BACTERIAL CAUSES OF DROP IN EGG PRODUCTION IN LAYING HENS AND PREVENTION BY VACCINATION

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ABSTRACT

Twenty-five laying flocks suffered from drop in egg production ranging from 3-10% were examined bacteriologically and the following bacteria were isolated: E.coli (21.9%), Staphylococcus aureus (17.2%) Pseudomonas aeruginosa(11.3%), Proteus vulgaris (6.74%), Enterococci (5.61%), klebsiellaoxytoca (5.41%), proteus mirabilis (5.41%),klebsiella pneumoniae (5%); klebsiellaozaenae (5%), Yersiniaenterocolitica (4.18%), Salmonella (4.08%), Streptococcus (3.77%), Actinomycesbiogenes(3.06%), and Citrobacterfreundii (1.22%). Two hundred and fifteen (215) E.coli isolates were obtained and serotyped as 0166 (45 isolate), 018 (31 isolate), 078 (29 isolate), 01 (28 isolate), 086(17 isolate), o20 (14 isolate) and untypedE.coli strains (13 isolate). The pathogenicity of these serotypes were determined in 9 days old chicks, the E.coli 0166-infected chicks exhibited the higher morbidity and mortality (42.9% - 42.9%) followed by E.coli 078, E.coli 0146,E.coli 020, E.coli 01 and E.coli 086 respectively. Experimental infection of laying hens with E.coli 0166 resulted in a significant decrease in egg production from the first week post infection up to the fifth week (13.5 – 35%) followed by 018 and 078. vaccinated hens showed higher egg production, significant reduction in faecal shedding and egg contamination.

INTRODUCTION

Escherichia coli remains one of the most important pathogens in poultry, causing respiratory infection, cellulites, septicemia, or other diverse clinical condition ***(Barnes and Gross, 1997)***. *E. coli* is present in litter and dust increasing the risk of infection and making colibacillosis difficult to control. In addition, chickens do not naturally develop immunity to pathogenic serotypes. Using antibiotics to control *E. coli* infection results in transient, unreliable protection and often results in subsequent development of drug resistant *E. coli* strains ***(cloud et al.,1985 and DuPont and Steele, 1987)***.

Thus, vaccination could be important in controlling *E. coli*., althoughmonovalent E. coli vaccines did not protect against other *E. coli* serotypes.

Therefore, an effective vaccine against colibacillosis should contain the serotypes of *E. coli* commonly isolated from local farms and associated with this infection ***(Gyimahet al.,1985)***.

Mahmood and Reza, (2010) isolated *E. coli* from oviduct of layer hens (30 – 68 w.), isolates belonged to 11 different serogroups including 01,02,08,015,020,025,036,078,086 and 0111. 078 was the most prevalent serotype followed by 02 and 01. Infection of 1 day old chicks via intraperitoneal or oral route with *E. coli* 0128 produced (90% - 70%) mortality respectively, for 078 (90- 50%), for 0166 (80% - 60%), for 029 (80% - 50%) and for 01 (70% - 60%) respectively, ***(Ahmed, 2012)***.

Hegazyet al., (2012) vaccinated two groups of breeder hens with *E. coli* sonicated or formalized vaccine twice at 20 and 25 week of age followed by Evaluation of antibodies by Elisa and indirect haemagglutination test from hens, eggs and hatched chicks. Sonicated vaccine produced significantly higher titer, protection and body weight than formalinized vaccine. Immunization of breeders protects offspring with maternal antibodies from E. coli infection up to the fourth week of age. The aim of this study is the detection of E. coli causing decreased egg production and prevention by vaccination.

MATERIAL AND METHODS

1- Samples:

One hundered and fifty cloacal swabs from apparently healthy layer chickens. Lung, liver, intestine, ovary and oviduct from 100 diseased layer chickens and 100 freshly dead laying chickens were collected from different farms which suffered from a decrease in egg production and / or high mortalities.

1- Bacterial isolation from collected samples according to ***Buchanan and Gibbons (1974)***.

2- Bacteriological identification, according to ***Cruickshank,et al.,(1975)***.

3- Serotyping, according to ***Edwards and Ewings (1972)***.

4- Streptomycin resistant challenge strains field isolates of *E. coli* 078, 0166, 01 and 018, were rendered streptomycin resistant before experimental use according to ***Barnhart et al.,(1999)***.

5- Bacterial titration: according to ***Sambrooket al.,(1989)***.

6- Preparation of whole cell, formalin inactivated polyvalent *E. coli*bacterin, according to the method of ***Panigraphyet al., (1983)***.

7- Bacteriological examination of eggs: three eggs per group were examined along 5 weeks post infection.

8- Cloacal swabs: six swabs per group were collected along 5 weeks post infection.

2- Birds:

a- Fifty six, 9 days old chicks housed on floor and received prepared ration (21% protein,5.14% fat, 2950 kc/kg).

b- Sixty 19 weeks old laying hens (Bovans) housed on flour and received a prepared ration 18% protein, 5.8% fat and energy 2780 kc/kg.

3- Experimental design:

Experiment (1) study the pathogenicity of the isolated *E.coli* to 9 day oldchicken, table (1).

Experiment (2) aimed to study the effect of isolated *E.coli* strains on egg production, faecal shedding and egg contamination in laying hens and prevention by vaccination table (2).

Statistical analysis: Data were analyzed using SAS statistical analysis system package ***(SAS, 2002)***. One way ANOVA was performed to determine the differences among groups.

Table (1): pathagenicity of *E.coli* serotypes in 9 days old chicks.

Group	Number	Infection	Route	Dose
1	7	*E.coli O78*	Intra tracheal	1×10^8 CFU/Ml
2	7	*E.coli O 18*		
3	7	*E.coli O166*		
4	7	*E.coli O146*		
5	7	*E.coli O20*		
6	7	*E.coli O1*		
7	7	*E.coli O86*		
8	7	Non infected		

Table (2): Efficacy of vaccination of laying hens with prepared polyvalent *E. Coli*bacterin (078, 0166, 01 and 0146) in prevention of *E. coli* infection (n = 6)

Group	No.	Vaccination				E. Coli			
		Type	Age	Dose	Route	Type	Age	Dose	Route
Vacc .ge	1	Poly valent inactivated *E.coli* vaccine	At 19 and 21w of age	0.5ml	S/C	O78	23 w	1×10^8 CFU/ml	Intra tracheal
	2					O 166			
	3					O 1			
	4					O 18			
	5					NI			
Positive control	6	NV				O 78			
	7					O166			
	8					O01			
	9					O18			
negative control	10	NV				NI			

NV = Non Vaccinated NI = Non Infected

RESULTS

Isolation of bacteria:

The incidence of the isolated bacteria was *E.coli* (21.9%), *Staphylococcusaurus* (17.2%), *Pseudomonas aeruginosa*(11.3%), *Proteus vulgaris* (6.74%),*Enterococci* (5.61%), *Klebsiellaoxytoca* (5.41%), *Proteus mirabilis* (5.41%), *klebsiellapneumoniae*(5%), *Klebsiellaozaenae*(5%), *Yersinia enterocolitica* (4.18%), *Salmonella* (4.08%), *Streptococcus species* (3.77%), *Actimomycesbiogenes* (3.06%)

and *Citrobacterfreundii* (1.22%) . Two hundered and fifteen (215) *E.coli* isolates were serotyped as 0166 (45 isolate), 0146 (38 isolate), 018(31 isolate), 078(29 isolate), 01 (28 isolate), 086 (17 isolate), 020 (14 isolate) and untyped*E.coli* strains (13 isolate)

Pathogenicity of *E.Coli* strains to 9 daysold chicks:

E.coli 0166 was more pathogenic for 9 days old chicks, and resulted in higher mortality and morbidity (42.9% and 42.9% respectively) followed by 018 (28.6% and 28.6 respectively). 01 (28.6% and 14.3% respectively), 0146 (14.3% and 28.6% respectively), 078 (zero % and 42.9% respectively), 020 (zero% and 14.3% respectively) and 086 (zero% and 14.3% respectively).

Effect of *E.coli* serotypes on Egg production:

E.coli 0166 infection to laying hens induced significant decrease in egg production starting from the 1st week post infection up to the 5th w.P.i. (16.67%- 35.77%) followed by *E.coli* 01 the drop was from the 2ndw.P.I.(11.43% - 28.57%)

Effect of vaccination with *E.Coli*bacterin on egg production, faecal shedding post challenge, and egg contamination:

No significant increase post vaccination and challenge by 078, 01, 018, while significant increase in egg production was observed post challenge with 0166 (group, 2) from the 1st W.P.I. up to the 5th W.P.I. (100%) in comparison with non vaccinated infected group (7) with egg production (64.23%) table(3). Faecal shedding decreased (078) post vaccination from the 1st WPI from 53.3% to 1.11% in the non vaccinated infected and vaccinated group respectively and reached to zero shedding at the 5th WPI in the vaccinated birdscompaired to 30% in infected non

vaccinated birds, while the bacterin decreased *E.coli* 0166 shedding from 27.8% to 5.55% in non vaccinated infected and vaccinated groups respectively and reached to zero at the end of the experiment in vaccinated birds. (Table, 4).The *E.coli*bacterin decrease the egg albumen contamination with *E.coli* 078, 0166 from 44.4% and 33.3% to zero respectively.

Table (3): Effect of tetravalent bacterin, (078, 0166, 01, 0146) on egg production in *E.coli*- infected Bovans hens

Groups	No of birds	Vaccin-ation	Infection	Egg production % weeks PI				
				1st week	2nd week	3rd week	4th week	5th week
1	6	+ve	*E.coli* 078	$92.86^{ab}\pm3.37$	$92.86^{ab}\pm3.37$	$92.86^{ab}\pm3.37$	$92.86^{ab}\pm3.37$	$95.24^{a}\pm3.07$
2	6	+ve	*E.coli*0166	$100^{a}\pm0.0$	$97.62^{ab}\pm2.3.8$	$97.62^{a}\pm2.38$	$100^{a}\pm0.0$	$90.47^{a}\pm4.96$
3	6	+ve	*E.coli*01	$90.47^{b}\pm4.96$	$90.47^{ab}\pm4.96$	$92.86^{a}\pm3.37$	$92.86^{ab}\pm3.37$	$92.86^{a}\pm3.37$
4	6	+ve	*E.coli*018	$97.62^{ab}\pm2.38$	$95.24^{ab}\pm3.07$	$92.86^{a}\pm4.96$	$90.47^{ab}\pm3.37$	$90.47^{a}\pm4.95$
5	6	+ve	Non infected	$97.62^{ab}\pm2.38$	$92.86^{ab}\pm3.37$	$97.62^{a}\pm2.38$	$97.61^{ab}\pm2.38$	$95.23^{a}\pm3.07$
6	5	-ve	*E.coli*078	$100^{a}\pm0.0$	$100^{a}\pm0.0$	$91.43^{a}\pm4.04$	$91.43^{ab}\pm4.04$	$91.43^{a}\pm4.04$
7	6	-ve	*E.coli*0166	$83.33^{c}\pm1.01$	$73.80^{c}\pm3.37$	$66.66^{c}\pm1.57$	$64.23^{c}\pm2.38$	$64.28^{c}\pm2.38$
8	5	-ve	*E.coli*01	$97.14^{ab}\pm286$	$88.57^{b}\pm4.04$	$80.00^{b}\pm0.0$	$71.43^{c}\pm5.95$	$74.29^{bc}\pm5.71$
9	5	-ve	*E.coli*018	$100^{a}\pm0.0$	$94.29^{ab}\pm3.69$	$91.43^{a}\pm4.04$	$85.71^{b}\pm3.69$	$88.57^{a}\pm4.04$
10	6	-ve	Non infected	$97.62^{ab}\pm2.38$	$100^{a}\pm0.0$	$95.24^{a}\pm3.07$	$100^{a}\pm0.0$	$97.38^{a}\pm2.38$

* All birds of groups (1,2,3,4,5) were vaccinated subcutaneously with 0.5 ml of polyvalent *E.coli*bacterin (078, 0166, 01 and 0146) twice at 19 and 21 weeks of age

**All birds groups (1,2,3,46,7,8,9) were infected intratrachealy with 1x 108 CFU/ml of different *E.coli* serotypes at 23 weeks of age

***Values within the same column bearing different superscripts are significant at $p\leq0.05$

Table (4): Effect of tetravalent *E.coli*bacterin (0.78, 0166, 01 and 0146) on *E.coli*faecal shedding in bovans layers.

Groups	No of birds	Vaccin-ation	Infection	*E.colifaecal* shedding											
				1st week		2nd week		3rd week		4th week		5th week		Total	
				No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
1	6	+ve	*E.coli* 078	2/18	1.11	2/12	16.66	0/12	0.0	1/12	8.33	0/12	0.0	5/66	7.5^{c}
2	6	+ve	*E.coli*0166	1/18	5.55	0/12	0.00	1/12	8.33	0/12	0.0	0/12	0.0	2/66	30.03^{d}
3	6	+ve	*E.coli*01	6/17	35.29	2/12	16.66	1/12	8.33	2/12	16.66	2/12	16.66	13/65	20^{bc}
4	6	+ve	*E.coli*018	5/18	27.77	2/12	16.66	0/12	0.0	3/12	25.0	3/12	25.0	13/66	19.7^{bc}
5	6	+ve	Non infected	0/18	0.0	0/12	0.0	0/12	0.0	0/12	0.0	0/12	0.0	0/66	0.0^{d}
6	6	-ve	*E.coli*078	8/15	53.33	4/10	40.0	2/10	20.0	5/10	50.0	3/10	30.0	22/55	40.0^{a}
7	6	-ve	*E.coli*0166	5/18	27.77	2/12	16.66	3/12	25.0	1/12	8.33	1/12	8.33	13/66	19.7^{bc}
8	6	-ve	*E.coli*01	5/15	33.33	4/10	40.0	1/10	10.0	3/10	30.0	2/10	20.0	15/55	25.9^{b}
9		-ve	*E.coli*018	8/15	53.33	4/10	40.0	4/10	40.0	4/10	40.0	3/10	30.0	23/55	39.7^{a}
10	6	Non vaccinated	Non infected	0/18	0.0	0/12	0.00	0/12	0.0	0/12	0.0	0/12	0.0	0/66	0.0^{d}

* All birds of groups (1,2,3,4,5) were vaccinated subcutaneously with 0.5 ml of polyvalent *E.coli*bacterin (078, 0166, 01 and 0146) twice at 19 and 21 weeks of age

** All birds groups (1,2,3,46,7,8,9) were infected intratrachealy with 1x 108 CFU/ml of different *E.coli* serotypes at 23 weeks of age

*** Values within the same column bearing different superscripts are significant at $p \leq 0.05$

Table (5): Effect of tetravalent *E.coli*bacterin (0.78, 0166, 01 and 0146) on reisolation of *E.coli* serotypes from egg albumen in bovans laying hens.

Groups	No of birds	Vaccin-ation	Infection	*E.coli*resisolation											
				1st week		2nd week		3rd week		4th week		5th week		Total	
				No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
1	6	+ve	*E.coli* 078	0/9	0.0	0/6	0.0	0/6	0.0	1/9	11.11	0/6	0.0	1/36	2.77^{b}
2	6	+ve	*E.coli*0166	1/9	11.11	0/6	0.0	0/6	0.0	0/9	0.0	1/6	16.66	2/36	5.55^{b}
3	6	+ve	*E.coli*01	1/9	11.11	1/6	16.66	0/6	0.0	0/9	0.0	1/6	16.66	3/36	8.33^{ab}
4	6	+ve	*E.coli*018	3/9	33.33	1/6	16.66	0/6	0.0	1/9	11.11	1/6	16.66	6/36	16.7^{ab}
5	6	+ve	Non infected	0/9	0.0	0/6	0.0	0/6	0.0	0/9	0.0	0/6	0.0	0/36	0.0^{b}
6	5	-ve	*E.coli*078	4/9	44.44	0/6	0.0	0/6	0.0	0/9	0.0	0/6	0.0	4/36	11.1^{ab}
7	6	-ve	*E.coli*0166	0/9	0.0	2/6	33.33	1/6	16.66	2/9	22.22	0/6	0.0	5/36	13.9^{ab}
8	5	-ve	*E.coli*01	1/6	16.66	2/6	33.33	0/6	0.0	09	0.0	1/6	16.66	4/33	12.1^{ab}
9	5	-ve	*E.coli*018	4/6	66.66	2/6	33.33	0/6	0.0	2/9	22.22	1/6	16.66	9/33	27.3^{a}
10	6	Non vaccinated	Non infected	0/9	0.0	0/6	0.0	0/6	0.0	0/9	0.0	0/6	0.0	0/36	0.0^{b}

*All birds of groups (1,2,3,4,5) were vaccinated subcutaneously with 0.5 ml of polyvalent *E.coli*bacterin (078, 0166, 01 and 0146) twice at 19 and 21 weeks of age

**All birds groups (1,2,3,4,6,7,8,9) were infected intratrachealy with 1x 108 CFU/ml of different *E.coli* serotypes at 23 weeks of age

***Values within the same column bearing different superscripts are significant at $p \leq 0.05$

Table (6): Effect of tetravalent *E.coli*bacterin (0.78, 0166, 01 and 0146) on reisolation of *E.coli* serotypes from egg yolk In Bovanslayers.

Groups	No of birds	Vaccin-ation	Infection	*E.coli* faecal shedding											
				1st week		2nd week		3rd week		4th week		5th week		Total	
				No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
1	6	+ve	*E.coli* 078	0/9	0.0	0/6	0.0	0/6	0.0	0/9	0.0	0/6	0.0	0/36	0.0^{c}
2	6	+ve	*E.coli*0166	1/9	11.11	1/6	16.66	0/6	0.0	0/9	0.0	0/6	0.0	2/36	$.55^{bc}$
3	6	+ve	*E.coli*01	0/9	0.0	1/6	16.66	1/6	16.66	1/9	11.11	1/6	16.66	4/36	11.1^{abc}
4	6	+ve	*E.coli*018	3/9	33.33	1/6	16.66	0/6	0.0	0/9	0.0	0/6	0.0	4/36	11.1^{abc}
5	6	+ve	Non infected	0/9	0.0	0/6	0.0	0/6	0.0	0/9	0.0	0/6	0.0	0/36	0.0^{c}
6	5	-ve	*E.coli*078	1/9	11.11	0/6	0.0	1/6	16.66	1/9	11.11	0/6	0.0	3/36	8.33^{abc}
7	6	-ve	*E.coli*0166	2/9	22.22	3/6	50.0	0/6	0.0	0/9	0.0	1/6	16.66	6/36	16.7^{ab}
8	5	-ve	*E.coli*01	2/6	33.33	1/6	16.66	2/6	33.33	1/9	11.11	1/6	16.66	7/33	21.2^{a}
9	5	-ve	*E.coli*018	0/6	0.0	2/6	33.33	2/6	33.33	2/9	22.22	0/6	0.0	6/33	18.2^{ab}
10	6	Non vaccinated	Non infected	0/9	0.0	0/6	0.0	0/6	0.0	0/9	0.0	0/6	0.0	0/36	0.0^{c}

*All birds of groups (1,2,3,4,5) were vaccinated subcutaneously with 0.5 ml of polyvalent *E.coli*bacterin (078, 0166, 01 and 0146) twice at 19 and 21 weeks of age

**All birds groups (1,2,3,46,7,8,9) were infected intratrachealy with 1x 108 CFU/ml of different *E.coli* serotypes at 23 weeks of age

***Values within the same column bearing different superscripts are significant at $p \leq 0.05$

DISCUSSION

Bacterial isolation from cloacal swabs and tissuesfrom laying hens resulted in *E.coli* (21.9%),*Staphylococcus aureus*(17.2%),*Pseudomonas aeruginosa*(11.3%),*Proteus vulgaris* (6.74%),*Enterococci* (5.61%), *klebsiellaoxytoca*(5.41%),*Proteus mirabilis*(5.41%),Klebsiellapnemonae (5%),*klebsiellaozaenae*(5%),*Yersinia enterocolitica* (4.18%),*Salmonella* (4.08%), *Streptococcus* species (3.77%), *Actinomycesbiogenes* (3.06%) and *Citrobacter freundii* (1.22%). ***Similarly Mubaraket al., (1998)*** isolated *E.coli* (8.1%), *Salmonella enteritidis* (14.5%), *Proteus Vulgaris* (4.8%), *S.typhyimurium* (14.5%), *Proteus mirabilis* (11.3%), *Klebsiellaoxytoca* (20.9%), *Klebsiellapnemoniae* (11.3%), *Citrbacter*

cloacae (6.5%) and *Yersinia enterocolitica* (8.1%) from ovary and oviduct of freshly dead laying hens. *E.coli* isolates were serotyped as *E.coli* O166 (20.9%), O146 (17.7%), O18 (14.4%), O78 (13.5%), O1 (13%), O86 (7.9%), and O20 (6.5 %), also several studies on laying hens suffered from mortalities and drop in egg production with positive isolation of *E.coli* from oviduct were recorded by ***Vandekerchoveet al., (2004), Zanellaet al., (2000), Mahmood and Reza (2010)*** and ***Oh et al.,(2011)***. Exprimentally *E.coli* O166 was more pathogenic for 9 days old chicks with high morbidity and mortality (42.9% , 42.9%) respectively followed by O18 (28.6% ,28.6%), O1 (14.3% , 28.6%) , O146 (28.6% , 14.3%) , O78 (42.9% , 0%) , O20 (14.3%, 0%) and O86 (14.3% , 0%) respectively.High mortality was recorded by ***Rosenberger et al., (1985)*** and ***Heller et al., (1990).***

Vaccination of laying hens with polyvalent *E.coli*bacterinfollowed by challenge with *E.coli* O166 two weeks p.v. induced significant increase in egg production througout the experiment period, while *E.coli* serotypes O78 and O18 showed non significant increase in egg production. Vaccination with polyvalent *E.coli* was effective in decreasing faecal shedding of *E.coli* serotypes O78, O166 and O18 as early as 1st week post infection. The significant protection from *E.coli* O166 and O1 ,could be attributed to lowering colonization of *E.coli* due to IgA which protect mucosa from *E.coli* infection (Ogra*et al.*,1994). Also vaccination with polyvalent *E.coli*bacterin decreased egg albumen contamination, No *E.coli* O78 reisolation along the experiment except the 4th week p.I. (1+ve from 9), O166 ,0% from the 2nd week up to the end, except the 1st and 5th w.(1 +ve only), while O1 and O18 was still found in faeces and egg albumin which may be due to colonization of

the oviduct with *E.coli* strains.Reisolation of *E.coli* strains from egg yolk post vaccination and infection revealed negative result for O78 along the experiment, while low rate ofreisolation of O166 and O18 was in the 1st two weeks p.i only and 0% up to the end O1 was still found but in lower frequency when compared with infected non vaccinated group. Similar protective effect was reported post vaccination by ***Gyimah and Panigraphy (1985)***, and ***Huang and Matsumoto (1998)***.

It was concluded that different *E.coli* serotypes infection in laying hens cause a variable decrease in the egg production,colonize the intestinal and reproductive tracts and contaminate eggs. Vaccination was efficient in controlling homologus and partially heterologous *E.coli* infection.It is clear that preparation of local vaccine from the most prevalent pathogenic *E.coli* serotypes circulating in the area was protective.

REFERENCES

- ***Ahmed, H.M.(2012):*** characterization of common antegens shared by E .coli strains causing diseases in poultry .Ph.D thesis, dep. of microbiology faculty of vet medicine kafrek sheikh in Egypt.
- ***Barnes, H.J. and gross, W.B. (1997):*** colibacillosis in disease of poultry, 10th ed. Jowa state university press, Ames.P.131-141
- ***Barnhart.E.,Caldwell ,D,croush , M.,Byra, J.,Corrier , D. and Hargis, B. (1999):*** Effect of lactose administration in the drinking water prior to and during feed withdrawal . poultrysci 78:211-214
- ***Buchanon, R.E. and Gibbons, N.E. (1974):*** Bergeyss manual of determinative bacteriology, 8thed Williams and wilkins .co, Blatimose.

- ***Cloud, S.S., Rosenberges , J.K. , Fries , P.A., Wilson , R.A. and Odor , E.M. (1985):*** invitro and invivo characterization of avian *E.coli* serotypes, metabolic activity and antibiotic sensitivity . Avian Dis. 29:1084-1093.
- ***Cruickshank, K.R., Duguiel, J.P., Marmion, B.P. and swain, R.H.A. (1975):*** Medical microbiology . 12th ED vol II Churchill .livingstone, Edenburg . London and New York.
- ***Dupont , H.L. and steele . J.H. (1987):*** use of antimicrobial agents in animal feeds, implications for human health. Rev. infect. Dis. 9:447-460
- ***Edwards, P. R. and Ewings, W. H. (1972):*** Identification of Enterobacteriacae. 3rdEd.,Burgess publishing Co. Unineopoles .
- ***Gyimah, J.E., Panigraphy, B.,Hall, C. F. and Williams, J. D. (1985):*** Immunogenicity of an - Emulsified *E.coli*bacterin against heterologous challenge. Avian Dis. 29 (2) :540-545.
- ***Hegazy ,A. M., Lebda, M. A., Abdelsamie, L. K., Abdallah, H.A. and Abdallah, E. M. (2010):*** Comparative study on sonicated and formalin inactivated *E.coli* vaccines in chickens. 7th Int. Sci. Conf. Mansura. 501-510.
- ***Heller, E.D., Leitner, G., Drabakin, N. and Meamed,D. (1990):*** Passive immunization of chicks against *E.coli* . Avian Pathology. 19: 345-354.
- ***Huang,H. J. and Matsumoto, M.(1998):*** Immunity against *E.coli* infection in chickens assessed by viable bacterial counts in internal organs .Avian Dis. 43: 469-475.

- ***Mahmood, S. and Reza, G.(2010):*** Characterization of *E.coli* isolates from commercial layer hens salpingitis. Amer.J. of animal and veterinary Sci.5(3): 208-214.
- ***Ogra, P.L., Strober, W., Mestecky, J., MeGhee, J. R., Lamm, M. E. and Bienenstock, J. (1994):*** Hand book of Mucosal immunology .Academic press, Inc., San. Diego. New York. Boston. London. Sydney. Tokyo. Toronto.
- ***Panigraphy, B.,Gyimah , J.E. , Hall, C.F. and Williams, J.D. (1983):*** Immunogenic potency of an oil emulsified *E.coli* bacteria . Avian Dis 28(2): 475-481.
- ***Rosenberges , J.K. , Fries , P.A. and Cloud , S.S. (1985):*** Invitro and invivo characterization of avian *E.coli* iii . Immunization Avian Dis. 29(4): 1108-1117.
- ***Sambrook , J., Fritsch , E.F. and Mawiatis , Y. (1989):*** Molecular cloning a laboratory
- ***Vandekerchove , D. , DeHerdt , P. , H. and Pasmans , F. (2004):*** colibacillosis in caged layer hens : characterization of disease and the etiological agent . Avian pathol . 33(2) :117-125.
- ***Zanella, A., Alborali, G.L., Basdotti , M., Candotti , P., Guadagnini, P.F. Annamostino , P. and Stonfer , M. (2004) :*** Severe *E.coli*oiii septicemia and polyserositis in hens at the start lay. Avian pathology 29,311-317

الملخص العربي

الأسباب البكتيرية لانخفاض انتاج البيض في الدجاج البياض و الوقاية بالتحصين في هذه الدراسة تم التعرف علي اهم الامراض البكتيرية و تأثيرها علي إنتاج البيض و الوقاية من الميكروب القولونى بالتحصين.

تم فحص 25 مزرعة دجاج بياض تعاني من نقص في انتاج البيض يتراوح من 3–10 % و ارتفاع في نسبة الوفيات وقد تم عزل البكتريا كالآتي:

E.coli (21.9%), *Staphylococcus aureus* (17.2%),*Pseudomonas aerogenosa* (11.3%), *Proteus vulgaris* (6.74%) *Enterococci* (5.61%) *Klebsiellaoxytoca* (5.41%)*Proteus mirabilis* (5.41%), *Klebsellapnemoniae* (5%) *Klebsiellaozaenae* (5%), *Yersinia enterocolitica* (4.18%), *Salmonella* (4.08%), *Streptococcus species* (3.77%), *Actinomycesbiogenes*(3.06%) and *Citrobacterfreundii*(1.22%)

وقد صنفت معزولات الميكروب القولوني (215) كالآتي:

O166 , O146 , O18 , O78 , O1 , O86 and O20

وتم اختبار ضراوة هذه المعزولات في كتاكيت عمر 9 ايام حيث كانت العترة (0166) هي الاكثر ضراوة و كانت نسبة الوفيات و الطيور المصابة ظاهريا عالية (42.9%–42.9%) مقارنة بالعترات الاخري و تلاها في الخطورة العترة (018) ثم (078) ثم (0146) ثم (020) ثم (01) و أخيرا العترة (086). وبالعدوي التجريبية في الدجاج البياض بالعترة 0166 كان هناك انخفاض معنوي في انتاج البيض بنسبة تتراوح بين (13.5%–35%) من الاسبوع الاول بعد العدوي الي نهايه التجربة ثم تلاها باقل خطورة علي الإنتاج العترات 078 و 018 و 01 .كما اظهر التحصن بلقاح محضر معمليا منالعترات (078 ، 0146 ، 01 ، 0166) كفاءة عالية ضد عدوي الميكروب القولوني حيث وجد ارتفاع معنوي في انتاج البيض بالقطيع المحصن والمعدي بالعترة 0166 كذلك ادي التحصين الي تقليل نسبة اعادة عزل الميكروب القولوني من الزرق و العزل من البيض.