.

The highest number of cases contaminated with antibiotic residues was observed in the beef samples collected from the Bakarmandi slaughterhouses, whereas the lowest was recorded in the samples of PAMCO slaughterhouses with a percentage of 28.57%. Beef samples (n=35) collected from Baker Mandi slaughterhouse demonstrated 26 samples with positive results that was 74.28% whereas 9 samples showed negative results. Samples of beef muscle tissue collected from PAMCO revealed 23 samples positive with presence of antibiotic residues, whereas 10 beef muscle tissues collected from the Zenith slaughterhouse after examination by microbial inhibition test exhibited absence of transparent zones indicative of negative results with no antibiotic residues. Out of 70%, the percentage contamination of antibiotic residues in the positive beef samples of different slaughterhouses of Lahore i.e. Baker Mandi, PAMCO, Zenith, M. Yousaf & Sons were recorded as 74.28, 28.57, 37.14 and 68.57%, respectively (Fig. 3).

The present study revealed that 30% of the samples (muscle tissue) appeared negative with no antibiotic residues after examination by field disc method and exhibited no transparent zones around beef sample discs. The lowest number of beef samples containing antibiotic residues was recorded from the Bakamandi (25.72%). The percentage of negative samples without any antibiotic residues ranged from 25.72-71.43% for all the four slaughterhouses of Lahore (Fig. 4).

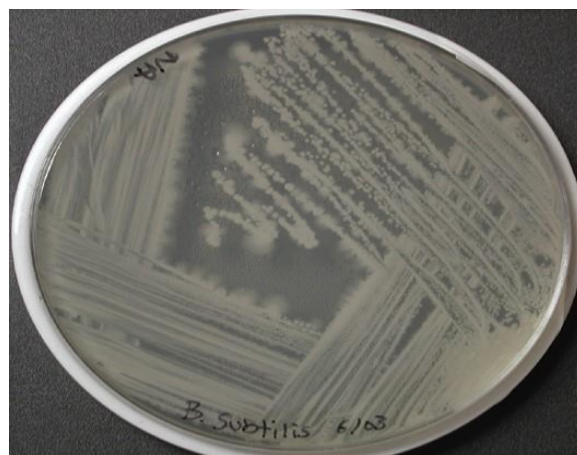


Fig. 1: Lawn of *Bacillus subtilis* for testing of antibiotic residues.

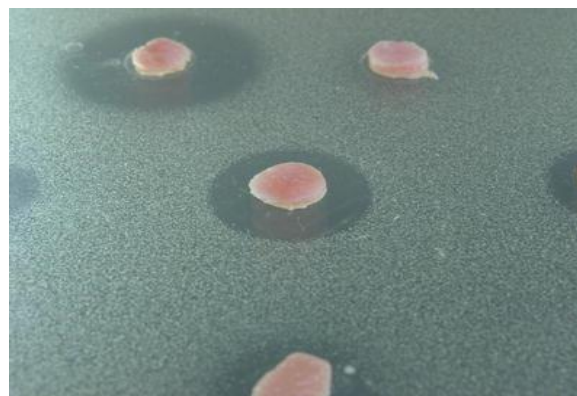


Fig. 2: Transparent zones of inhibition around beef samples

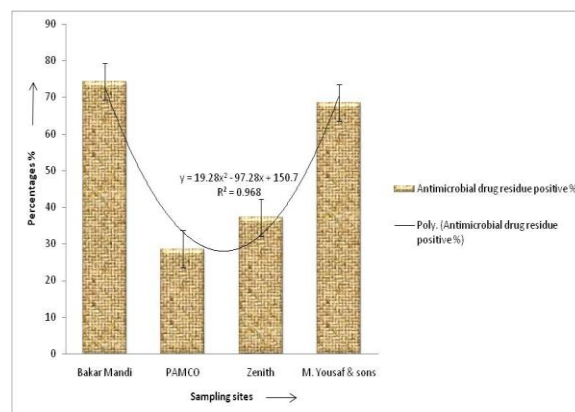


Fig. 3: Percentage of positive samples for antibiotic drug residues in different slaughterhouses of Lahore.

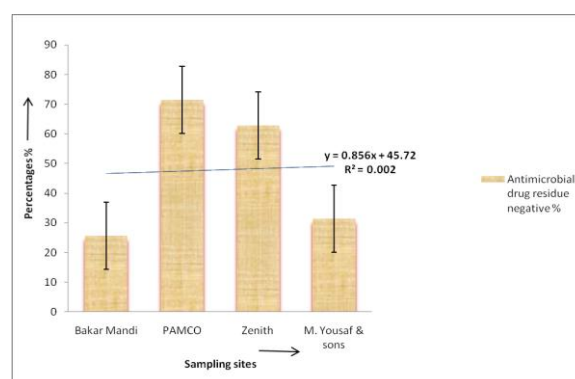


Fig. 4: Percentage of negative samples for antibiotic drug residues in different slaughterhouses of Lahore.

DISCUSSION

Drug residues in meat and other commodities are a growing community health concern due to negligence of proper handling, especially in countries where rules and regulations implemented by official quality assurance authorities for use of antibiotics are bypassed. Consumers are conscious about contamination of meat items by drugs, pesticides and antibodies due to their potential health hazards carcinogenicity, allergic reactions and increased bacterial resistance in humans (Ferrini *et al.*, 2006; Mangsi *et al.*, 2014).

During present study, detection of residues of antibiotics in the collected meat samples of four slaughterhouses (Bakermadi, Pamco, Zenith and M. Yousaf & Sons) of Lahore was performed by Microbial inhibition test. 98 samples out of 150 showed the positive results, i.e., the presence of antibiotic drug residues by showing the zone of inhibition around the samples after incubation of 24 hours. 42 samples did not show any change in them, so they were considered negative for the presence of antibiotic drug residues.

The residual contamination appeared high at the Bakarmandi slaughterhouse with 74.28% positive cases, whereas, it was lowest at PAMCO (28.57%) followed by Zenith (37.14%). The positive cases showing the presence of antibiotic residues at Bakarmandi (74.28%) and M. Yousaf & Sons (68.57%) might indicate that the animals were not held long enough before slaughter or the source of animals at these facilities is different. The lack of lairage facility might be the crucial factor responsible for a higher percentage of positive cases. Hakem *et al.* (2013) also reported 124 cases out of 145 meat samples with high percentage (85.51%) of positive cases having antibiotic residues due to non-compliance of the withdrawal period after antibiotic administration. Ramatla *et al.* (2013) analyzed 150 raw meat samples and 56% were tested positive for antibiotic residues.

The present study demonstrated a high contamination of beef samples with antibiotic residues with 70% positive cases. The results of this study are in agreement with Hamdi *et al.* (2008) who reported 50% positive cases of meat samples containing antibiotic residues. The percentage of positive cases with antibiotic residues showed significant ($P < 0.05$) variations in different studies carried out in various countries as Sanz *et al.* (2015) tested 53% antibiotic residues positive cases in Spain; Randrianomen *et al.* (2006) reported 36.72% positive cases. However, a low percentage of 9.8% has been reported by Alambedji *et al.* (2004) in Senegal and 9.56% by Saitanuet *et al.* (1993) in Thailand. The similar results were reported by Al Ghamdi *et al.* (2000) concerning the quantification of residues of tetracyclines in poultry products in the Eastern Province of Saudi Arabia.

Emergence of more virulent and new disease causing strains of bacteria and human health issues due to consumption of meat contaminated by antibiotic-resistant strains necessitate surveillance and detection of antibiotic residues in meat and meat products to save humans from worst consequences (Wulf and Voss, 2008; Price *et al.*, 2012; Ewers *et al.*, 2012; Liu *et al.*, 2016). During current investigation, 70% of beef samples collected from different slaughterhouses of Lahore exhibited significant

antibiotic residues in meat put on the market for human consumption. Babapour *et al.* (2012) reported 22.8% antibiotic residues after screening 500 beef and mutton samples. However, high level of antibiotic residues and elevated contamination noted in beef samples during present study might be attributed to frequent, prolonged and uncontrolled use of antibiotics by local farmers and livestock farmers at livestock farms without proper guidance and consultation of veterinarians against pathogens to treat the infections.

The high levels of antibiotic residues in beef are also indicative of the early slaughtering of animals without following the recommended withdrawal periods after administration of antibiotics to animals and lack of lairage. Ezenduka *et al.* (2014) also detected antibiotic residues in 42 cases out of 70 with prevalence percentage of 60% in poultry meat by microbial inhibition test. The results are in close agreement with Agmas and Adugna, 2018. Detection of antibiotic residuals might be noted due to before time slaughtering of animals without following withdrawal period recommended for human safety.

Conclusions: Beef samples of all the under-study slaughterhouses of Lahore i.e., Bakermadi, PAMCO, Zenith and M. Yousaf & Sons demonstrated high level of antibiotic residues. Considerable contamination of meat sold at these slaughterhouses might be due to inappropriate management practices, lack of lairage facility and withdrawal periods of the antibiotics after treatment were not followed by the local livestock farmers.

Authors contribution: FM designed the research. AZ, SS and AK conducted the research. FM, GJ and TK analyzed the data and wrote the article. Manuscript was read carefully by all the authors.

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