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### LIST OF ABBREVIATIONS

A/E	Attaching and effacing.
APEC	Avian pathogenic <i>Escherichia coli</i> .
CRD	Chronic respiratory disease.
D+HUS	Diarrhea-associated hemolytic uremic syndrome.
dNTPs	Deoxy nucleoside triphosphates.
E.coli	Escherichia coli.
eae A	Attaching and effacing gene.
EAEC	Enteroaggregative Escherichia coli.
EHEC	Enterohemorrhagic Escherichia coli.
EIEC	Enteroinvasive Escherichia coli.
EMB	Eosine methyline blue.
EPEC	Enteropathogenic Escherichia coli.
ETEC	Enterotoxigenic Escherichia coli.
ExPEC	Extraintestinal pathogenic Escherichia coli.
FC	Fecal coliform.
HC	Hemorrhagic colitis.
HUS	Hemolytic uremic syndrome.
LT	Heat-Labile enterotoxin.
MR/VP	Methyl red/Voges-Proskauer.
PCR	Polymerase chain reaction.
PFGE	Pulsed field gel electrophoresis.
RAPD	Random amplified polymorphic DNA.
SHS	Swollen head syndrome.
ST	Heat-Stable enterotoxin.
St×1	Shiga toxin 1.
St×2	Shiga toxin 2.
STEC	Shiga toxin- producing Escherichia coli.
TBE	Tris buffered EDTA.
TMA	Thrombotic microangiopathies.
TSI	Triple sugar iron agar medium.
TTP	Thrombotic thrombocytopenic purpura.
UPEC	Uropathogenic Escherichia coli.
VFs	Virulence Factors.
VP	Voges-Proskauer.

### 6. Summary

In recent years great extensions was achieved in poultry industry in Egypt, many farms were constructed but most of them not consider the hygienic rules either in construction or breeding system and this leads to enhancement of many avian pathogens. One of these pathogens is *E.coli*, which is a worldwide bacterial avian pathogen affecting all ages and all types of birds, leading to economic losses either due to increase in mortality rate or increase in feed consumption or increase in broiler carcasses rejection at slaughter houses.

The present study includes:

# A. Isolation and identification of *E.coli* from environmental samples:

- A total of 160 samples, air (40), litter (40), feed (40) water (40), were collected from different farms. The samples collected in winter and summer seasons, the result of isolation and identification of *E.coli* from those samples were as follow:

1-The percentage of *E.coli* isolated from environmental samples from different farms was 35.6%.

2-The highest percentage of *E.coli* was recorded in water samples 45% followed in order by litter 40%, air 32.5% and feed 25%.

3-The percentage of *E.coli* was found to be higher in winter than in summer for all samples as air (35 and 30%), water (50 and 40%), feed (30 and 20%) and litter (45 and 35%) for winter and summer respectively.

# **B.** Isolation and identification of *E.coli* from broiler chickens samples:

A total of 40 chickens samples were collected, those samples were
(30) cloacal swabs and (10) internal organs from freshly dead chickens.

1-The percentage of *E.coli* isolated from broiler chickens was 47.5%.

2-The highest percentage of *E.coli* which recovered in winter season 50% which is lower than that recovered in summer season 45%.

# C. Isolation and identification of *E.coli* from persons in contact with chickens:

24 samples from persons in contact with chickens were collected from different farms.

1-The percentage of *E.coli* isolated from persons I contact with chickens was 41.66%.

2-The highest percentage of *E.coil* which recovered in winter season 42.85% which is lower than that recovered in summer season 40%.

#### D. In vitro virulence assay of *E.coli* isolates:

1-The percentage of *E.coli* isolates showing haemolysis with blood agar media was 46.66%.

2-The percentage of *E.coli* isolates showing motility was 72%.

#### E. Serological characterization of *E.coli* isolates:

8 isolates of *E.coli* recovered from broiler chickens and samples subjected to serological identification resulted in:

*E.coli* serogroups recovered from chickens were O<sub>148</sub> (1) and O<sub>26</sub> (2).

*E.coli* serogroups recovered from environmental samples were O<sub>55</sub>,
O<sub>18</sub>, O<sub>27</sub> (1) and O<sub>128</sub> (2).

#### F. Antibiotic sensitivity test of *E.coli* isolates:

In the present study we use 8 different antibiotic discs by using the disc-diffusion method and found that all isolates were resistant to ampecillin (100%) and most of isolates were resistant to tetracycline (85.71%). Also, in this work isolated *E.coli* strains showing moderate degree of resistance to gentamycin (57.14%), norfloxacin (42.85%) and neomycin (28.57%), and lower resistance to tobramycin (71.42%) and streptomycin (42.85%).

# G. Molecular typing of *E.coli* isolates by random amplified polymorphic DNA (RAPD-PCR):

The present study reported the use of RAPD-PCR analysis as a mean genetic typing of avian *E.coli* strains, and investigate the degree of relatedness between strains isolated from chickens and strains isolated from environmental samples.

The results of RAPD-PCR technique carried out on six groups that represent *E.coli* serogroups present in chicken  $1^{st}$  group, air  $2^{nd}$ , litter  $3^{rd}$ , water  $4^{th}$ , and feed  $5^{th}$  and persons  $6^{th}$  groups, cleared that.

1-Different degrees of similarity were found between isolates of chickens and other four groups of environmental samples and human samples as follow:

- The degree of similarity between chickens and litter was 45%.
- The degree of similarity between chickens and water was 52%.
- The degree of similarity between chickens and air was 63%.

- The degree of similarity between chickens and feed was 26%.
- The degree of similarity between chickens and persons samples was 29%.

2-No correlation has been found between the result of *E.coli* serogrouping and that of RAPD-PCR technique.

The obtained results and the hygienic importance of *E.coli* isolated from different samples were discussed.

From the results presented in this study it could be concluded that, *E.coli* could isolated from examined samples in different farms under investigation in either winter or summer seasons. Also the isolation rate was higher in winter than in summer season and so the following hygienic measures should be undertaken to avoid spreading of *E.coli* in poultry farms.

1-A sanitary control measures should be applied to litter by using periodically sound fresh litter.

2-Improvement of air quality especially inside open system broiler farms, this may be achieved by hygienic construction of the farms, good ventilation system and avoid overcrowding of birds.

3-A strict hygienic measure should be applied to improve the water quality, this could be achieved by efficient chemical treatment of water before its use in poultry houses.

4-The choice of feed should be undertaken with strict rule, also hot pelted feed is preferred to be used than fine mashed feed, and also the hygienic conditions of feeders should be considered to avoid contamination of feed from litter or bird droppings. 5-Periodical examination of environmental samples inside the farm to determine the degree of contamination and determine the source of infection with *E.coli*.

6-The down time period should be long enough to allow cleaning and disinfection of the farm, feed and water utensils, also to decrease the survival of microorganisms.

7-Serological identification of *E.coli* revealed that *E.coli* isolated represented by limited number of serogroups in different farms, and so bacterines of the native *E.coli* serogroups should be prepared according to the prevalent serogroups in Egypt.

8-RAPD-PCR technique found to be excellent technique for determination of the genetic relatedness of avian *E.coli* isolates and so it's recommended to be used for tracing the sources of infection of *E.coli* to chickens.

### **Advisor's Committee**