# Molecular detection of *E.coli* virulence genes causing broiler cellulitis at Ismailia Governorate

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**Abstract**

This study was conducted to investigate the existence of virulence genes among *Escherichia coli* strains isolated from cellulitis lesions in broiler chickens**.** Total of (103) defeathered broiler carcasses were collected from different slaughter houses during carcasses inspection at Ismailia province. Samples were collected from moist lesion in subcutaneous tissue and fibrinopurulent material for bacteriological examination. Biochemical and serological identification of *E.coli* isolates were carried out as well as detection of virulence genes (*iss*, *iutA*, *tsh*, *kpsMTII*, *papC*, *stx1*, *traT* and *cvaC***)** *by* PCR protocol. The result showed that *E.coli* was isolated from 51 out of 103 examined carcasses (49.51%). The isolated *E.coli* serologically belonged to O125, O158, O169 and untyped ones. Virulence genes were detected among selected *E.coli* strains derived from cellulitis lesions with variable percentage. All examined isolates were positive for presence of *traT* and *papC* genes, 80% of isolates were positive for *iss*, *iutA* and *kpsMTII* genes, 60% of isolates were positive for *cvaC* gene and 20% of isolates were positive for *tsh* gene. None of the isolates harbored the *stx1* gene.

Key words: Virulence Genes – *Escherichia coli* - Cellulitis - Broiler Chickens - PCR.

## Introduction

*Escherichia coli* causes a variety of diseases in poultry, including respiratory tract infection, omphalitis, swollen-head syndrome, enteritis, septicemia and cellulitis and these diseases are responsible for major economic losses in the chicken industry ***(Gross, 1994 and Norton, 1997).*** Avian cellulitis is an economically important disease of broiler chickens was first reported in 1984 in Great Britain ***(Randall et al, 1984).*** It is an infection of the subcutaneous tissues of the skin, which leads to the production of fibrinous plaques ***(Gross, 1994).*** It has also been called infectious process, inflammatory process, or necrotic dermatitis ***(Barnes and Gross, 1997; Norton, 1997; Dho-Moulin and Fairbrother, 1999).*** Generally, it is associated with a scratch or skin break on the thighs

or lower abdomen of broiler chickens ***(Norton, 1997)***. Cellulitis is not associated with clinical illness and does not seem to affect the growth of the bird ***(Elfadil et al, 1996)***. The infection is generally detected at the time of slaughter ***(Gross, 1994).***

*Escherichia coli* were the predominant bacteria isolated from cellulitis lesions. It appears that the *E.coli* is capable of causing cellulitis must have some special characteristics such as virulence factors. The virulence factors include aerobactin, colicin and cytotoxin are required for the *E.coli* strain to be causative ***(Peighambari et al, 1995 and Norton et al, 2000).*** The virulence genes that permit certain intestinal commensal *E.coli* to become avian pathogenic *E.coli* (APEC) and infect extraintestinal sites include those encoding for the adhesins type 1fimbriae and temperature-sensitive hemagglutinin (*tsh*), iron-scavenging systems and the protectin (*iss*) ***(Dziva and Stevens, 2008).***

Most of these genes are often carried on Colicin V (*ColV*) or other large plasmids and are thought to enable APEC strains to adhere to host tissues survive within host fluids and resist host immune defenses. Different APEC strains may have unique combinations of different virulence factors that have similar functions with regards to disease establishment ***(Johnson et al, 2006 and Mellata et al, 2010).***

The objective of this study was molecular detection of *E.coli* virulence genes causing broiler cellulitis in Ismailia Governorate using polymerase chain reaction (PCR) as a rapid diagnosis for genotypic characterization.

## Material and Methods

### 1- Collection of samples

A total of 103 defeathered broiler carcasses with gross skin lesions (abnormal area characterized by brown discoloration, thickened and scabby) were collected from different flocks in slaughter houses at Ismailia province. The samples were transported in ice to the laboratory, immediately examined and subjected to isolation and identification of *Escherichia coli*.

### 2-Isolation and identification of *Escherichia coli*

A sterile swab was used to collect fibrinopurulent material for bacteriological examinations. All swabs were inoculated in buffer peptone water and incubated at 37˚C for 18±2 hrs under aerobic condition. A loopful from each sample was separately streaked onto MacConkey's agar and Eosin Methylene Blue agar (EMB) and incubated at 37˚C for 24hr. Suspected colonies were picked up, subjected to morphological and Gram staining reaction then streaked on nutrient agar slope for further identification and also in semi-solid agar for detection of motility and finally for preservation. Suspected isolates were confirmed

by a series of biochemical identification according to ***(Murray et al, 2003)*** as well as serotyping by slide agglutination test according to ***(Lee et al, 2009)*** using standard polyvalent and monovalent *E.coli* antisera.

**2-Molecular characterization of *E.coli* isolates using conventional PCR (cPCR):**

Five *E.coli* isolates were subjected to cPCR for detection of these virulence genes **(***iss*, *iutA*, *tsh*, *kpsMTII*, *papC*, *stx1*, *traT* and *cvaC*).

3.1. Extraction of *E.coli* DNA: using **ABIOpure Genomic DNA extraction kit instructions** (Cat. No. M501DP100).

3.2. Preparation of PCR Master Mix According to **Emerald Amp GT PCR master mix (Takara)** Code No. RR310A kit as shown in **Table (2).**

3.3. Cycling conditions of the primers during cPCR: Temperature and time conditions of the primers during PCR are shown in **Table (3)** according to **specific authors** and **Emerald Amp GT PCR master mix (Takara)** kit.

3.4. Agarose gel electrophoreses **(Sambrook *et al*., 1989)**.

DNA Molecular weight marker: using **Gene ruler 100 bp plus DNA ladder** (cat. no. SM0323) supplied from Fermentas with size range: 100-3000 bp.

**Table (1):** *Oligonucleotide primers sequences source: Midland Certified Reagent Company_oilgos (USA).*

| Gene | Primer Sequence 5'-3' | Amplified product | Reference |
|---|---|---|---|
| *tsh* | GGTGGTGCA CTG GAG TGG | 620 bp | **Delicato *et al*. (2003)** |
| | AGT CCA GCG TGA TAG TGG | | |
| *papC* | TGATATCACGCAGTCAGTAGC | 501 bp | **Wen-jie *et al*. (2008)** |
| | CCGGCCATATTCACATAA | | |
| *iss* | ATGTTATTTTCTGCCGCTCTG | 266 bp | **Yaguchi *et al*. (2007)** |
| | CTATTGTGAGCAATATACCC | | |
| *iutA* | GGCTGGACATGGGAACTGG | 300 bp | |
| | CGTCGGGAACGGGTAGAATCG | | |
| *cvaC* | CACACACAAACGGGAGCTGTT | 760 bp | |
| | CTTCCCGCAGCATAGTTCCAT | | |
| *kpsMT*II | CAGGTAGCGTCGAACTGTA | 280 bp | **Ewers *et al*. (2007)** |
| | CATCCAGACGATAAGCATGAGCA | | |
| *stx1* | ACACTGGATGATCTCAGTGG | 614 bp | **Dipineto *et al*. (2006)** |
| | CTGAATCCCCCTCCATTATG | | |
| *traT* | GATGGCTGAACCGTGGTTATG | 307 bp | **Kaipainen *et al*. (2002)** |
| | CACACGGGTCTGGTATTTATGC | | |

**Table (2):** *Preparation of PCR master mix*

| Component | Volume/reaction |
|---|---|
| Emerald Amp GT PCR master mix (2x premix) | 12.5 $\mu l$ |
| PCR grade water | 4.5 $\mu l$ |
| Forward primer (20 pmol) | 1 $\mu l$ |
| Reverse primer (20 pmol) | 1 $\mu l$ |
| Template DNA | 6 $\mu l$ |
| Total | 25 $\mu l$ |

**Table (3):** *Cycling conditions of the different primers during cPCR*

| Gene | Primary denaturation | Secondary denaturation | Annealing | Extension | No.of cycles | Final extension | Reference |
|---|---|---|---|---|---|---|---|
| ***pho*A** | 94˚C 10 min. | 94˚C 45 sec. | 58˚C 40 sec. | 72˚C 45 sec. | 35 | 72˚C 10 min. | **Hu *et al* (2011)** |
| ***tsh*** | 94˚C 5 min. | 94˚C 45 sec. | 54˚C 45 sec. | 72˚C 45 sec. | 35 | 72˚C 10 min. | **Delicato *et al* (2003)** |
| ***papC*** | 94˚C 5 min. | 94˚C 45 sec. | 59˚C 45 sec. | 72˚C 45 sec. | 35 | 72˚C 10 min. | **Wen-jie *et al* (2008)** |
| ***iss*** | 94˚C 5 min. | 94˚C 30 sec. | 54˚C 30 sec. | 72˚C 30 sec. | 35 | 72˚C 7 min. | **Yaguchi *et al* (2007)** |
| ***iutA*** | 94˚C 5 min. | 94˚C 30 sec. | 63˚C 30 sec. | 72˚C 30 sec. | 35 | 72˚C 7 min. | |
| ***cvaC*** | 94˚C 10 min. | 94˚C 1 min. | 63˚C 1 min. | 72˚C 1 min. | 35 | 72˚C 10 min. | |
| ***kpsMT*II** | 94˚C 5 min. | 94˚C 30 sec. | 54˚C 30 sec. | 72˚C 30 sec. | 35 | 72˚C 7 min. | **Ewers *et al*. (2007)** |
| ***stx1*** | 94˚C 10 min. | 94˚C 1 min. | 58˚C 1 min. | 72˚C 1 min. | 35 | 72˚C 10 min. | **Dipineto *et al* (2006)** |
| ***traT*** | 95˚C 5 min. | 94˚C 1 min. | 55˚C 1 min. | 72˚C 1 min. | 35 | 72˚C 7 min. | **Kaipainen *et al* (2002)** |

## Results

### 1- Carcasses inspection:

In all 103 examined cases, the cellulitis lesion was located between the thigh and the ventral midline or in the region of the back and the thighs with yellowish brown discolorations of broiler chicken skin. In some samples, both sides were involved. The lesions varied from small localized lesions (<1 cm) to very extensive ones (>10 cm). The presence of a loosely attached yellow fibrinous plaque

in the subcutaneous tissue was a common feature of most lesions , Ulcerative lesion of cellulitis with underlying pus formation located at thigh as shown in **photo (1).** Other showed thickened and brown discolouration skin in **photo (2)**.

**2- Bacteriological examination:**

The results of bacteriological examination were presented in **Table (4).**

**3- Serotyping:**

The *E.coli* isolates taken from cellulitis lesions were distributed among 3 different O serotype groups (O125, O158 and O169) besides untypable ones.

**4- Molecular characterization of *E.coli* isolates using conventional PCR (cPCR):**

Photo (3), (4), (5), (6), (7) and Table (5) showed the amplification and the detection of 8 virulence genes (*iss*, *iutA*, *tsh*, *kpsMTII*, *papC*, *stx1*, *traT* and *cvaC*) in the five *E.coli* isolates.

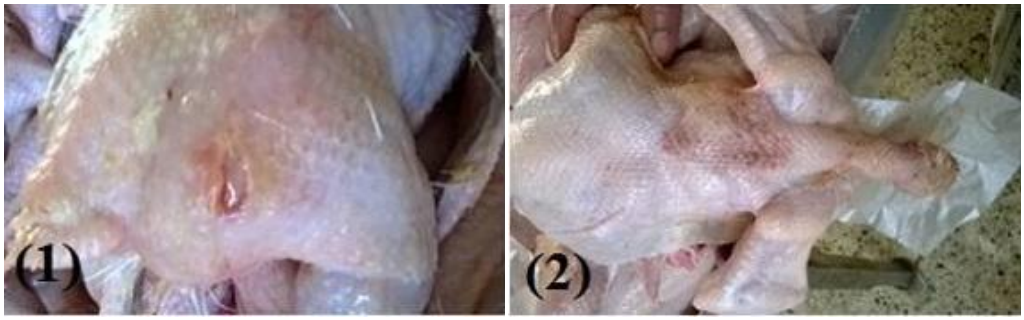


**Table (4):** *Prevalence of E.coli isolated from cellulitis lesions from broiler chickens*

| Type of samples | Total number of the collected samples | Positive *E.coli* isolates | |
|---|---|---|---|
| | | No. | % |
| Thigh | 43 | 20 | 39.2 |
| Breast | 60 | 31 | 60.8 |
| Total S.C samples | **103** | **51** | **49.51** |

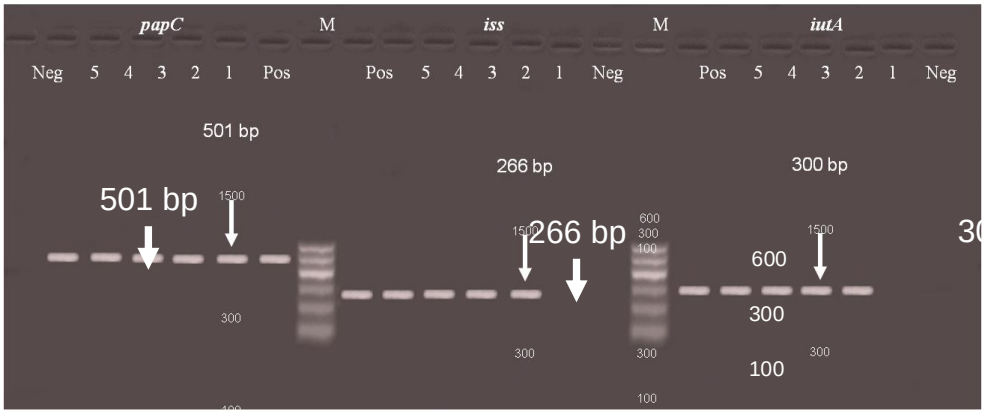


**Photo (3):** Agarose gel electrophoresis of amplified *papC* (501 bp), *iss* (266 bp) and *iutA* gene PCR product (300 bp).

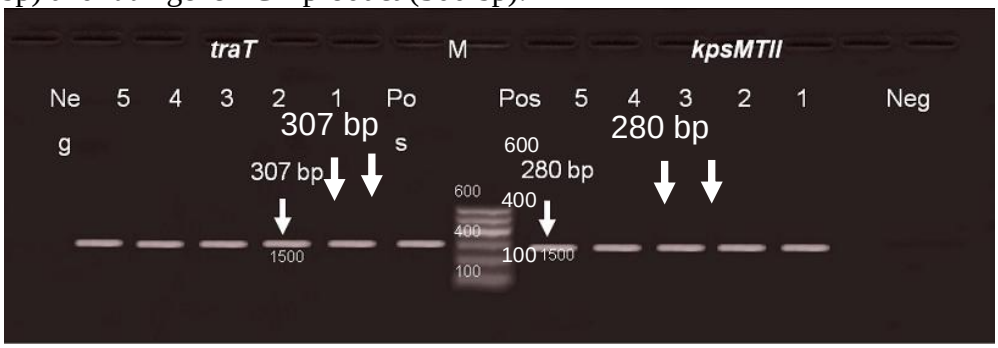


**Photo (4):** Agarose gel electrophoresis of amplified *traT* (307 bp) and *kpsMTII* gene PCR product (280 bp).

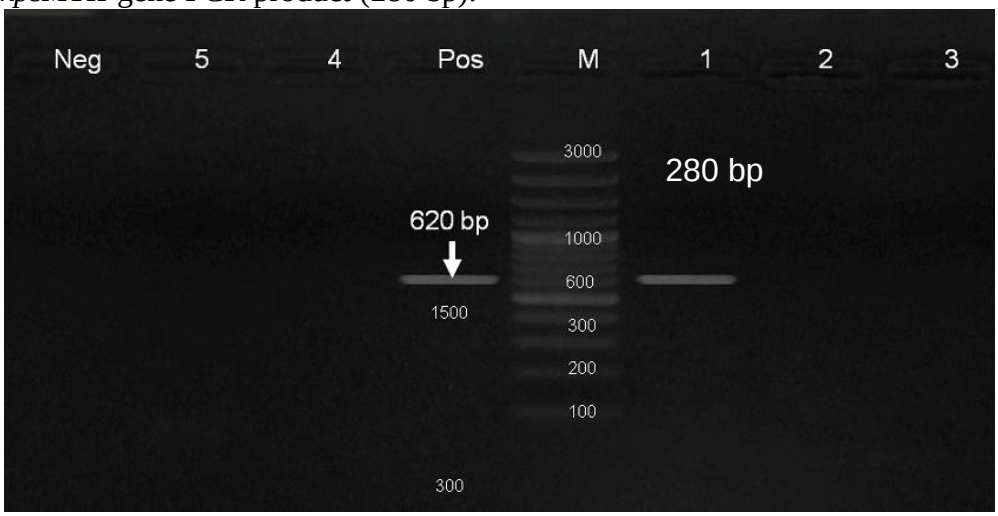


**Photo (5):** Agarose gel electrophoresis of amplified *tsh* PCR product (620 bp).

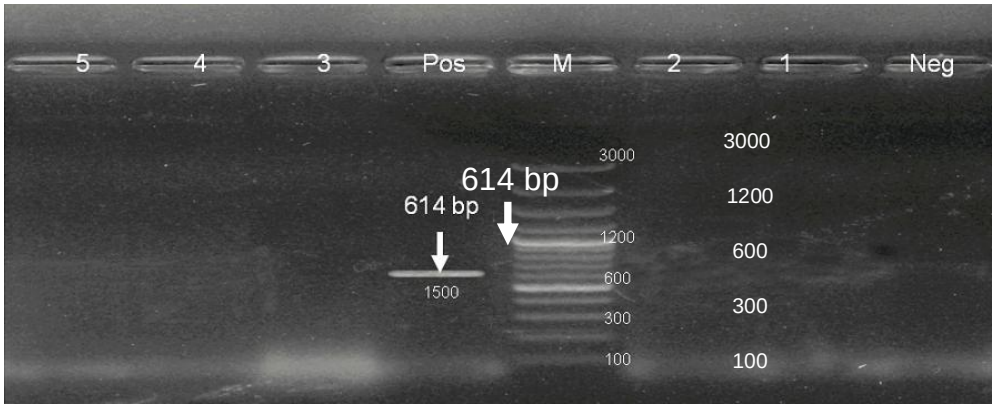


**Photo (6):** Agarose gel electrophoresis of amplified *stx1* PCR product (614 bp).

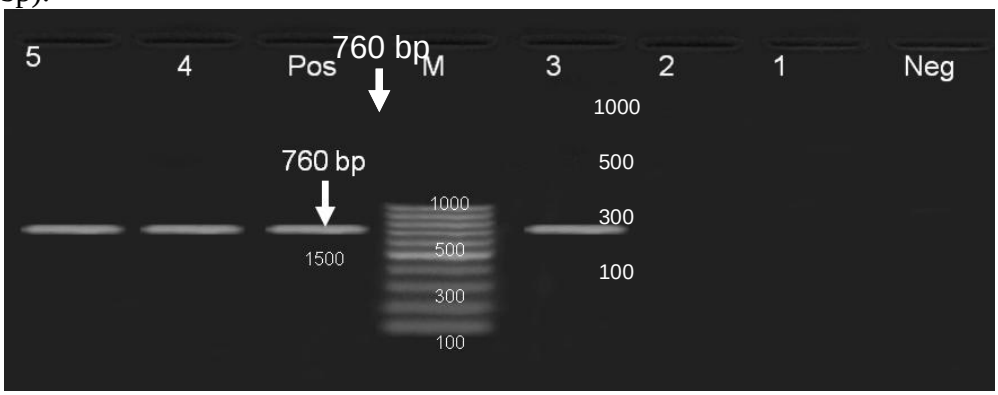


**Photo (7):** Agarose gel electrophoresis of amplified *cvaC* PCR product (760 bp).

**L1:** *E.coli* O169.
**L2:** Untypable *E.coli* (No. 1).
**L3:** *E.coli* O158.
**L4:** *E.coli* O125.
**L5:** Untypable *E.coli* (No. 2).

**Table (5):** *Prevalence of virulence genes (iss, iutA, tsh, kpsMTII, papC, stx1, traT and cvaC) detected by cPCR among examined E.coli strains.*

| Virulence genes | *E.coli* isolates | % |
|---|---|---|
| *iss* | 4/5 | 80 |
| *iutA* | 4/5 | 80 |
| *tsh* | 1/5 | 20 |
| *kpsMTII* | 4/5 | 80 |
| *papC* | 5/5 | 100 |
| *stx1* | 0/5 | 0 |
| *traT* | 5/5 | 100 |
| *cvaC* | 3/5 | 60 |

**Discussion:**

Coliform cellulitis has emerged as a significant disease since its description in 1984 ***(Randall et al, 1984)*** because of increased condemnations, downgrading at processing, and higher labor costs to process affected flocks ***(Lisa et al, 2013).***

Cellulitis is one of the most prevalent extraintestinal infections caused by avian pathogenic *E.coli* (APEC) in broiler chickens and is characterized by the presence of subcutaneous fibrinonecrotic plaques and inflammation of the overlying chicken skin, resulting in rejection of part or all of the carcasses at processing ***(De Brito et al, 2003).***

The results of carcass inspection and gross lesions revealed that , in 103 examined cases, the cellulitis lesions was located between the thigh and the ventral midline or in the region of the back and the thighs with yellowish brown discolorations of the skin of broiler chicken. In some cases, both sides were involved. The lesions varied from small localized lesions (<1 cm) to very extensive ones (>10 cm). The presence of a loosely attached yellow fibrinous plaque in the subcutaneous tissue was a common feature of most lesions . Muscles under the fibrinous plaques usually showed streaky hemorrhages or focal hyperemia, and fluid was present at the site of lesions or ulcerative lesion with underlying pus formation located at thigh in some cases. Other showed thickened and brown discoloration skin. Similar gross lesions were recorded during carcass inspection with cellulitis by ***(Derakhshanfar and Ghanbarpour, 2002 and Lisa et al, 2013).***

*Escherichia coli* isolation incidence was 49.51% from broilers with cellulitis lesions which was lower than that reported by many authors; ***Gomis et al (1997)*** isolated *E.coli* in pure culture from cellulitis lesion by (97.5%). ***Onderka et al (1997)*** isolated *E.coli* from cellulitis lesion by **(**83%). Also ***Derakhshanfar and Ghanbarpour (2002)*** isolated *E.coli* by (91.8%) from cellulitis lesions of examined broiler's carcasses. While, recently ***Barros et al (2013)*** isolated *E.coli* from cellulitis lesion by (82.5%).

Concerning, serotyping results revealed that *E.coli* isolates were distributed among 3 different O serotype groups (O125, O158 and O169) besides untypable ones. The obtained results agreed to some extent with O serotype groups reported by other researchers as ***Gomis et al (1997 & 2001)*** who isolated *E.coli* (O125) and (O158) from cellulitis lesions. While ***Allan (2012)*** isolated *E.coli* (O158) from birds suffered from cellulitis. Also ***Mohamed et al (1997)*** isolated *E.coli* O125 from chickens with facial cellulitis.

However, genotyping characterization has shown that the prevalence of pathogenic *E.coli* in a broiler house was independent of

the prevalence of other *E.coli* in the environment. DNA fingerprinting identified the presence of endemic populations of specific cellulitis-associated *E.coli* existing in the broiler house environment. These organisms persist for at least 6 months, irrespective of partial or complete cleaning and disinfection and cause coliform cellulitis in successive flocks ***(Lisa et al, 2013).*** Although many techniques can be used to identify virulence factors, the PCR still a powerful technique for detection of pathogens because of its rapidity, specificity and sensitivity. It is an effective procedure for generating large quantities of a specific DNA sequence in vitro **(Holland et al, 2000).**

The *E.coli* isolates were subjected for the detection of the virulence genes (*papC, iss, iutA, tsh, kpsMTII, stx1, traT* and *cvaC*) ***(De Brito et al, 2003).***

*E.coli* strains were examined for detection of gene associated with outer membrane protein (genes encoding P fimbriae *papC*) and specific primers were utilized. The results demonstrated that virulence gene (*papC*) was carried by all examined *E.coli* strains isolated from broiler chickens with cellulitis. These results go ahead nearly with ***Gomis et al (2001)*** who found that *papC* gene expressed in 62.4% of *E.coli* isolates.

Some of the virulence factors such as aerobactin, fimbrial antigens and cytotoxins may enhance the ability of *E.coli* isolates to grow within the subcutaneous tissue of broilers ***(Ngeleka et al, 1996).*** Adherence of *E.coli* to the deeper and superficial tissue layers of the skin appears to be important in development of lesion and may be promoted by type 1fimbriae ***(Gyles et al, 2004).*** Most APEC strains express type 1 fimbriae (F1 adhesin) characterized by their ability to bind D-mannose and thus bind to many types of eukaryotic cells. Binding is mediated via FimH subunit which is a minor component of the fimbriae ***(La Ragione and Woodward, 2002).***

*iss* gene in an avian *E.coli* strain is a good indicator of bacterial capacity for causing disease, although its absence does not warrant lack of virulence of *E.coli* ***(Pfaff-McDonough et al, 2000).*** Moreover, the *iss* gene occurs more frequently in APEC than strains from apparently healthy birds ***(McPeake et al, 2005; Rodriguez-Siek et al, 2005).***

*E.coli* strains were examined by PCR for detection of the virulence gene (*iss*) and the result explained that 80% of the examined *E.coli* strains isolated from broiler chickens with cellulitis carried this gene. This concern correlated with ***Jeffrey et al (2002)*** who detected *iss* gene in 72% of *E.coli* isolates, while ***De Brito et al (2003)*** found *iss* gene in 83% and ***Barros et al (2013)*** identified *iss* gene by 87.9 % in *E.coli* isolates from the cellulitis samples of broiler chickens. From

human health hazard point of view this argument leads us to talk about the risks to public health where, the presence of *iss* gene in current assay may indicate a possible pathogenic potential of *E.coli* strains for human since the broiler chickens used in the assay were for human consumption.

*E.coli* uses iron for oxygen transport and storage, DNA synthesis, electron transport, and metabolism of peroxides ***(Neilands et al, 1985).*** Almost all iron in biological fluids is, however, complexed with host iron proteins. A host defense mechanism against bacterial infection is to further reduce the amount of iron available to the invading pathogen ***(Weinberg, 1978).*** Thus, bacteria need effective systems to meet their iron needs during infection. In *E.coli*, the siderophore aerobactin is the most effective of the several iron chelation systems employed by enteric bacteria for iron acquisition ***(Neilands et al, 1985).*** Isolates with the aerobactin system have a growth advantage in low-iron conditions ***(Montgomerie et al, 1984)*** and, in comparison to the other major specialized siderophore, enterobactin, aerobactin is more effective. In most isolates of *E.coli*, aerobactin is expressed by 5 operons, *iucA-D* and *iutA*. *IutA* is coding for the OMP receptor protein. Aerobactin determinants are found both on plasmids and on the bacterial chromosome ***(De Lorenzo et al, 1986).***

The present result showed that *E.coli* isolates harbor aerobactin receptor gene (*iutA*) by 80% of the examined isolates from broiler chickens with cellulitis. This concern completely agreed with ***Gomis et al (2001)*** who found that *iutA* gene identified in *E.coli* isolates by 82.9%, also ***Jeffrey et al (2002)*** and ***De Brito et al (2003)*** detected *iutA* gene by about 92% of *E.coli* isolates.

***De Brito et al (2011)*** evaluated influence of the *iss* and *iutA* genes in the experimental pathogenicity of *Escherichia coli* and declared that the combination of these two genes results in worse lesions, which shows the characteristics of this multifactorial disease. They concluded that the *iss* and *iutA* genes serve as molecular markers for the virulence of cellulitis-producing *E.coli* isolates.

Resistance to normal serum has been associated with *E.coli* causing generalized infection in poultry and extra-intestinal infections in other species. This process is a complement-mediated event and is unrelated to the presence of specific antibodies. The majority of strains derived from birds with cellulitis and other lesions were resistant to killing by normal chicken serum and contained the *traT* gene, suggesting that serum resistance was also advantageous during the production of cellulitis ***(Harel et al, 1993 and Dozois et al, 1994).***

*E.coli* isolates were examined for detection of outer membrane

protein serum resistance gene (*traT*) and it was found that the virulence gene (*traT*) was carried by all the examined *E.coli* strains isolated from broiler chickens with cellulitis. The obtained result agreed with that of ***Barbieri et al (2013)*** who found *traT* gene in 90% (129/144) of *E.coli* isolates from cellulitis lesions and with ***Ngeleka et al (1996)*** detected *traT* gene from *E.coli* isolated from cellulitis lesions by 72% (28/39). In the same context ***Gomis et al (2001)*** and ***De Brito et al (2003)*** detected *traT* gene in 60% of *E.coli* isolates from cellulitis lesions.

The virulence gene (*kpsMTII*) capsule polysaccharide export protein group 2 (K1-K5) was carried by 80% of the examined *E.coli* strains isolated from broiler chickens with cellulitis. This percentage was higher than that reported by ***Barbieri et al (2013)*** who detected *kpsMTII* gene in 37% (53/144) of *E.coli* isolates from cellulitis lesions and ***De Brito et al (2003)*** who detected *kpsMTII* gene in 31% (16/52) of *E.coli* isolates from cellulitis lesions. While the result do not match with ***Ngeleka et al (1996)*** and ***Jeffrey et al (2002)*** who found that the percentage of *kpsMTII* gene in *E.coli* strains isolated from broiler chickens with cellulitis was 8% and 6%, respectively. Where, capsules have been associated with highly virulent avian *E.coli* strains ***(Gross, 1991).*** Capsules are also known to function in the prevention of phagocytosis by host's immune cells ***(Sussman, 1997).*** In the same context, the most common capsule types in avian *E.coli* are K1 and K80 ***(Gross, 1994).*** K1 capsules are poorly immunogenic and increase the serum resistance of *E.coli* strains ***(Dho-Moulin and Fairbrother, 1999)***, and strains that have these capsules are highly virulent ***(Ngeleka et al, 1996).***

Concerning the temperature sensitive hemagglutinin gene (*tsh*), the result revealed that only 20% of the examined *E.coli* strains isolated from broiler chickens with cellulitis carried the virulence gene (*tsh*). This concern goes parallel with ***De Brito et al (2003)*** who detected *tsh* gene in 19% (10/52) of *E.coli* isolates from cellulitis lesions. While this result not matched with ***Barbieri et al (2013)*** who detected *tsh* gene in 66.6% (96/144) of *E.coli* isolates from cellulitis lesions.

Shiga toxin 1 gene (*stx1*) was not detected in any examined *E.coli* strains isolated from broiler chickens with cellulitis. This result agreed to some extent with ***De Brito et al (2003)*** who reported that *stx1* gene carried by only 6% (3/52) of *E.coli* strains isolated from cellulitis lesions from broiler chickens.

Although the role of colicin V in *E.coli* is controversial, production of this bacteriocin may indicate that an organism possesses a battery of properties associated with ColV plasmids, including serum resistance and an aerobactin iron uptake ***(Waters and Crosa, 1991).***

Previous reports indicate that the majority of *E.coli* isolates causing generalized infection in poultry produced colicin V and the siderophore aerobactin ***(Lafont et al, 1987). Ngeleka et al (1996)*** suggested that production of colicin V and production of aerobactin are also common characteristics of *E.coli* strains that cause cellulitis.

It was found that structural gene of colicin V (*cvaC*) was carried by 60% of the examined *E.coli* strains isolated from broiler chickens with cellulitis. This obtained result go ahead with ***Barbieri et al (2013)*** who reported that *cvaC* gene carried by 60% (83/144) of *E.coli* strains isolated from cellulitis lesions from broiler chickens. While ***De Brito et al (2003)*** detected *cvaC* gene in 48% (25/52) of *E.coli* strains isolated from cellulitis lesions from broiler chickens. On the other hand the obtained result was lower than that reported by ***Ngeleka et al (1996)*** who found that *cvaC* gene carried by 92% (36/39) of *E.coli* strains isolated from cellulitis lesions from broiler chickens.

It could be concluded that, the results of this study indicated that cellulitis is caused by pathogenic *E.coli* isolates from different O serotyped groups and non-typed ones, which possess virulence-associated genes varied in their distribution within these O serotypes. Examined strains of *E.coli* possess a characteristic set of virulence factors and that some of these virulent factors may be of public health concern. The occurrence of cellulitis and other diseases caused by *E.coli* and multiple lesions are not estimated, because other types of lesions may not be detected at the time of inspection, as the birds condemned for cellulitis are not examined. *E.coli* avian cellulitis may be associated with other virulence factors that have not been investigated.

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# التحديد الجزيئي لبعض جينات الضراوة للإيشيرشيا كولاي المسببة للإلتهاب الخلوي فى بداري التسمين بمحافظة الإسماعيلية

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**الملخص العربى**

إجريت هذه الدراسة للكشف عن مدى تواجد بعض جينات الضراوة فى عترات الميكروب القولونى المعزولة من إصابات الإلتهابات النسيج الخلوى لبدارى التسمين. تم تجميع عدد ١٠٣ عينة من بدارى التسمين المذبوحة منزوعة الريش من بعض مجازر الدواجن بمحافظة الإسماعيلية. وقد تم تجميع عينات من الأنسجة الرطبة والمتقيحة تحت الجلد وذلك للفحص البكتيرى وقد تم إجراء التصنيف البيوكيميائي و السيرولوجى للعترات المعزولة وكذلك الكشف عن وجود بعض جينات الضراوة مثل(*iss, iutA, tsh, kpsMTII, papC, stx1, traT, cvaC*) بإستخدام إختبار البلمرة المتسلسل. وقد أظهرت النتائج أن النسبة العامة لعزل ميكروب الإيشيرشيا كولاي كانت ٤٩,٥١% (بواقع ١٠٣/٥١). وقد وجد أن عترات الميكروب القولونى المعزولة تنتمى سيرلوجيا إلى O125 ، O158 ، O169 وعترات أخرى لم تصنف سيرولوجيا. وقد خلصت الدراسة إلى أن جميع المعزولات كانت إيجابية لوجود جين *traT* وجين *papC* ، وأن ٨٠% من المعزولات كانت إيجابية لجين *iss* وجين *iutA* وجين *kpsMTII* ، وأن ٦٠% من المعزولات كانت إيجابية لجين *cvaC* ، وأن ٢٠% من المعزولات كانت إيجابية لجين *tsh* ، بينما جميع المعزولات كانت لا تحتوى على جين *stx1*.