Study of the boiling heat on the antibiotic residues in the carcasses of poultry by four plate test

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Background
The discoveries of antibiotics and sulfonamides have been among the great achievements of our time with therapeutic applications in both human and veterinary medicine. They have played an important role in the reduction of morbidity and mortality due to infectious diseases. Subtherapeutic applications are widely used for disease prevention, growth promotion and feed efficiency in livestock and poultry production; they also have applications in control of wildlife, fish, plant diseases and food spoilage. Such a large use of antimicrobial agents are expected to have a great impact on human and animal health, agriculture, ecology and public health. Among possible problems associated with antibiotic use are contamination of food chain products and selection of bacterial populations which are antibiotic resistant. (Smith 1977, Linton 1977, Nazer 1980). The latter may become serious when resistant genes become linked to those which determine pathogenicity.

Objective
The purpose of the present study was to determine the boiling heat effects on the antibiotic residues in the chicken tissues by four plate method.

Methods
Extract samples of breast muscle, liver and kidney were applied to four plate Muler-Hinton agar medium, three of which were inoculated with Bacillus subtilis spores at pH 6, 7.2 and 8 and Staphylococcus aureus at pH 8. Trimethoprim was incorporated into the pH 7.2 medium to enhance the sensitivity of the test for sulphonamide residues. Diffusion of the active antibiotic was detected by the formation of zones of inhibition on one or more plates after overnight incubation.

Results
It was found that 31(12.4%) of 250 carcasses had the residual of antibiotics at one more sites. 11.2% of the breast muscles, 6.8% of the kidneys and 8% of the livers were shown to be contaminated with antibacterial substances (Table 1). For detection of sulphonamide residues, the plates containing trimethoprim were used. Out of 250 samples tested the contamination rate of antibiotics in breast muscles, kidneys and livers were 1.8, 0.8 and 0.2 percent, respectively (Table 2). There was no significant effect of boiling heat treatment on the residual of antibiotics of the specimens collected from slaughterhouse.

Conclusions
A study was conducted to evaluate the residues of antibacterial substance on 250 poultry carcasses. A total of 750 specimens were collected from kidneys, livers and breast muscles of the carcasses. The diffuse four plate test method with the pH of 6, 7.2 and 8 employed for each specimen. Bacillus subtilis and staphylococcus aureus which were sensitive to the inhibitor action of the antibacterial drugs were used.

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Thirty carcasses which didn't show any residual antibiotics were experimentally contaminated with streptomycin, neomycin, oxytetracycline and furazolidone. Boiling heat treatment was administrated on those specimens which were positive for residual of antibiotics as well as 30 specimens from experimentally contaminated carcasses. There was no significant effect of boiling heat treatment on the residual of antibiotics of the specimens collected from slaughterhouse, but there was a significant reduction in residual of antibiotics of the carcasses being contaminated by the streptomycin and furazolidone.

Literature
Linton, A.H., 1977. Antibiotic resistance. *Veterinary Record* 100: S354-360.
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Smith, H.W., 1977. Antibiotic in clinical practice. Pitman Medical, Third Edition: pp. 19-20.

Table 1: Number & percentage of positive antibiotic residues in different organs of chicken extract at different pH values.

pH (organism): Organs tested*	6.0 (B. subtilis)	7.2 (B. subtilis)	8.0 (B. subtilis)	8.0 (S. aureus)	Total
Breast muscle	21 (8.4)	22 (8.8)	21 (8.4)	20 (8.0)	28 (11.2)
Kidney	8 (3.2)	10 (4.0)	7 (2.8)	5 (2.0)	17 (6.8)
Liver	10 (4.0)	12 (4.8)	10 (4.0)	7 (2.8)	20 (8.0)

*Number of samples = 250

Table 2: Number & percentage of positive sulfonamide residues in different organs of chicken extract using trimethoprim at pH 7.2.

Organism: Organs tested*	B. subtilis	S. aureus	Total
Breast muscle	4 (1.6)	5 (2.0)	9 (1.8)
Kidney	2 (0.8)	2 (0.8)	4 (0.8)
Liver	0 (0.0)	1 (0.4)	1 (0.2)

*Number of samples = 250