Antimicrobial Resistance Genes and Some Virulence Factors in *Escherichia coli* and *Streptococcus pyogenes* Isolated from Mansoura University Hospitals

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ABSTRACT

**Aim:** The present study aims to determine the antimicrobial resistance genes and some virulence factors produced by clinical isolates of *E. coli* and *S. pyogenes* isolated from different sources. **Methods:** A total of 60 *E. coli* and 42 *S. pyogenes* were studied for antibiotic susceptibility pattern, resistance genes, different resistance enzymes production and some virulence factors such as serum resistance, haemolysin, bacterial ability for colonization, haemagglutination, gelatinase, streptokinase and streptodornase. **Results:** Nearly 79.4 % of the isolates are multiple resistant as they were resistant to all antimicrobial classes used in this study. All *E. coli* isolates and only 2 *S. pyogenes* were β-lactamase producers. Detection of bla-TEM gene revealed that it was amplified on plasmid DNA of 23 isolates out of 25 *E. coli* isolates. AGIEs prepared from 19 *E. coli* and 11 *S. pyogenes* isolates exhibited variable inactivation percentages on different aminoglycosides. Chloramphenicol acetyltransferase (CAT) enzyme was detected among 55% of *E. coli* isolates. Gyr A gene was successfully amplified by a specific primer set on plasmid DNA of 36 isolates out of 39 tested *E. coli* isolates which were quinolone resistant. bla-TEM gene was amplified on plasmid DNA of 23 isolates out of 25 B-lactam resistant *E. coli* isolates. While for *S. pyogenes*, Gyr A was detected and amplified on genomic DNA of 14 quinolone resistant isolates and bla-TEM was detected and amplified on genomic DNA of 28 B-lactam resistant isolates. Among 60 *E. coli* isolates, 42 (70%) were serum resistant, 20 (33.3%) showed mannose-resistant haemagglutination, 16 (26.7%) showed mannose-sensitive haemagglutination, 37 (61.7%) were slime producers, 10 (16.7%) were hemolytic and none of *E. coli* isolates was positive gelatinase producer. Among 42 *S. pyogenes* isolates, streptodornase production was detected in 28 (66.7%) of *S. pyogenes* isolates while streptokinase activity was detected in 23 (55%) of *S. pyogenes* isolates.

INTRODUCTION

The discovery and use of antibiotics has been one of the major scientific achievements of the 20th century. During the early period of antibiotic usage, antibiotics were being used to cure potentially lethal infections. However, widespread antibiotic use has promoted the emergence of antibiotic-resistant pathogens, including multidrug resistant strains[1]. Antibiotic resistance is now well recognized as a major problem in the treatment of infections in hospitals and, with increasing and alarming frequency, in the community[2].

*E. coli* is one of the commensals in human intestinal tract. As commensals, it contributes to maintenance of health of a person. However, *E. coli* when enter into unnatural sites, can cause variety of infectious diseases such as urinary tract infections, wound infections, bacteraemiae, meningitis and other soft tissue infections[3]. The ability of *E. coli* to cause extraintestinal infections depends largely on several virulence factors, which help in the survival of *E. coli* under adverse conditions present in those sites[4].

Urinary tract infection (UTI) is one of the most prevalent bacterial infections, and the financial burden on family and society is substantial[5]. Uropathogenic *Escherichia coli* (UPEC) are the most common microorganisms causing UTIs. UPEC strains possess specialized virulence factors, enabling them to colonize and invade to the host, disrupt the host defense mechanisms, injure host tissues, and/or stimulate a noxious host inflammatory response. Virulence factors of recognized importance in the pathogenesis of UTI include diverse adhesins, toxins, siderophores, and polysaccharide coatings[6].

Because UPEC yearly affects a large proportion of the population, they are a major target of antimicrobial therapy. However, the clinical management of UTI is complicated by the increasing incidence of infections caused by
Strains of UPEC that are resistant to commonly used antimicrobial agents\(^9\). The knowledge of drug resistance pattern in a geographical area and the formulation of an appropriate hospital antibiotic policy will go a long way in the control of these infections. Therefore, it is necessary to know the antibiotic susceptibility pattern of UPEC to select the correct antibiotics for proper treatment of infections caused by it\(^9\).

Streptococcus pyogenes (frequently referred to as group A Streptococcus; GAS) is an important human pathogen and causes both mild infections, such as pharyngitis and impetigo, and severe disease, such as toxic-shock-like syndrome and necrotizing fasciitis. GAS infection can also give rise to sequelae such as acute rheumatic fever and acute glomerulonephritis\(^9\). The ability of Streptococcus pyogenes to cause infection depends on a wide array of virulence factors including the M protein or M related proteins, fibronectin binding proteins, the hyaluronic acid capsule, the cysteine protease Spe B, streptokinase, streptolysins O and S, and several phage-encoded exotoxins. Among them, the M protein is the most extensively studied virulence factor of GAS\(^9\). Erythrogenic toxins are major virulence factors of S. pyogenes of utmost importance\(^11\).

The clinical approach to the treatment of streptococcal infections, particularly in the community setting, is traditionally empirical. Therefore, current regional resistance patterns and the mechanisms of resistance should be taken into account when selecting an antimicrobial therapy for a given infection\(^12\). S. pyogenes is globally susceptible to β-lactams; thus, therapy with penicillin or its derivatives is recommended. Nevertheless, it has been described that penicillin has decreased efficacy against severe invasive S. pyogenes infections, such as the toxic shock-like syndrome\(^13\). Macrolides are therapeutic alternatives to penicillin for treating S. pyogenes infections in patients allergic to β-lactams. However, S. pyogenes is frequently resistant to macrolide antibiotics, particularly in European and Asian countries\(^14\). Macrolide resistance in S. pyogenes is usually developed by efflux (encoded by mef A)\(^18\) or target modification due to ribosomal methylation (encoded by erm B or erm A)\(^16\). Resistance of S. pyogenes to fluoroquinolones, although not frequent at present\(^17\), is of concern, and such resistance has been associated with mutations in the quinolone resistance-determining region of both par C and gyrA\(^18\).

The objectives of the present study were to demonstrate the virulence factors and drug resistance in E. coli and S. pyogenes isolated from Mansoura University Hospitals.

**MATERIALS & METHODS**

**I- Clinical strains:**

Sixty strains of E. coli and forty two strains of S. pyogenes were isolated from 177 clinical specimens in Mansoura University Hospitals, Dakahlia governate, Egypt. Specimens were obtained from a variety of clinical lesions including urine, sputum, blood, feces, wounds and throat swabs. The samples were processed immediately using standard procedures and were identified according to Barrow and Feltham, 1993 (19) and Maxted, 1953. (20)

**II- Antibiotic susceptibility testing:**

All E. coli and S. pyogenes isolates were screened for susceptibility to sixteen antimicrobial discs namely; ampicillin (AM, 25 µg), carbenicillin (CB, 100 µg), amoxycillin /clavulanic acid (AMC, 20µg/10µg), cefotaxime (CTX, 30µg), cefazidime (CAZ, 30µg), cefoperazone (CEP, 75µg), ceftriaxone (CRO, 30µg), gentamicin (CN, 10µg), tobramycin (TOB, 10µg), amikacin (AK, 30µg), netilmicin (NET, 30µg), nalidixic acid (NA, 30µg), norfloxacin (NOR, 10µg), ciprofloxacin (CIP, 5µg), levofoxacin (LEV, 5µg) and chloramphenicol (C, 30µg) using the standard disc diffusion method of Chah, and Oboegbulem21. All discs were supplied from Bioanalyse Company.

**III- Qualitative detection of β- lactamase enzyme:**

β- lactamase enzyme was detected according to the method of Chah, and Oboegbulem\(^21\) using nitrocefin (Calbiochem, Germany) solution.

**IV- Bioassay of aminoglycoside-inactivating enzymes (AGIEs) by agar diffusion method:**

Aminoglycoside-inactivating enzymes AGIEs were assayed by agar diffusion method according to the method of Doi et al\(^22\) using Bacillus subtilis ATCC 6633 strain as an assay test organism (it was obtained from the Department of Microbiology, Faculty of pharmacy, Mansoura University).

**V- Detection of Chloramphenicol acetyltransferase (CAT) enzyme:**

All E. coli isolates were tested for the production of CAT enzyme. According to the method of Satish et al\(^23\).
VI- Detection of some resistance genes
1. Plasmid extraction:
   Plasmid DNA was extracted from *E. coli* isolates by alkaline lysis method[24].

2. Polymerase Chain Reaction for the Amplification of Resistance Genes:
   The resistance genes (gyrA and bla-TEM) were detected and amplified using the following reaction: 12.5 µl Hot Star Taq master mix (dNTPs, MgCl2 and polymerase), 1 µl of forward primer (10 µM), 1 µl of reverse primer (10 µM) (table 1), 5 µl of the DNA template and 5.5 µl of nuclease free water were added for a total of 25 µl per reaction[25]. Colony PCR was used for amplification of resistant genes in *S. pyogenes*. The pipette was held like a pencil to touch a colony to a tip. Only a single colony, not agar was picked. The colony was transferred to the PCR mixture and the reaction mixture was pipetted up and down 4 or 5 times[26].
   
   The PCR was started with initial DNA denaturation and polymerase activation for 15 min at 95°C, followed by 30 cycles of denaturation for 30 sec. at 94°C, annealing for 90 sec. at 55°C, and extension for 90 sec. at 72°C and a final extension step for 10 min at 72°C[27].

| Table (1): Specific amplification primer sets for the tested resistance genes |
|--------------------------|------------------|---------------------|
|Gene name | Type | Sequence |
| GyrA | Fw | CTGGAAACAAGCCTATAAAT |
| | Rv | Universal-ACCGAGAAGCCTGCGCAGT |
| Bla-TEM | Fw | CCCCGAAGAAGCTTCCAT |
| | Rv | Universal-CTTACCATCTGGCCCAT |

Fw: Forward primer  Rv: Reverse primer

VII- Studies on some virulence factors of *E. coli*:
1. Serum resistance:
   One hundred and fifty microlitres of serum was mixed with 50 µl of bacterial suspension in a 96 well micro plate. The initial absorbance at 620 nm was measured, and compared with the absorbance after 3 hrs of incubation, using a micro plate reader[28].

2. Haemagglutination:
   *E. coli* isolates were inoculated into 1% nutrient broth and incubated at 37 °C for 48 hrs for full fimbriation. On a glass slide, one drop of the RBC suspension was added to a drop of the broth culture and slide was rocked at room temperature for 5 min. Presence of clumping was taken as positive for haemagglutination[2].

3. Bacterial ability for colonization
   The bacterial ability to colonize the abiotic surface was quantified by slime test. The tested isolates were cultivated in tubes of nutrient broth and incubated at 37 °C for 24 hrs and thereafter the cultures tubes were further emptied and stained with safranin alcoholic solution 1 % for 30 min, washed three times with distilled water and left at room temperature for 24 hrs. The intensity of the red ring on the tube wall was recorded as ++, +++, ++++[29].

4. Hemolysin production
   All *E. coli* isolates were tested for production of hemolysin on blood agar plates according to the method of Panus et al.[29].

5. Gelatinase test:
   Gelatin agar plates were inoculated with the tested organism and incubated at 37°C for 24 hrs. The plate was then flooded with mercuric chloride solution. Development of opacity in the medium and zone of clearing around colonies were considered positive for gelatinase production[8].

VIII- Studies on some virulence factors of *S. pyogenes*:
1. Streptodornase production
   The detection of streptodornase depends on DNase activity observed in agarose gel electrophoresis by fading or disappearance of DNA band under UV-lamp[30].

2. Streptokinase activity
   Two hundred microlitres of thromboplastin prewarmed at 37°C in water bath for 30 min was added to 100 µl of diluted plasma (1:5) in 0.1 M phosphate buffer pH 7.2 prewarmed at 37°C in water bath for 30 min and clot formation was allowed to take place. One ml of the solution to be tested for streptokinase activity was added. Clot
lysis was observed within 30 min depending on streptokinase activity\[31\].

RESULT

Antibiotic susceptibility testing

All *E. coli* isolates were resistant to ampicillin, carbenicillin and ceftazidime. Most isolates of *E. coli* were resistant to cefotaxime (78%), cefoperazone (85%), ceftriaxone (93%), nalidixic acid (78%), norfloxacin (75%), ciprofloxacin (82%), levofloxacin (67%) and amoxicillin / clavulanic acid (67%). Regarding *S. pyogenes*, most isolates were resistant to both carbenicillin and nalidixic acid (98%), ceftazidime (95%), ampicillin (88%). Only 60% of *S. pyogenes* were resistant to amikacin, 57% were resistant to tobramycin, 50% were resistant to cefotaxime, 45% were resistant to gentamicin, 43% were resistant to norfloxacin and 38% were resistant to ciprofloxacin and netilmicin. In contrast, most *S. pyogenes* isolates were sensitive to amoxicillin /clavulanic acid (83%), levofloxacin (79%), Chloramphenicol (76%) and cefoperazone (71%).

Qualitative detection of β- lactamase enzyme

Using the nitrocefin solution, all *E. coli* isolates (100%) showed red color and considered positive β-lactamase producers Fig. (1). In contrast, only 2 isolates (4.7%) of *S. pyogenes* (isolates No. 17 and 36) showed color change from yellow to red and so considered positive β-lactamase producers.

**Bioassay of Aminoglycoside-inactivating enzymes (AGIEs) by agar diffusion method:**

Crude extracts prepared from 19 *E. coli* and 11 *S. pyogenes* isolates were used for determination Aminoglycoside Phosphotransferases (APHs) and Aminoglycoside Nucleotidyltransferases (ANTs) using ATP as a cofactor and Aminoglycoside Acetyltransferases (AACs) using acetyl co-A as a cofactor. The percentage inactivation of gentamicin, amikacin and streptomycin was shown in Fig. (2 -3).
Fig (2): Determination of % inactivation of gentamicin, amikacin and streptomycin by AGIEs isolated from different *E. coli* isolates using ATP and acetyl co-A as cofactors.
Fig. (3): Determination of % inactivation of gentamicin, amikacin and streptomycin by AGIEs isolated from different *S. pyogenes* isolates using ATP and acetyl co-A as cofactors.
Detection of (CAT) enzyme in *E. coli* isolates:

Seven isolates (No. 5, 35, 38, 43, 44, 50 and 59) reduced complete inactivation of Chloramphenicol and 26 isolates produced partial inactivation of Chloramphenicol where they produced percentage decrease in inhibition zone ranging from 3% to 47% and so considered CAT producers. In contrast 27 isolates showed 0% decrease in inhibition zone diameter compared with the control and so considered non CAT producers (fig. 4)

![Detection of CAT in E. coli isolates from No. 45 to 50](image)

*Fig. (4)*: Detection of CAT in *E. coli* isolates from No. 45 to 50 C : Control Chloramphenicol. Isolate No. 45 and 48 were non CAT producers. Isolate No. 46, 47, 49 and 50 were non CAT producers.

Polymerase chain reaction for the amplification of quinolone and β-lactamase resistance genes:

*GyrA* gene was amplified in all tested *E. coli* isolates except isolates No. 15, 22 and 59 and was amplified on genomic DNA of 14 quinolone resistant *S. pyogenes* isolates. On the other hand *bla-TEM* gene was amplified in all tested *E. coli* isolates except isolates No. 15 and 18 (fig 5-7).

![Agarose gel electrophoresis of bla-TEM amplicons generated by PCR](image)

*Fig. (5)*: Agarose gel electrophoresis of *bla-TEM* amplicons generated by PCR. Lane M was 1 Kb ladder molecular weight marker. Lanes 1 to 13 were amplicons from *E. coli* isolates No. 9, 14, 15, 16, 17, 18, 19, 20, 21, 24, 25, 31 and 33 respectively.
**Serum Resistance:**

The remaining absorbance after 3 hr (OD 620,3 h) was higher than 100% relative to the initial absorbance in 42 *E. coli* isolates (70%) so they were designated serum resistant while the remaining absorbance after 3 hrs was less than 100% relative to the initial absorbance in 18 isolates (30%) therefore they were considered serum sensitive.

**Haemagglutination:**

Clumping of erythrocytes in the presence of D-mannose was observed in 20 *E. coli* isolates (33.3%) and this was considered mannose resistant haemagglutination (MRHA) while clumping of erythrocytes only in absence of mannose was observed in 16 *E. coli* isolates (26.7%) and this was considered mannose sensitive haemagglutination (MSHA). On the other hand 24 *E. coli* isolates (40%) produced no haemagglutination.

**Bacterial Ability for Colonization:**

Red rings of different intensities were noted in 37 *E. coli* isolates (61.7%) so they have colonization ability to abiotic surface while 23 isolates (38.3%) showed no red ring and considered non slime producers (fig. 8).

**Hemolysin production:**

Ten *E. coli* isolates (16.7%) showed complete hemolysis (beta-hemolysis). The remaining 50 isolates showed no hemolysis (gamma-hemolysis)

**Gelatinase test:**

All *E. coli* isolates were considered non gelatinase producers.

**Detection of Streptodornase**

Twenty eight isolates out of 42 *S. pyogenes* isolates (67%) were considered streptodornase producers (fig. 9).

**Detection of Streptokinase Activity**

Clot lysis within 30 min was observed in 23 *S. pyogenes* isolates (55%) and were considered streptokinase producers.

**Gelatinase activity of streptodornase in 14 S. pyogenes isolates. Lane D was blank DNA. Lanes from 1 to 14 were mixtures of DNA and culture supernatant of S. pyogenes isolates from No. 1 to 14 respectively. All the fourteen isolates are producers except No. 6 and 13.**
DISCUSSION

Resistance of bacteria to most antibiotics used was seen as a consequence of excessive antibiotic use[32]. The emergence of antibiotic resistant pathogenic bacteria is becoming an increasingly worrying clinical problem. It has been caused by the widespread use, and sometimes abuse of antibiotics not only in medical practice but also in animal husbandry[33]. In the present study resistance of *E. coli* was observed to most antibiotics. The greater prevalence of resistance to common antibiotics has also been reported by other workers[34]. Maximum number of isolates (100%) was resistant to ampicillin, carbenicillin and ceftazidime and the lowest to chloramphenicol (24%).

In the present study resistance of *E. coli* was observed to most antibiotics. The greater prevalence of resistance to common antibiotics has also been reported by other workers[34]. Maximum number of isolates (100%) was resistant to ampicillin, carbenicillin and ceftazidime and the lowest to chloramphenicol (24%). These results are consistent with the previous studies on drug resistance in *E. coli*[35].

World-wide, penicillins resistance has not been identified in clinical strains of *S. pyogenes*[36] and even the existence of penicillin tolerance in *S. pyogenes* seems doubtful[37]. Other β-lactam antibiotics, e.g. amoxicillin and cefaclor, have also been used successfully in the treatment of *S. pyogenes* infection since there is no resistance developed to most β-lactam[38,39].

In contrast, the present data confirms increased resistance of *S. pyogenes* to β-lactam antibiotics. Maximum number of isolates (98%) was resistant to both carbenicillin and nalidixic acid and the lowest to chloramphenicol (24%).

Qualitative detection of β-lactamase activity among *E. coli* isolates using nitrocefin solution showed that 100% of *E. coli* isolates are positive β-lactamase producers. Similar results were reported in the study of Chah and Obogbulem[21]. In contrast Qualitative detection of β-lactamase activity among *S. pyogenes* isolates using nitrocefin solution showed that only 2 isolates showed color change to red and so considered positive β-lactamase producers, this was in accordance with the previous study of Brazier et al.[40]. The gradual acquisition of mutations has recently led to a dramatic increase in the number of β-lactamase variants observed, which is well exemplified by TEM, SHV, and CTX-M enzymes[41]. In the present study, the β-lactamase gene; bla-TEM was detected and amplified on plasmid DNA of 25 β-lactam resistant *E. coli* isolates (most resistant to amoxicillin). Bla-TEM gene was detected in 23 isolates. Only isolates No. (15 and 18) seem to lack bla-TEM gene on their plasmid DNA.

Resistance to aminoglycosides through enzymatic deactivation, although seemingly straightforward, is in reality a very complex phenomenon, involving three different classes of enzyme, each with many variants. These enzymes transfer an acetyl group (derived from acetyl-Co A), an adenyl group or a phosphate group (the latter two both derived from ATP). Thus these AGIEs are broadly classified as N-acetyltransferase, O-adenylyltransferases or O-Phosphotransferases, and together they comprise over 50 different enzymes[42].

In the present study, the crude extracts prepared from 19 *E. coli* isolates and 11 *S. pyogenes* (the most resistant to aminoglycosides) were tested for aminoglycoside Acetyltransferase (AACs) using acetyl coenzyme A as a cofactor and aminoglycoside phosphotransferases (APHs) and aminoglycoside nucleotidyltransferases (ANTs) using ATP as a cofactor. The enzymes exhibited variable inactivation percentages on different aminoglycosides using different cofactors but it was clear that most AGIEs inactivated gentamicin then amikacin and finally streptomycin. This may be due to the excessive use of gentamicin in medical practice.

Chloramphenicol is a broad-spectrum antibiotic exerts its action through inhibition of microbial protein synthesis and it is effective in the treatment of several infectious diseases[43]. Microbial chloramphenicol resistance is mainly caused by drug inactivation mediated by chloramphenicol acetyltransferase (CATs) [44]. The present study revealed that 55 % produced partial or complete inactivation chloramphenicol. These results are consistent with the previous studies on chloramphenicol resistance[23].

Mechanisms of bacterial resistance to fluoroquinolones fall into 2 principal categories, alteration in drug target enzymes and alteration that limit the permeation of drug to the target[45]. In the present study, one quinolone resistance determinant genes: gyrA was amplified in DNA of some selected *E. coli* and *S. pyogenes* isolates (the most resistant isolates). GyrA gene was successfully amplified by a specific primer set in all tested *E. coli* isolates except isolates No. 15, 22 and in all the tested isolates of *S. pyogenes*.

In humans, strains of *E. coli* can be commensal (since they are part of the normal intestinal microbial flora) and/or the cause of various infectious diseases (intestinal and extra-intestinal infections)[46]. The barrier between...
commensalism and virulence results from a complex balance between the status of the host and the presence of virulence factors in the bacteria[5]. In the present study several virulence factors were studied. Serum resistance is the property by which the bacteria resist killing by normal human serum due to the lytic action of complement system[9]. In the present study 70% of E. coli isolates were resistant to serum bactericidal activity; this was in accordance with Siegfried et al.[47].

In the present study, mannose resistant haemagglutination was detected in 33% of E. coli isolates while mannose sensitive haemagglutination was detected in 26.7% of E. coli isolates and 40% of E. coli isolates produced no haemagglutination of red blood cells. These results agree to some extent with the results of Vagarali et al.[48] who reported mannose resistant haemagglutination in 25% of the isolates, mannose sensitive haemagglutination in 34.4% of the isolates and no haemagglutination in 40.6 % of the isolates.

Slime production is postulated to be a mechanism by which bacteria attach to and colonize indwelling medical devices[49]. It was noted in 37 E. coli isolates (61.7%) while 23 isolates (38.3%) were non slime producers. Haemolysin production is associated with pathogenicity of E. coli, especially the more severe forms of infection[9]. In this study, haemolysin production was detected in only 10 E. coli isolates (16.7%). These ten isolates were isolated from urine so 20% of E. coli isolates isolated from urine were considered positive hemolysin producers. In contrast, none of E. coli isolates obtained from wounds, sputum, blood, throat and feces was positive hemolysin producer. Similar results have also been reported by other workers[8].

In this study, it was observed that all hemolysin producing isolates were quinolone sensitive. This was in accordance with the study of Pitout et al.[80] as they concluded that fluoroquinolone resistant E. coli isolates typically exhibit a decreased prevalence of certain virulence factors and a seemingly reduced invasive capacity. On the other hand gelatinase production testing among all E. coli isolates reveals that none of the isolates were positive gelatinase producers; this is in accordance with the study of Sharma et al.[8] where they found that only 4 isolates out of 152 E. coli isolates were positive gelatinase producers.

Regarding S. pyogenes streptodornase is considered one of the most important virulence factors elaborated by S. pyogenes, but it was not until 2005, that its role as a virulence factor had been proved by Sumby et al.[81]. It acts as spreading factor by liquefying the viscous DNA and nucleoproteins remaining after the degradation of host cells facilitating the infiltration of the pathogen through the host tissues. In this study 66.7% of S. pyogenes isolates were considered streptodornase producers. This is consistent with the study of Kenawy[82]. Another enzyme streptokinase is produced by various strains of β-hemolytic streptococci. The enzyme is a single-chain polypeptide that exerts its fibrinolytic action indirectly by activating the circulatory plasminogen[83]. In the present study, Streptokinase activity was detected through clot lysis within 30 min. It was observed in 55% of S. pyogenes isolates.

In conclusion, the level of antibiotic resistance among hospitals has steadily increased and become a major health problem globally. Misuse of antimicrobial agents such as third generation cephalosporins and quinolones is responsible for the increased incidence and prevalence of resistance to these antimicrobials. This would have a serious impact on remaining therapeutic options. In addition bacterial pathogens elaborate a variety of virulence factors that enable them to colonize the nutrient-rich tissues, disrupt the host defense mechanisms, and/or stimulate a noxious host inflammatory response.

REFERENCES


المقاومة البكتيرية وبعض عوامل الضاوطة في كل من عيارات الإشيشيا كولات والستريتوينوكس المعزولة من مستشفيات جامعة المنصورة

 رمضان حسن، رشا برو، خان شحاته
قسم الميكروبيولوجي، كلية الصيدلة، جامعة المنصورة، جمهورية مصر العربية

يتناول البحث دراسة المقاومة البكتيرية وبعض عوامل الضاوطة في كل من الإشيشيا كولات والستريتوينوكس. في هذه الدراسة تم إجراء اختبار الحساسية لـ120 عينة من عيارات الإشيشيا كولات والستريتوينوكس في قسم الميكروبيولوجي، كلية الصيدلة، جامعة المنصورة. كما تم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها. حيث يتم استخدام محلول الببتيد ثينوكسافين وعمل التقييم الميكروبيولوجي لها.去哪里。