American Journal of Food Science and Health
Vol. 1, No. 2, 2015, pp. 57-62
http://www.aiscience.org/journal/ajfsh

Exploration the Causes of Infectious Illness and Detection of Antibiotic Residues in Warehouse Poultry

Razia Khatun[1], Md. Abdul Jabbar Howlader[2], Md. Nazrul Islam[1], Md. Khorshed Alam[1], Mohammad Showkat Mahmud[1], Md. Hafizur Rahman[1, *]

[1]Bangladesh Livestock Research Institute, Savar, Dhaka, Bangladesh
[2]Department of Zoology, Jahangirnagar University, Savar, Dhaka, Bangladesh

Abstract

The present study was designed to exploration the causes of infectious illness and detection of antibiotic residues in warehouse poultry of Sirajgonj district, Bangladesh. A total of 96 samples including 24 live broiler birds, 24 samples of poultry litters, water and feed (24 x3=72) were collected from different warehouse of study areas. Among the 96 samples, 24 live broilers were subjected to post mortem examination followed by histopathological examination; in which 5(21%) were suspected to positive for *Salmonella* spp. and 3(13%) were *Mycoplasma* spp. infection. After that serum samples from these birds were tested and positive for agglutination test using specific antiserum of *Salmonella pullorum* (SP) and *Mycoplasma gallisepticum* (MG). In addition 24 samples of litters, water and feed were also tested by microbiological examination (Culture and biochemical test) for exploration the causes of infectious illness in warehouse birds. The microbiological study showed that 91% (22/24) litter samples were infected by *E. coli* and 83% (20/24) by *Salmonella* spp.; 75% (18/24) water samples had mixed infection with *E. coli* and *Salmonella* spp.; and 25% (6/24) feed samples by *Salmonella* spp. and 33% (8/24) by *E. coli*. The ELISA test from meat samples of 24 live broiler birds showed that 16.70% (4/24) breast muscle has residues for Sulfadiazine and Oxytetracycline; 29.17% (7/24) thigh muscle for Oxytetracycline and 16.70% (4/24) for Sulfadiazine; 20.83% (5/24) liver for Oxytetracycline and Sulfadiazine; 29.17% (7/24) for Ciprofloxacin and Enrofloxacin, 8.33% (2/24) for Chloramphenicol and Furazolidone. No Neomycin or Gentamycin residues were detected in all tested samples. The above results showed that bacterial loads in the items used at birds selling centers were high and the content of different drug residues in poultry meat showed great concern for public health ground and need to be more attention.

Keywords

Poultry, Antimicrobial Residues, Microorganisms, Warehouse

Received: April 9, 2015 / Accepted: April 25, 2015 / Published online: May 28, 2015

1. Introduction

Antibiotic residues in different poultry products are a burning issue nowadays for public health ground and the poultry meat is not an exception. The natural products of a microorganism or identical synthetic products or semi-synthetic product that inhibit the growth of or destroy microorganisms are called antibiotics (Kirbis A et al. 2007). In veterinary medicine, antibiotics are excessively used as therapeutic, prophylactive, growth promoters and nutritive purposes in livestock and poultry production (Donoghue D. J et al. 2003 and Committee on Drug Use in Food Animals Panel on Animal

*Corresponding author
Email address: hafiz.vet@gmail.com (M. H. Rahman), rkbaby96@yahoo.com (R. Khatun), jabbarjuiv@gmail.com (M. A. J. Howlader), dgblri56@yahoo.com (M. N. Islam), mkasumon@gmail.com (M. K. Alam), msmvet@yahoo.com (M. S. Mahmud)

Health, Food Safety, and Public Health 1999). Under circumstances when the withdrawal periods of drug is not applied, antibiotic residues may appear in the different animal products such as milk, eggs, meat as reported by various studies (Nonga H. E et al. 2010; Addo K. K et al. 2011; Adi M et al. 2012 and Hind A et al. 2012; Adams H.R 2001). Various antibiotics take different time periods to be excreted from the body. The continuous exposure to antibiotics has been found to lead to serious health problems in humans-such as allergies, spreading of drug-resistant microorganisms, carcinogenic effect and potential harmful effect on human intestinal microflora due to consumes these animal products (Nonga H. E et al. 2010; Ferrinie A. M et al. 2006 and Jafari M.T et al. 2007; Khachatourians G.G and Simonsen G.S et al. 1998; Popelka A et al. 2003; Mariel G. and Pikkemaat 2009).

The presence of antibiotic residues in the foodstuffs of animal origin is one of the most important indexes for their safety. Many livestock producers treat their animals themselves, even if they use the same drugs as veterinarians, they have little understanding of the conditions and quantities to administer or the waiting periods. Currently, approximately 80% of all food-producing animals receive medication for most their lives (Lee H. J et al. 2001). The indiscriminate use of antimicrobials in food-producing animals may result in the presence of residues in foodstuffs of animal origin. The most frequently identified substance in urine, feces and tissue is the parent tetracycline. As much as 30% is excreted unchanged in feces. Tetracycline's are reversibly bound to plasma proteins and are widely distributed. Tetracycline's diffuse throughout the body and found in highest concentrations in kidney, liver, spleen, and lungs. It's also deposited at active sites of ossification (Riviere J. E et al. 2001). Microbial resistance to antibiotics is a worldwide problem in human and veterinary practices (Hassan M. M et al. 2014 and Ahaduzzaman M et al. 2014). Generally it is accepted that the main risk factor for the increase in antibiotic resistance is an extensive use of antibiotics. The antimicrobial agents used in animal care are also significant, not only in increasing the resistance in animal pathogen, but also in bacteria transmitted from animals to humans (Shahid M. A et al. 2007).

The poultry meat consumption practice is very vague here in Bangladesh as there are very few hygienically slaughter houses or warehouses. Moreover people are very reluctant to buy from the slaughter houses or warehouses rather they are very fond of processing live bird from the open market. The people in the market are not very aware about the hygienic lair, slaughter and meat processing. As such the birds are affected by several diseases in the warehouse and the marketing people are forced to use different antibiotics to keep the birds alive/healthy. This might be the major sources of antibiotics in the finished poultry products.

The availability of simply and reliable screening systems for the detection of antibiotics is an essential tool in assuring the safety of food products. In this study a simple and fast method was surveyed for detection of antibiotics residue in chicken meat. Enzyme Linked Immunosorbent Assay (ELISA) test was found sensitive, exact and reliable method for the detection of antibiotic residues. Post mortem and histopathological, serological, cultural and biochemical examinations are available methods for the detection of causative agents of poultry diseases.

From the above facts, it may be mentioned that antibiotic residues might be potential hazards for human as well as animal health and a great obstacle to export poultry meat from Bangladesh. In this context, this research work was undertaken in this country to detect antibiotic residues in warehouse poultry in Sirajgonj district of Bangladesh with encountered the objectives is to give an advice or a suggestion to the people and the policymakers of the country about the source of antibiotic residues in poultry meat and give them a message to change their present poultry meat consumption practice through a biological assessment of the selling centre of poultry birds.

2. Materials and Methods

2.1. Collection of Sample

A total of 96 samples including 24 live broiler birds, 24 samples of water, poultry litters and feeds (24x3=72) were collected from different market points of Sirajgonj district of Bangladesh. Serum samples also were collected from 24 live broiler birds aseptically and transferred to the Bangladesh Livestock Research Institute (BLRI), Savar, Dhaka, Bangladesh for necropsy, histo-pathological examination, Slide agglutination test, ELISA and isolation and identification of micro organisms.

Postmortem examination: The gross examinations of the female reproductive tract followed by postmortem examinations of all the cases were performed at Bangladesh Livestock Research Institute (BLRI), Savar, Dhaka, Bangladesh. The entire tracts were examined carefully following the method of Mc Enttee K (1983).

Histo-pathological examination: During necropsy, various organs having gross lesions were collected, fixed in 10% buffered neutral formalin for histopathological studies. Formalin fixed tissue samples were processed and stained as per standard method (Luna L.G 1968).

2.2. Agglutination Test

Agglutination test was performed on 24 serum samples from 24 live broiler birds against *Salmonella Pullorum* (SP) and *Mycoplasma gallisepticum* (MG) using specific antisera of SP and MG.

Microbiological Examination: For isolation and purification of bacteria from warehouse associated samples various culture media were used included: EMB agar, MacConkey agar, Salmonella-Shigella agar and nutrient agar. Inoculated media were incubated aerobically at 37 ºC for 24 hours. Isolated bacteria were then identified by morphology, Gram stain and Biochemical test e.g. MIU test (Motility, indol and Urease test), TSI (triple sugar iron), Voges Proskauer (VP) test. All biochemical tests were done according to Forbes B.A et al. (2007).

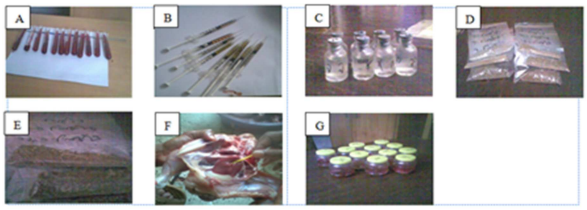

Figure 1. A-B: Blood & serum samples, C: Water, D: Feed, E: Litter, F: Liver swabs, G: Meat samples

2.3. Detection of Antimicrobial Drugs Residue with ELISA

Collected tissue samples were immediately transferred to the Bangladesh Livestock Research Institute with necessary safety measures and maintaining cold chain properly. The tissue samples were stored at -20^0 C until performing the test. Residues of antimicrobial drugs such as Sulfonamide, Oxytetracycline, Enrofloxacin, Chloramphenicol, Furazolidone, Gentamycin and Ciprofloxacin were investigated in collected samples by using Enzyme Linked Immunosorbant Assay (ELISA) according to manufacturer's instructions.

3. Results

Detailed postmortem examination was performed on 24 live broiler birds. On postmortem examination, 5 cases (21%) were detected as *Salmonella pullorum* (SP) and 13% (3/24) were *Mycoplasma gallisepticum* (MG) which were confirmed by histopathology and slide agglutination test using specific antiserum against SP and MG (Table 1).

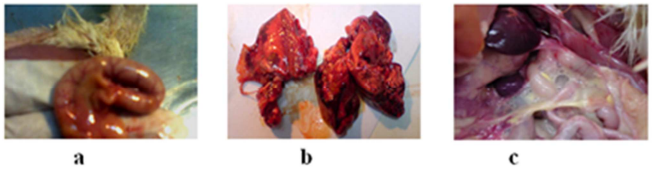

Figure 2. Congestions and hemorrhages in duodenum (a), lungs (b) for Salmonella spp. infection and airsaculitis (c) for Mycoplasma spp. infection.

Table 2 shows that 91% (22) litter samples were infected by *E. coli* and 83% (20) by *Salmonella* spp. In case of water sample 75% (18) had mixed infection with *E. coli* and *Salmonella* spp. and 25% (6) feed samples by *Salmonella* spp. and 33% (8) by *E. coli*.

Table 1. Isolation and identification of bacteria from broiler birds by slide agglutination test.

Sample used	Sample No.	Test	Antisera	Positive	Results
Serum	24	Agglutination test	SP MG	21% (5) 13% (3)	*Salmonella pullorum* (SP) *Mycoplasma gallisepticum* (MG)

Table 2. Isolation and identification of bacteria from water, litter and feed samples of poultry warehouse by cultural and biochemical test.

Sample type	Tested sample	Positive	Results
Litter	24	91% (22) 83% (20)	*E. coli* *Salmonella* spp.
Water	24	75% (18)	*E. coli* and *Salmonella* spp.
Feed	24	25% (6) 33% (8)	*Salmonella* spp. *E. coli*

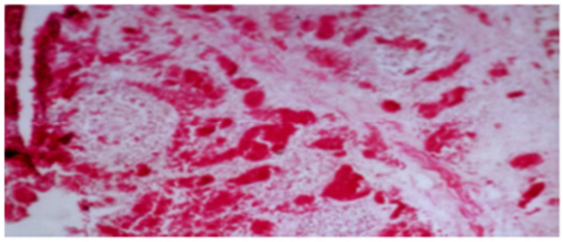

Figure 3. The section of lung in salmonella spp. infection showing severe congestion and infiltration of heterophils, macrophages and lymphocytes in the wall of the bronchus (H & E staining, ×83).

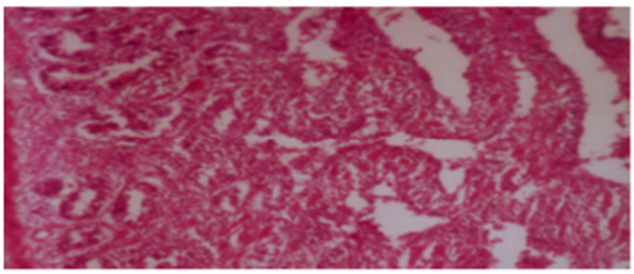

Figure 4. The section of duodenum in salmonella spp. infection showing severe infiltration of leukocytes in the submucosa of the duodenal wall (H & E staining, ×83).

Table 3. Detection of drug residue in poultry meat by ELISA method.

Sample type	Tested sample	% Positive	Drug residue
Breast muscle	24	16.70% (4)	Sulfadiazine and Oxytetracycline
Thigh muscle	24	29.17% (7)	Oxytetracycline
		16.70% (4)	Sulfadiazine
		20.08% (5)	Sulfadiazine and Oxytetracycline
Liver	24	29.17% (7)	Ciprofloxacin and Enrofloxacin
		8.33% (2)	Chloramphenicol and Furazolidone

ELISA test was performed for detection of seven antibiotics and for each type of stored poultry product (liver, breast and thigh muscle samples). The test results showed that 16.70% (4/24) breast muscle has residues for Sulfadiazine and Oxytetracycline; 29.17% (7/24) thigh muscle for Oxytetracycline and 16.70% (4/24) for Sulfadiazine; 20.83% (5/24) liver for Oxytetracycline and Sulfadiazine; 29.17%(7/24) for Ciprofloxacin and Enrofloxacin, 8.33%(2/24) for Chloramphenicol and Furazolidone (Table-3).

4. Discussion

Detailed postmortem examination was performed on 24 live broiler birds. On postmortem examination and agglutination test, 5 cases (21%) were detected as *Salmonella pullorum* (SP) and 13% (3/24) which is supported by Hossain M.S et al. (2006). They noted that out of 70 samples, 8 isolates were identified as *Salmonella* spp. (11.42%), 5 isolates were identified as *Salmonella gallinarum* (7.14%), one isolate was characterized as *Salmonella pullorum* (1.43%). Islam M.M et al. (2006) also found similar results in their experiments such as 33 (21.02%) *Salmonella* spp. were isolated from live and dead birds.

Microscopically, in and lung severe lymphoid cell infiltration was evident in the lumens of the tertiary bronchi, and normal structures of the bronchi and submucosa of duodenum. Heterophilic granulomas with coagulation necrosis in the centers that contained degenerated heterophils and were surrounded by multinucleated giant cells were seen in the lungs (Fig.1 & 2). The above results have similarity with the results of Latife B et al. (2010); Tonu N. S. et al. (2011) and Islam M. M et al. (2006) where they found severe hyperemia and hemorrhagine in the lungs and duodenum.

The microbiological examination performed on 24 samples of litter, water and feeds collected from warehouse revealed that 91% (22/24) litter samples were infected by *E. coli* and 83% (20/24) by *Salmonella* spp. which was agreed with the results of Furtula V et al. (2010); Webb K. E and Fontenot J. P (2013). They reported that broiler litter is a source of antimicrobial residues and represents a reservoir of multiple antibiotic-resistant *E. coli* In case of water sample 75% (18/24) were mixed infection with *E. coli* and *Salmonella* spp. and feed samples, 25% (6/24) were positive for *Salmonella* spp. and 33% (8/24) positive for *E. coli*. Antibiotic residues and antibiotic resistant bacteria from untreated animal wastes (soil, water and litter) can lead to the proliferation of antibiotic resistance in the environment (Li et al. 2010). In case of feed samples, 25% (6/24) were positive for *Salmonella* spp. and 33% (8/24) positive for *E. coli*.

For the detection of antibiotic residues**,** ELISA test was performed for seven antibiotics and for each type of stored poultry product (liver, breast and thigh muscle samples). The ELISA test results showed that 16.70% (4) breast muscle had residues for Sulfadiazine and Oxytetracycline; 29.17% (7) thigh muscle for Oxytetracycline and 16.70% (4) for Sulfadiazine; 20.83% (5) liver for Oxytetracycline and Sulfadiazine; 29.17% (7) for Ciprofloxacin and Enrofloxacin, 8.33% (2) for Chloramphenicol and Furazolidone. No Neomycin or Gentamycin residues were detected in all samples tested which were similar with the results of Shareef A. M et al. (2009); Fujita K et al. (2009); Wang S et al. (2008); Zhang H et al. (2010); Tajik H et al. (2010); Popelka P et al. (2003); Liousia M et al. (2012) and Islam M. M et al. (2006). They tested poultry products like liver, breast and thigh muscle samples for the presence of four (Oxytetracycline, Sulfadiazine, Neomycin, and Gentamycin) antibiotics residue using thin layer Chromatography. They tested 25 samples of each of liver, breast and thigh muscle samples and found 28% of liver and breast muscle were positive for Sulfadiazine and Oxytetracycline while 28% of thigh muscle was positive for Oxytetracycline and 16% samples were positive for Sulfadiazine. No Neomycin or Gentamycin residues were detected on TLC plates in all samples tested.

5. Conclusion

Antibiotic residues in poultry products are crucial issues at the moment for public health. Screening of poultry meat in this study showed that the detectable levels of antibiotic residues which may indicate the widespread misuses of antibiotic in poultry farms as well as in warehouse and the lack of awareness of farmers regarding the recommended withdrawal periods of drugs. The use and sometimes misuse of antimicrobials in food animal production have resulted in the emergence and dissemination of resistant pathogens and resistance genes. Antimicrobial resistant bacteria in food animals can affect not only animal health, but also public health when they enter the food chain. It is recommended that the awareness of the farmers regarding the misuses of drugs, their effect on human health and the rearing systems at birds selling centre need to be improved. It is also necessary

to establish a routine screening program for antibiotic residues by the appropriate authorities.

References

[1] Adams, H. R. (2001). Veterinary pharmacology and therapeutics. 18 edition. Iowa state press. USA, 310-311.

[2] Addo, K. K., Mensah, G. I., Aning, K.G., Nartey, N., Nipah, G. K., Bonsu, C., Akyeh, M. L., & Smits, H. L. (2011). Microbiological quality and antibiotic residues in informally marketed raw cow milk within the coastal savannah zone of Ghana. Tropical Medicine and International Health, 16: 227.

[3] Adi, M., Hind, A., Salman., Nasri, E., Intisar, A. M. & Osman (2012). Detection of antibiotic residues in milk using Delvo test kit and disc assay methods in Khartoum State, Sudan. U of K. Journal of Veterinary Medicine & Animal Production, 3: 3-15.

[4] Ahaduzzaman, M., Hassan, M. M., Alam, M., Islam, S. K. M. A. & Uddin, I. (2014). Antimicrobial resistance pattern against *Staphylococcus aureus* in environmental effluents. Research Journal for Veterinary Practitioners, 2(1): 13-16.

[5] Donoghue, D. J. (2003). Antibiotic Residues in Poultry Tissues and Eggs: Human health concerns. Poultry Science, 82, 618-621.

[6] Ferrinie, A.M., Mannoni, V. & Aurel, P. (2006). Combined Plate Microbial Assay (CPMA): A 6-plate method for Simultaneous First and Second Level Screening of Antibacterial Residue in Meat. Food additives and Contaminants, 23:16.

[7] Forbes, B. A., Sahm, D. F. & Weissfeld, A. S. (2007). Bailey and Scotts' Diagnostic microbiology. 12th ed. Elsevier.

[8] Fujita, K., Ito, H., Yamaguchi, T., Yoneyama, S., Sengoku, K., Sumiya, Y., Watanabe, M., Mizutani, Y. & Hirose, T. (2009). Inter-laboratory validation studies of biological method for determination of tetracyclines in royal jelly. Antibiot Khimioter, 54(9-10), 53-60.

[9] Furtula, V., Farrell, E. G., Diarrassouba, F., Rempel, H., Pritchard, J. and Diarra, M. S. (2010). Veterinary pharmaceuticals and antibiotic resistance of Escherichia coli isolates in poultry litter from commercial farms and controlled feeding trials. Poultry Science, 89(1), 180-8.

[10] Hassan, M. M., Amin, K. B., Ahaduzzaman, M., Alam, M., Faruk., M. S. A. & Uddin, I. (2014). Antimicrobial resistance pattern against *E. coli* and *Salmonella* in layer poultry. Research Journal for Veterinary Practitioners, 2(2): 30-35.

[11] Hind, A., Nasri, E., Adi M., Salman., Intisar, A. M. & Osman (2012). Detection of Antibiotic residues in table eggs using Disc assay and Premi test in Khartoum State, Sudan. U of K. Journal of Veterinary Medicine & Animal Production, 3:16-27.

[12] Hossain, M. S., Chowdhury, E. H., Islam, M. M., Haider, M. G. & Hossain, M. M. (2006). Avian salmonella infection: isolation and identification of organisms and histopathological study. Bangladesh Journal of Veterinary Medicine, 4 (1), 07–12.

[13] Islam, M. M., Haider, M. G., Chowdhury, E. H., Kamruzzaman, M. & Hossain, M. M. (2006). Seroprevalence and pathological study of salmonella infections in layer chickens and isolation and identification of causal agents. Bangladesh Journal of Veterinary Medicine, 4 (2), 79–85.

[14] Jafari, M. T., Khayamian, T., Shaer, V. & Zarei, N. (2007). Determination of Veterinary Drug Residues in Chicken Meat using Corona Discharge Ion Mobility. Spectrometry Analytics Chimica Act, 581: 147.

[15] Khachatourians, G. G. (1998). Agricultural use of antibiotics and the evolution and transfer of antibiotic-resistant bacteria. *Canadian Medical Association Journal*, 159(9), 1129 – 1136.

[16] Kirbis, A. (2007). Microbiological screening method for detection of aminoglycosides, B-lactames, macrolides, tetracyclines and quinolones in meat samples. Slovenian veterinary Researc, 44(1/2): 11-8.

[17] Latife, B., Ayhan, A., Fuat, A., Gumüşsoy, K. S. & Seçil, A. (2010). Pathological and clinical findings and tissue distribution of *Salmonella gallinarum* infection in turkey poults. Turky Journal of Veterinary and Animal Science, 34(2), 101-110.

[18] Lee, H. J., Lee, M. H. & Ruy, P. D. (2001). Public health risks: Chemical and antibiotic residues. Asian Australian Journal of Animal Science, 14(1): 402-413.

[19] Li, Xu., Snow & Daniel (2010). Mitigate and Treat Antibiotic Residues and Antibiotic Resistance Genes in Soil and Water. Report as of FY2010 for 2010NE207B.

[20] Liousia, M., Gousia, P., Economou, E., Sakkas, H., Levidiotou, S., Papadopoulou, C. & Ioannina, G. R. (2012). Detection of antibiotic residues in pork and chicken meat using the STAR test. Polish Journal of Veterinary Science, 15(4), 773-9.

[21] Luna, L. G. (1968). Manual of Histologic Staining Methods of the Armed Forces Institute of pathology. 3rd Erd, McGraw Hill Book Company, New York.

[22] Mariel, G. & Pikkemaat (2009). Microbial screening methods for detection of antibiotic residues in slaughter animals. Analytical and Bioanalytical Chemistry, 395, 893–905.

[23] Mc Entee K. (1983). 15th FAO/SIDA International postgraduare course on Animal Reproduction. College of Veterinary Medicine Uppsala Sweden. Vol 111.

[24] Nita, K. P. (2007). Introduction to the pharmaceutical sciences. Lippincott Williams and Wilkins, 301-304.

[25] Nonga, H. E., Simon, C., Karimuribo, E. D. & Mdegela, R. H. (2010). Assessment of antimicrobial usage and residues in commercial chicken eggs from small holder poultry keepers in Morogoro municipality, Tanzania. Zoonoses.Public Health, 57:339.

[26] Pavlov, A., Lashev, L. & Rusev, V. (2005). Studies on the residue levels of tobramycin in stored poultry products. Trakia Journal of Science, 5, 20-22.

[27] Popelka, P., Nagy, J., Popelka, P. A., Marcincák, S., Jevinová, P. & Hussein, K. (2003). Comparison of bsda and premi® test sensitivity to penicillin standards in poultry meat and after administration of amuril. Folia Veterinaria, 7(3), 73-81.

[28] Riviere, J. E. & Spoo, J. W. (2001). Tetracycline antibiotics. In: Adams, R.H. editor. Veterinary Pharmacology and Therapeutic. 7th ed. Iowa State University Press, Ames, Iowa, USA, 784-796.

[29] Shahid, M. A., Siddique, M., Abubakar, M., Arshed, M. J., Asif, M. & Ahmad, A. (2007). Status of oxytetracycline residues in chicken meat in Rawalpindi/Islamabad area of Pakistan. Asian Journal of Poultry Science, 1(1): 8-15.

[30] Shareef, A. M., Jamel, Z. T. & Yonis, K. M. (2009). Detection of antibiotic residues in stored poultry products. Iraqi Journal of Veterinary Science, 23, 45-48.

[31] Simonsen, G. S., Haaheim, H., Dahl, K. H., Kruse, H., Lovseth, A., Olsvik, O. & Sundsfjord, A. (1998). Transmission of Van A-type vancomycin resistant enterococci and Van A resistance elements between chicken and humans at avoparcin-exposed farms. Microbiology Drug Resist Winter, 4 (4), 313 -318.

[32] Tajik, H., Malekinejad, H., Razavi, R. S. M., Pajouhi, M. R., Mahmoudi, R. & Haghnazari, A. (2010). Chloramphenicol residues in chicken liver, kidney and muscle: A comparison among the antibacterial residues monitoring methods of Four Plate Test, ELISA and HPLC. Food and Chemical Toxicology, 48 (8–9), 2464–2468.

[33] The Use of Drugs in Food Animals: Benefits and Risks (1999). Committee on Drug Use in Food Animals Panel on Animal Health, Food Safety, and Public Health. Academy press Washington, D.C.USA. 71-88.

[34] Tonu, N. S., Sufian, M. A., Sarker, S., Kamal, M. M., Rahman, M. H. & Hossain, M. M. (2011). Pathological study on Colibacillosis in Chickens and Detection of *Escherichia Coli* by PCR. Bangladesh Journal of Veterinary Medicine, 9(1), 17 – 25.

[35] Wang, S., Zhang, H. Y., Wang, L., Duan, Z. J. & Kennedy, I. (2008). Analysis of sulphonamide residues in edible animal products: a review. Journal of Immunology Methods, 337(1):1-6.

[36] Webb, K. E. & Fontenot, J. P. (2013). Medicinal drug residues in broiler litter and tissues from cattle fed litter. Journal of Animal Science, 41(4).

[37] Zhang, H., Zhang, Y. & Wang, S. (2010). Development of flow-through and dip-stick immunoassays for screening of sulfonamide residues. Journal of Dairy Science, 93(9), 3961-4.