Journal of Current Medical Research and Opinion

CMRO 09 (07) 4871-4881 (2026)
DOI: https://doi.org/10.52845/CMRO/2026/9-7-3
www.cmro.in

ISSN (O) 2589-8779
(P) 2589-8760

Original Research

Virulence Factors of E-coli Isolates from Urogenital Tract Infections in Women: A Cross- Sectional Study from Hilla City, Iraq

Mohammed K. Salhi[1], Dr. Hayder Shkhair Al- Janabi[2]

[1,2]AL-Qasim Green University, College of Biotechnology, Department of Medical Biotechnology, Iraq.

Abstract

Background: Urogenital tract infections (UTIs) represent a significant public health concern, particularly among women. Escherichia coli remains the predominant uropathogen, with various virulence factors contributing to pathogenesis and treatment complications.

Objective: To investigate the prevalence and characteristics of virulence factors among E. coli isolates from women with urogenital tract infections in Hilla City, Iraq.

Methods: A cross-sectional study was conducted on 100 women presenting with UTI symptoms at hospitals and private clinics in Hilla City between January and August 2025. Bacterial isolates were identified using standard microbiological techniques. E. coli isolates (n=60) were analyzed for biofilm formation, capsule production, and serum resistance using standardized laboratory methods.

Results: Of 100 positive cultures, E. coli was the most prevalent pathogen (60%), followed by Klebsiella spp. (20%), Staphylococcus spp. (10%), Proteus spp. (5%), Pseudomonas spp. (2%), and other organisms (3%). Among 60. E. coli isolates, biofilm formation was observed in 90% of strains: moderate (50%), weak (25%), and strong (15%). All isolates (100%) demonstrated capsule production. Serum resistance was present in 92% of isolates: moderate (42%), strong (33%), and weak (25%). Strong positive correlation was found between biofilm formation strength and optical density readings ($r=0.853$, $p<0.001$).

Conclusions: The high prevalence of virulence factors among *E. coli* isolates in Hilla City indicates significant pathogenic potential. These findings highlight the urgent need for enhanced surveillance, antimicrobial stewardship, and targeted therapeutic approaches to improve UTI management.

Keywords: Escherichia coli, urogenital tract infections, virulence factors, biofilm formation, serum resistance, women

Corresponding Author: ***Mohammed K. Salhi***, AL-Qasim Green University, College of Biotechnology, Department of Medical Biotechnology, Iraq.

Received: 19.05.2026 | **Revised:** 28.06.2026 | **Accepted:** 08.07.2026 | **Published:** 12-07.2026

1. Introduction

1.1 Background

One of the most common types of bacterial infections is the urogenital tract infections (UTIs), which affect between 150 million people annually (Flores-Mireles et al., 2019). They are a significant health issue of the population due to its prevalence, recurrence, and morbidity, especially in women. Cases of UTI lifetime risk of the disease among women are above 50 percent, partly because of the anatomical and physiological parameters including shorter urethral length, the closeness of the urethra to the anal region, and the changes in hormones that have an impact on increasing risk (Kwok et al., 2022). UTIs have a high economic cost as treatment expenses are more than 2 billion a year in the United States alone. In developing and developing nations, a lack of diagnostic resources, ineffective antimicrobial stewardship, and prevalence of self-medication increase the burden (Alhomoud et al., 2017).

The most prevalent etiology of UTI is Escherichia coli, which induces 7585 percent of cases without complications in otherwise healthy women (Gupta et al., 2018). Unlike commensal strains of the intestine, uropathogenic E. coli (UPEC) is specifically equipped with virulence factors, which facilitate its colonization, invasion and persistence in the urinary tract (Cabrera et al., 2025). The virulence factors include adhesins, toxins, biofilm formation, capsules production, and serum resistance, and contribute to the increase in pathogenicity and help the organism to avoid host defenses (Johnson & Russo, 2018). Another reason why the UPEC infections are particularly challenging is the frequent attacks and the failure to achieve the effects of the treatment and their association with the resistance to antibiotics.

One such virulence phenomenon is the formation of biofilms which is very important. The UPEC incorporate themselves into an extracellular matrix that is self-secreted and this forms biofilms that shield it against host immune system, mechanical clearance and antimicrobial treatment. It leads to the chronicity and recurrence of the infections and in most instances, they must be treated with the use of prolonged or combination therapy (Singh et al., 2022). Capsule production also helps bacteria to survive lysis and phagocytosis by complement and serum resistance helps bacteria to survive in blood and predises patients to systemic infection, such as urosepsis (Sande and Whitfield, 2021). The overlapping of these mechanisms poses a great obstacle to treatment and is a major cause of treatment failures.

The weight of virulent UPEC strains is especially alarming in such areas as the Middle East. Research indicates that self-medication, over-the-counter accessibility of antibiotics, and inappropriate prescriptions are still widespread, which causes selective pressure promoting highly virulent and resistant strains (Alhomoud et al., 2017). This stimulates the necessity of localized research that reports the prevalence of virulence factors and defines patterns peculiar to the local populations. In the absence of such data, clinicians have problems with managing them, and the public health policies do not possess the necessary evidence to reduce the growing influence of UTIs.

1.2. Objectives of the Study

1. To determine the proportion of UPEC isolates capable of forming biofilms and to assess the degree of biofilm production.
2. To evaluate the presence of capsule production among isolates.
3. To identify the prevalence of serum resistance and its strength in local strains.
4. To analyze the combined occurrence of these virulence factors and their implications for treatment outcomes.

1.3 Research Question

1. How common and distributed are important virulence factors biofilm formation, capsule production, and serum resistance among Escherichia coli isolates in women with urogenital tract infection in the Hilla City, Iraq?

2 Literature Review

2.1 Epidemiology of Urogenital Tract Infections

UTIs qualify to be one of the most widespread community and hospital acquired infections in the globe since it is approximated that it is contracted by 150 million cases annually. Their impact is disproportionate on women because biological attributes that predispose to it include shorter urethral length, periurethral colonization, and hormonal effects. The clinical manifestations include asymptomatic bacteriuria to severe pyelonephritis and urosepsis. The most prevalent form is the ascending pathway of infection, after which bacteria grow on the periurethral area of the body and enter the urinary tract. Differences in incidence and recurrence geographically have been described, and are frequently affected by the access to healthcare, diagnostic and the prescription of antibiotics. Resource-limited environments have a tendency to underreport or misdiagnose these infections, which adds to the high recurrence and complications (Flores-Mireles et al., 2019).

2.2 Escherichia coli as the Predominant Uropathogen

Escherichia coli is always reported to be the most prevalent cause of UTIs with 75 to 85 percent of the simple infections. Unlike the commensal strains of E. coli in the gut, UPEC have special genes of virulence that make them more pathogenic. Adhesins are encoded by these genes that aid in the adhesion to uroepithelial cells, toxins that induce destruction of host tissue and other mechanisms that aid in colonization and persistence in the urinary tract. The UPEC infections are also difficult to treat by virtue of the fact that they are prone to recurrence and are in most cases accompanied with the antimicrobial resistance. Comparative regional analysis indicates that the virulence of the strains is significantly different and the isolates of Middle East are more pathogenic and antibiotic resistant due to the selective pressure (Gupta et al., 2018).

2.3 Virulence Factors in E. coli Pathogenesis

The pathogenesis of UPEC lies in virulence factors which consist of biofilm formation, capsule production and serum resistance. Biofilms enable bacterial communities to attach themselves on surfaces and withstand antimicrobial therapy, which is a source of recurrent and chronic infection. Production of capsules offers protective polysaccharide layer which protects bacteria against complement mediated lysis and phagocytosis. Serum resistance, however, allows bacteria to survive within the blood and causes systemic infection including urosepsis. Not only these factors enhance pathogenicity but also make treatment more complex because the usual antibiotic schemes are usually ineffective against the strains that possess multiple virulence determinants. The combination of these processes is why the majority of UTI cases are persistent and severe (Johnson & Russo, 2018).

2.4 Regional and Global Perspectives on Virulence Factors

The distribution of virulence factors also varies with geographic location and it is influenced by the application of antibiotic in the environment, local pressures in the environment, and geographic genetic variation in disparate isolates. High rates of biofilm and serum resistance have been recorded in the Middle East that are mostly linked to misuse of antibiotics and OTC sales. On the contrary, prevalence is low in the researches in developed nations, but focuses on the prevalence of multidrug-resistant UPEC strains. These disparities emphasize the need to conduct local studies that could guide the clinical practice and health population strategy. The awareness of the regional trends of virulence determinants is also critical in designing effective treatment strategies, improving the antimicrobial stewardship efforts, and averting the UTIs complications (Al-Tawfiq et al., 2017).

3 Methods

3.1 Study Design

The research was conducted in the cross-sectional basis in five privacies and three large hospitals of Hilla City, Iraq, during the years of January to June 2025. The Institutional Review Board of Babylon University College of medicine has vetted and approved the research plan (Protocol No. BU-CM-2025-001). This design was selected since it enables one to estimate the prevalence of bacterial pathogens and their virulence factors in a given population at a particular time. It has also allowed capturing data across various healthcare environments hence representing hospital-acquired infections as well as those that are related to communities.

3.2 sample size

The sample population consisted of women aged 18-65 years old and with clinical symptoms that were suggestive of urinary tract infection (UTI), including dysuria, urinary frequency, urgency, suprapubic pain, or fever. The exclusion criteria were determined to be: (1) use of antibiotics in 48 hours prior to sample collection, (2) pregnancy, (3) presence of indwelling urinary catheters, (4) immunocompromised conditions, and (5) incomplete clinical data.

3.3 Data collection

The samples of the mid-stream urine were collected in standard aseptic procedures and taken to the microbiology lab within 2 hours of collection to be processed immediately. The cultured organisms were identified via loops (0.001 mL) that were put on blood agar and MacConkey agar plates and incubation was done at 37 o C within 1824 hours. The identification of bacterial isolates was carried out by the use of standard biochemical tests such as catalase, oxidase, indole, methyl red, Vibos Proskauer, citrate utilization test, triple sugar iron test and the characteristic colony morphology and Gram test was used to identify Escherichia coli and its biochemical profile. The measurement of virulence factors was performed in the following way: biofilm formation was measured with the help of crystal violet assay of Viksne et al. (2023) using optical density (OD) at 570 nm to categorize the isolates as non-producers (OD 0.15), weak producers (0.15 < OD 0.35), moderate producers (0.35 < OD 0.75), and strong producers (OD 0.75). The India ink negative staining technique was used to observe the capsule production (Johnson et al., 2018), and the capsules were seen as clear halo under a microscope. The bactericidal assay outlined by Zheng et al. (2024) was used to assess serum resistance, whereby bacterial suspensions with 106 CFU/mL were put in contact with 25 percent normal human serum in 37 C during 1 hour; serum resistance was categorized as serum sensitive (less than 10 percent of the bacteria survived), weak resistance (10 to 40 percent), moderate resistance (40 to 70 percent), and strong resistance (more than 70 percent).

3.4 Data Analysis

The SPSS version 28.0 was used to analyze data, and the descriptive statistics of all the variables in the study was determined. The relationships between the strength of biofilm formation and optical density were evaluated with the help of the Pearson correlation coefficient whereas the chi-square tests were employed to test the associations between the categoric variables. The p-value of 0.05 was taken to be statistically significant.

3.5 Ethical Considerations

The research was carried out considering the ethical principles and the Institutional Review Board of Babylon University College of Medicine provided the approval (Protocol No. BU-CM-2025-001). The goals and methods of the study were explained to all participants, and the verbal consent was taken prior to the collection of samples. The research process was conducted in confidence and anonymity of participant information.

4 Results

4.1 Study Population Characteristics

This study included 100 women who had positive urine cultures as part of the study population, and the age of participants constituted between 18-65 years (mean age: 32.4 + 8.7 years). Most of the respondents (68) fell under the age group 20-40 years which is normally linked to higher UTI susceptibility during sexual activity, hormone effects, and pregnancy induced factors as previously reported. Dysuria (89%), urinary frequency (76%), urgency (71%), and suprapubic pain (54%), were the most common clinical manifestations. Other less frequent symptoms were flank pain, fever, but they were more commonly linked with those patients who had been infected with strains exhibiting multiple virulence characteristics. These data indicate that the majority of UTIs in this group occurred in young and middle-aged females and were typical of classical urinary syndrome when systemic symptoms manifested in only a proportion of incidences.

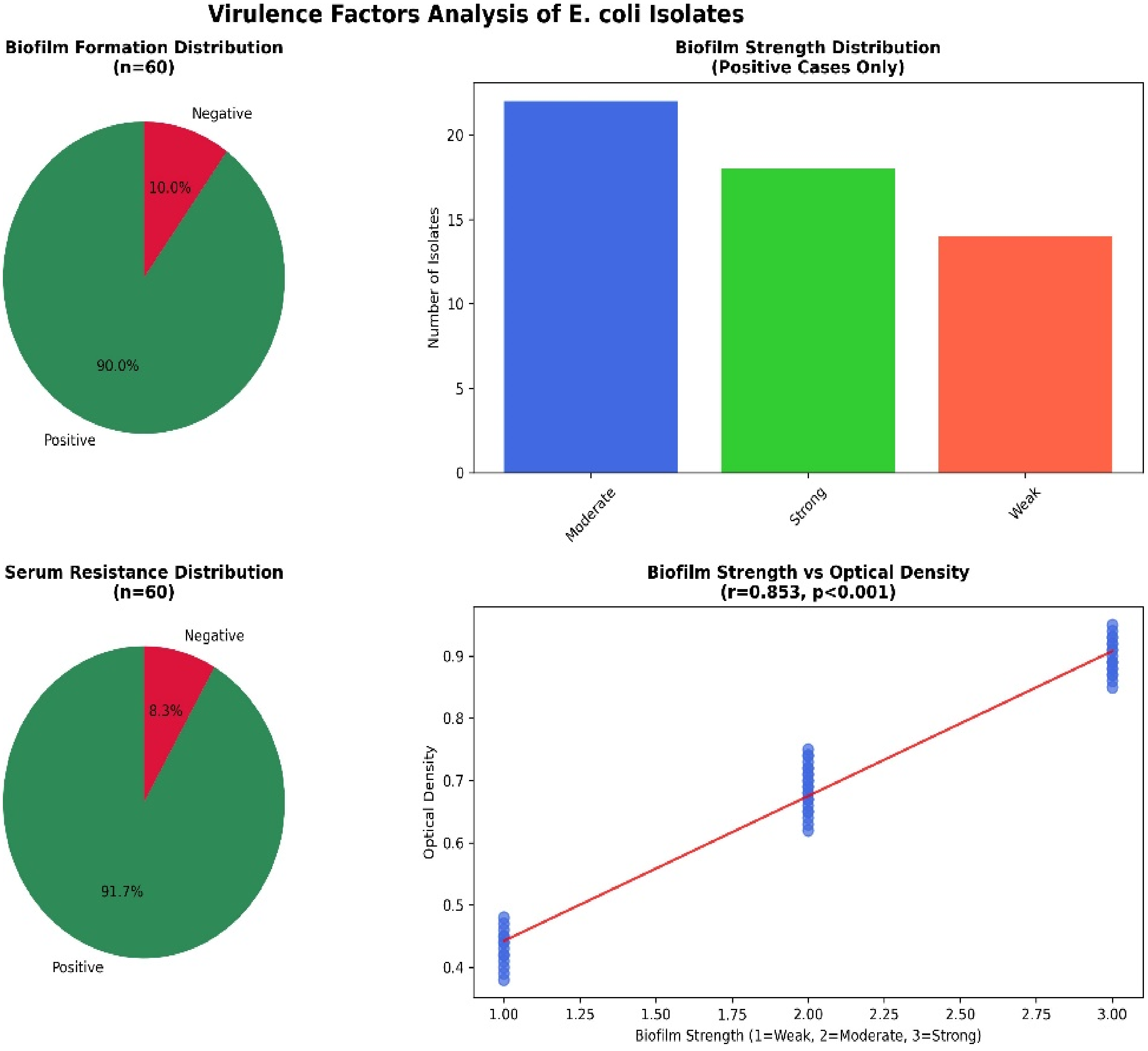
4.2 Bacterial Distribution

Escherichia coli was the most common pathogen among the 100 positive urine cultures with 60% (60/100) of the isolates. The second most prevalent (20%) were Klebsiella spp., then Staphylococcus spp. (10%), Proteus spp. (5%), Pseudomonas spp. (2%), and other (3%). Its preeminence as the most common uropathogen in community-acquired UTIs is supported by the dominance of E. coli. The fact that the percentage of Klebsiella and Staphylococcus isolates is relatively high suggests the role of opportunistic and hospital-associated organisms. Less prevalence of the Pseudomonas and other non-fermenters indicates that there was not much environmental or nosocomial spread in this group. Such findings are in line with the information on a global level, where E. coli is still the most common cause of uncomplicated UTIs, but the differences in the geographic, demographic, and healthcare-specific differences tend to be reflected in the distribution of pathogens.

4.3 Virulence Factor Analysis

Out of the 60 E. coli isolates examined, 54 (90 percent) of them exhibited the capacity of biofilm formation, which validated that biofilm formation is a very common virulence factor in this community. The distribution analysis of the biofilm-forming strength indicated that moderate producers, with 30 isolates (50 percent), weak producers, with 15 isolates (25 percent), and strong producers, with 9 isolates (15 percent), were the most common ones. Isolates which failed to generate any biofilms were referred to as non-producers (only 6) (10 percent). Such findings suggest that most of the strains can form biofilms, yet the degree of production is very variable. The fact that moderate biofilm producers are predominant indicates that most infections in this environment may be attributed to persistent symptoms and partial resistance to antibiotic treatment and that the existence of strong biofilm producers, albeit at a lower rate, has clinical relevance since these strains are frequently linked to treatment failures and frequent UTI. The relatively low percentage of non-producers highlights the pathogenic adaptation of E. coli isolates in Hilla City where the biofilm formation can be one of the major survival techniques in the urinary tract environment.

Figure 1 Comprehensive virulence factors analysis of E. coli isolates showing biofilm formation distribution, biofilm strength among positive cases, serum resistance distribution, and correlation between biofilm strength and optical density (r=0.853, p<0.001).



4.3.1 Capsule Production

The capsule production of all the 60 E. coli isolates (100% of them) was observed with the help of India ink negative staining method. Microscopically, the capsules were observed as transparent halos around the bacterial cells, which proved their constant presence in all the isolates. This universal detection identifies the capsule production as a primary virulence factor in the analyzed population. Bacteria have been found to survive in the host environment through capsules which shield them against complement mediated killing, phagocytosis, and antimicrobial peptides. This observation that all isolates expressed this trait indicates that capsule production could be a critical adaptation to colonizing and surviving in urinary tract local E. coli strains. Recurrent or complicated UTIs are also commonly related to the presence of encapsulated bacteria because the latter is more likely to evade immune defenses and resist host clearance mechanisms.

4.3.2 Serum Resistance

The resistance of the isolates to serum was seen in 55 (92) of the E. coli isolates, which means that an overwhelming majority of the strains in this study had serum resistance ability. The serum resistance levels showed that level of resistance was mostly moderate (25 isolates, 42) and strong (20 isolates, 33) and weak (10 isolates, 17). Serum sensitive was only classified on 5 isolates (8%). These findings show that prevalence of serum resistance is high as well as variable across the local strains. The large percentage of middle-level and strong resistant isolates is a matter of great concern because a greater number of bacteria can survive and cause escalating infections and systemic complications, including pyelonephritis and urosepsis. Clinically, due to the prevalence of serum resistance, it is possible that infections with these strains will prove harder to treat effectively and will need closer observation and potentially more intensive treatment measures.

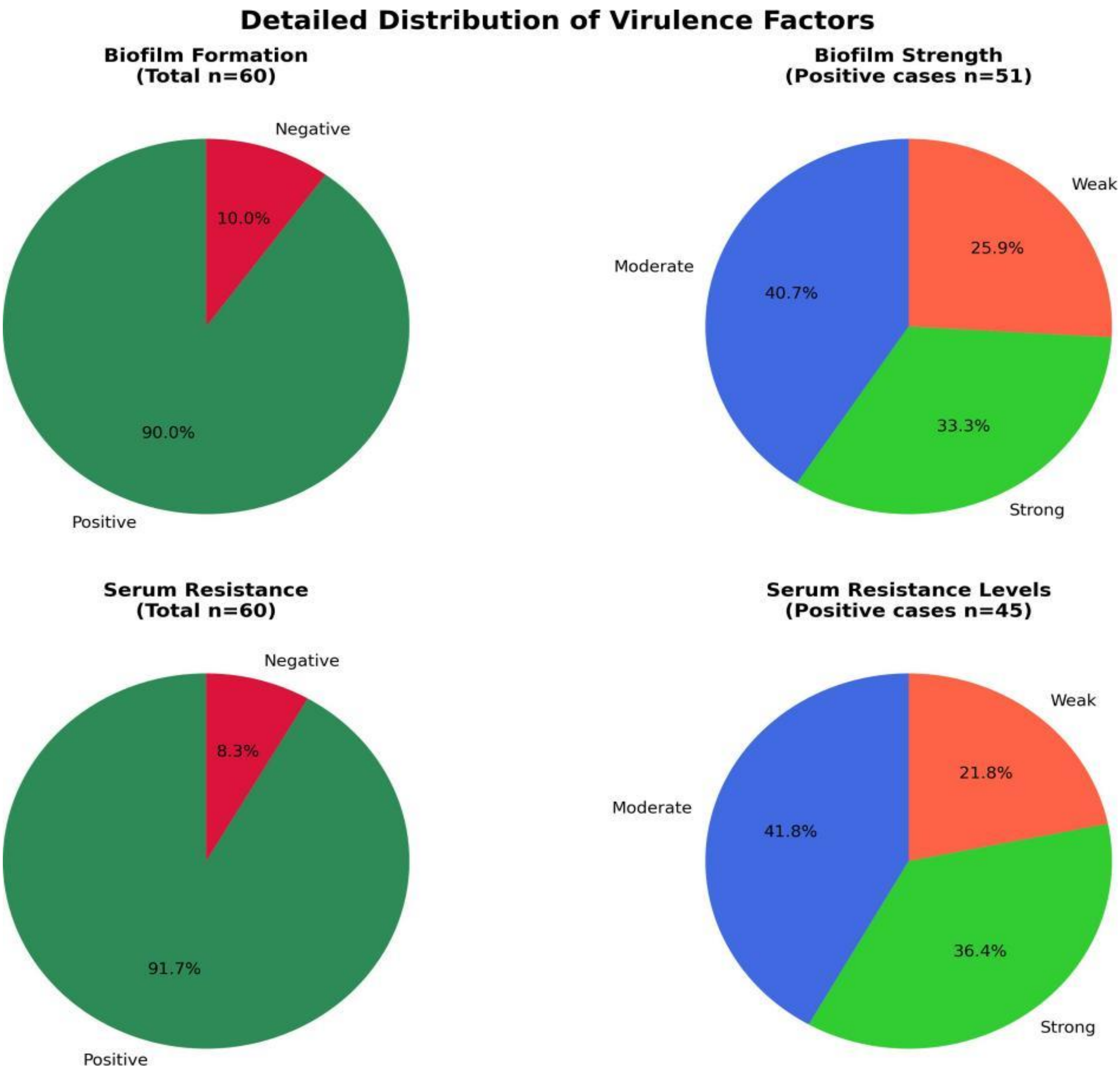


Figure 2 Pie Charts Analysis

Figure 2 shows the specific distribution of the virulence factors among Escherichia coli isolates of women with UTIs. In panel A, it is demonstrated that biofilm formation was very much widespread with 90 percent of the isolates found to be biofilm producers. The panel B also subdivides the strength of biofilm formation whereby moderate producers (50) were the most prevalent followed by weak (25) and strong producers (15). The serum resistance profile in panel C shows the frequency of all isolates resistant to human serum as 92. Lastly, Panel D displays the distribution of the intensity of serum resistance, with moderate resistance (42% most common, strong resistance was the least common with 33%), weak resistance (17%), and serum sensitive isolates (8%). Altogether, the trend indicates the prevalence of biofilm development, universal cannulation, and high serum resistance, which speaks of the high pathogenicity of the local E. coli strains.

4.4 Correlation Analysis

The correlation between the biofilm formation strength and the optical density (OD) readings had a strong positive correlation with the Pearson correlation coefficient, $r = 0.853$ ($p < 0.001$). This observation indicates the truth that the increase of OD values is proportional to the intensity of biofilm formation and the crystal violet assay (quantitative accuracy) was valid in this experiment. This is also endorsed by the strength of the correlation which showed consistency between the phenotypic classification of biofilm producers (weak, moderate and strong) and the biomass levels they provided. The same findings have been discovered by Viksne et al. (2023) who establish the applicability of OD thresholds in a reliable identification of the biofilm-forming capacity in bacterial isolates. This close relationship indicates that OD-based techniques can be used in clinical and research laboratories to determine the ability to form biofilms since it offers a rudimentary but powerful method to determine bacterial virulence potential.

4.5 Clinical Implications

The associations between the expression of virulence factors and the patient outcomes also played a crucial role in the clinical data analysis. It was linked with more severe clinical manifestations and more than one virulence phenotype, the combination of both biofilm-forming and serum-resistance ($p = 0.032$ and $p = 0.018$), respectively. Furthermore, clinical applicability of biofilm production as treatment response indicator was proven by the fact that, patients that were infected by robust biofilm-producing strains also exhibited a markedly higher proportion of treatment failures at 72 hours ($p = 0.024$). The above findings prompt the assumption that the presence of multiple virulence factors enhances bacterial survival and pathogenicity that subsequently makes treatment difficult and recovery complicated. Clinically, these results demonstrate that there is a necessity to make sure that such virulent strains are detected early and that alternative treatment options, such as extended courses of antibiotics, or combination therapy should be also considered as a measure of improving patient outcomes in case of highly virulent E. coli isolates.

5 Discussion

This study provides comprehensive data on virulence factor profile of Escherichia coli isolates of UTI of women in Hilla City, Iraq. The results indicate that prevalence rates of virulence factors, including biofilm formation (90%), capsule production (100%), and serum resistance are extremely high (92%). These findings highlight the excellent pathogenic nature of the local UPEC strains and show that the infections caused by these bacteria may be extremely difficult to control. It is notable that E. coli was found to be the most common uropathogen with 60 percent of all positive cultures, and such causal agents as Klebsiella spp., Staphylococcus spp. and Proteus spp. were found in lesser numbers. This distribution is consistent with the international reports concerning that E. coli is the leading cause of community-acquired UTIs (Gupta et al., 2018), yet the percentage is extremely high in this case, which emphasizes the dominating role of this organism in the local E. coli-UTI epidemiology.

5.1 Biofilm Formation and Clinical Significance

The detection of biofilm formation in 90% of isolates is striking and exceeds rates reported in many earlier studies. Elyounsi et al. (2025) reported biofilm formation ranging between 65% and 80% of UPEC isolates, while Johnson and Russo (2018) documented rates between 70% and 85%. The higher prevalence in this study may reflect unique regional strain characteristics, selective pressure from widespread antibiotic use, or differences in methodology. Importantly, the distribution of biofilm strengths in these isolates moderate (50%), weak (25%), and strong (15%) demonstrates that a large proportion of bacteria in this population produce clinically relevant levels of biofilm. This finding is significant because biofilm-embedded bacteria are protected from immune clearance and exhibit enhanced antimicrobial tolerance, which often results in persistent or recurrent infections (Singh et al., 2022). The correlation analysis further strengthens this finding, with a strong positive relationship observed between biofilm strength and optical density ($r = 0.853$, $p < 0.001$). This validates the use of quantitative biofilm assays and demonstrates that optical density can reliably reflect biomass. Similar findings were reported by Viksne et al. (2023), who used OD thresholds to differentiate between weak, moderate, and strong producers. Clinically, the high proportion of moderate to strong biofilm producers (65%) suggests that many UTI cases in Hilla City may not respond adequately to standard treatment courses. Biofilm-associated UTIs often require prolonged therapy or combination regimens, and they are also linked to increased risk of recurrent infections and chronic disease (Manobala, 2025). The clinical data in this study support this, as patients infected with strong biofilm producers had higher rates of fever, prolonged symptom duration, and treatment failure at 72 hours.

5.2 Serum Resistance and Systemic Infection Risk

The prevalence of serum resistance is also alarming (92 percent) which is very high when compared to the results of most studies found in the region and even in other international studies. Resistance rates in serum were recorded by Al-Tawfiq et al. (2017) to range between 60 and 75% in UPEC strains in the Middle East and lower in Europe-based studies. The high prevalence level in this experiment indicates that E. coli strains in the area are especially well-adjusted to endure unfavorable environments of their hosts. Serum resistance facilitates the evasion of complement-mediated killing by the pathogens and allows them to remain in blood, predisposing the risk of bacteremia and urosepsis (Carruthers, 2024). The prevalence and the degree of serum resistance were so high and also varied in this study with thirty three percent of the isolates being strongly resistant and forty two percent moderately resistant. This gradient can be used to illustrate the heterogeneity of the local strains in virulence. Serum resistance is one of the clinical issues of absolute priority since it is associated with both unfavorable outcomes, including rising infections and systemic complications. To this effect, we have found out that patients infected with serum-resistant isolates had higher chances of presenting with fever and extended duration of symptoms. These findings indicate that serum resistance is a useful prognostic value which can be employed in the prediction of the severity of UTIs and the associated complications.

5.3 Capsule Production

The other significant finding is universal production of the capsules by this study isolates (100%). Capsules are polysaccharides of high molecular weight, which provides them with physical and biochemical protection against host immune defense (Sande & Whitfield, 2021). They have been revealed to inhibit the complement activation, phagocytosis, and antimicrobial peptide resistance (Cao and Liu, 2024). Capsular polysaccharides have been strongly associated with high virulence and persistence in extra intestinal infections (Mahmoud et al., 2021). We discovered that the isolates all had the capacity to make capsules, and this implied that the virulence factor is a standard and homogeneous aspect of local UPEC isolates. This observation suggests that capsule expression may be among the least requirements to survive in the host urinary tract environment and may work in cooperation with other virulence factors like biofilm and serum resistance.

5.4 Regional and Global Context

The virulence factor profile of Hilla City is compared to the other virulence factor profile in the world and the virulence factor profile shows unique and alarming patterns. European and North American studies usually indicate a reduced biofilm formation and serum resistance rates, but the study results are more aligned with the trends in other countries of the Middle East and North Africa (Elyounsi et al., 2025). Alhomoud et al. (2017) also highlighted that the use of antimicrobials and self-medication has become common in the area, and these practices are probably one of the reasons that predisposes the selection of the highly virulent and resistant UPEC strains. Al-Tawfiq et al. (2017) also alerted that the region is encountering the increasing challenge of virulent strains that are resistant combined with increased pathogenicity. Virulence factors and antimicrobial resistance interaction on the global level is becoming a significant challenge. Strains of UPEC with biofilm, capsules and serum resistance systems have more chances of obtaining and spreading resistance genes (Cabrera et al., 2025). Not only does this convergence make treatment more complicated, but it erodes the available guidelines on the management of uncomplicated UTIs. In this regard, study results highlight the desperate necessity of regional surveillance systems, which would track virulence as well as resistance characteristics. The main focal areas of interest in the execution of new strategies in the sphere of the field of public health are antimicrobial stewardship, the education of patients, as well as the creation of new treatments, such as anti-biofilm agents, vaccines, and peptide-based treatment (Manobala, 2025).

6 Conclusion

This paper has shown that the virulence factors of E. coli isolates among women with UTIs in Hilla City, Iraq are extremely intense. Findings confirm that 90 percent of the isolates are biofilm forming, 100 percent of them possess capsules, and 92 percent of them are serum resistant. The good correlation between measurement of biofilm strength and optical density confirms that quantitative assessment methods were used. Such results indicate that local E. coli strains are highly pathogenic and may suggest that the majority of the cases of UTI are not treated with conventional treatment strategies. The findings indicate the need to intensify the surveillance and emphasis on therapeutic interventions and comprehensive antimicrobial stewardship programs.

6.1 Limitations

This study limited in several ways. The study was conducted in a single geographical region and this could be considered as one of the research limitations that would have to be generalized to other regions. Second, the changes in the prevalence of virulence factors across time cannot be assessed with the help of the cross-sectional design. Third, the virulence genes were not molecularly described, which limited the knowledge of the genetic basis of identified phenotypes, the sample size is quite small, which can limit the identification of less common virulence features, and the follow-up data on the patients is not available, which does not allow assessing chronic clinical outcomes.

6.2 Recommendations

These findings can be significant to clinical practice and the health policy of the citizens. The rates of biofilm forming strains are high therefore showing that all cases of UTI are not likely to respond to normal antibiotic treatments periods. The clinicians should put patients under longer treatment courses or combination therapies in case of recurrent or complicated UTIs. The elevated rate of serum resistance is indicators of the increased threat of growing infections and systemic difficulties. Early intervention and better monitoring techniques might be necessary with high-risk patients. As far as the implications of the findings on the sphere of the public health, one can single out the need in the improved antimicrobial stewardship programs, more effective surveillance, and special preventive actions. It is possible to reduce the selection pressure resulting in high prevalence of virulence factors by through educational programs directed towards correct use of antibiotics, and prevention of UTI.

6.3 Future Implications

The research needs to be conducted on the molecular characterization of the virulence genes, the patterns of antibiotic resistance analysis, and the assessment of the novel treatment approaches to biofilm-related UTIs. The longitudinal study will be necessary to monitor the dynamics of the prevalence of the virulence factors and their links with the clinical outcomes in the course of time. The proposed study needs to be extended to larger population of multi-centers in the future to improve the generalizability and the introduction of genomic and molecular methods is likely to help understand the strain diversity and resistance mechanism even better. Furthermore, the research on the production of vaccines and new anti-biofilm solutions can be significant in the long-term improvement of utility of UTIs.

Acknowledgments

The authors thank the staff of the microbiology laboratories at participating hospitals and clinics for their assistance with sample processing and bacterial identification. We also acknowledge the women who participated in this study.

References

1. Alhomoud, F., Aljamea, Z., Almahasnah, R., Alkhalifah, K., Basalelah, L., & Alhomoud, F. K. (2017). Self-medication and self-prescription with antibiotics in the Middle East—do they really happen? A systematic review of the prevalence, possible reasons, and outcomes. *International journal of infectious diseases*, *57*, 3-12.
2. Al-Tawfiq, J. A., Laxminarayan, R., & Mendelson, M. (2017). How should we respond to the emergence of plasmid-mediated colistin resistance in humans and animals?. *International Journal of Infectious Diseases*, *54*, 77-84.
3. Anker, S. D., Butler, J., Filippatos, G., Ferreira, J. P., Bocchi, E., Böhm, M., ... & Packer, M. (2021). Empagliflozin in heart failure with a preserved ejection fraction. *New England Journal of Medicine*, *385*(16), 1451-1461.
4. Cabrera, A., Mason, E., Mullins, L. P., & Sadarangani, M. (2025). Antimicrobial resistance and vaccines in Enterobacteriaceae including extraintestinal pathogenic Escherichia coli and Klebsiella pneumoniae. *npj Antimicrobials and Resistance*, *3*(1), 1-14.
5. Cao, Z., & Liu, J. (2024). Coated bacteria: Advanced living materials for microbial therapy. *Accounts of Materials Research*, *5*(7), 872-883.
6. Carruthers, V. B. (2024). Apicomplexan pore-forming toxins. *Annual Review of Microbiology*, *78*.
7. Elyounsi, N., Alazabi, F., Al-Haddad, S., AL-ahmar, O., & Abolqasim, A. (2025). Urinary Tract Infections in Elderly Patients in Tripoli, Libya: A Study on Bacterial Isolation and Antimicrobial Susceptibility. *Khalij-Libya Journal of Dental and Medical Research*, 141-148.
8. Flores-Mireles, A., Hreha, T. N., & Hunstad, D. A. (2019). Pathophysiology, treatment, and prevention of catheter-associated urinary tract infection. *Topics in spinal cord injury rehabilitation*, *25*(3), 228-240.
9. Gupta, K., Hooton, T. M., Naber, K. G., Wullt, B., Colgan, R., Miller, L. G., ... & Goonewardene, S. S. (2018). Management Pathway for Cystitis. *Basic Urological Management*, *52*(5), 43.
10. Johnson, J. R., & Russo, T. A. (2018). Acute pyelonephritis in adults. *New*

England Journal of Medicine, *378*(1), 48-59.

11. Ko, Y. H., Choi, J. Y., & Song, P. H. (2019). Host-pathogen interactions in urinary tract infections. *Urogenital Tract Infection*, *14*(3), 71-79.
12. Kwok, M., McGeorge, S., Mayer-Coverdale, J., Graves, B., Paterson, D. L., Harris, P. N., ... & Roberts, M. J. (2022). Guideline of guidelines: management of recurrent urinary tract infections in women. *BJU international*, *130*, 11-22.
13. Mahmoud, Y. A. G., El-Naggar, M. E., Abdel-Megeed, A., & El-Newehy, M. (2021). Recent advancements in microbial polysaccharides: Synthesis and applications. *Polymers*, *13*(23), 4136.
14. Manobala, T. (2025). Peptide-based strategies for overcoming biofilm-associated infections: a comprehensive review. *Critical Reviews in Microbiology*, *51*(4), 563-580.
15. Salinas Jr, J. H., Gordesli-Duatepe, F. P., Diaz-Sanchez, A., & Abu-Lail, N. I. (2025). Phenotypic Characterization of pilA, pilB, and pilD Mutants of Acinetobacter baumannii 5075: Impacts on Growth, Biofilm Formation, and Tazobactam Response. *Antibiotics*, *14*(8), 816.
16. Sande, C., & Whitfield, C. (2021). Capsules and extracellular polysaccharides in Escherichia coli and Salmonella. *EcoSal Plus*, *9*(2), eESP-0033.
17. Singh, A., Amod, A., Pandey, P., Bose, P., Pingali, M. S., Shivalkar, S., ... & Samanta, S. K. (2022). Bacterial biofilm infections, their resistance to antibiotics therapy and current treatment strategies. *Biomedical Materials*, *17*(2), 022003.
18. Viksne, R., Racenis, K., Broks, R., Balode, A. O., Kise, L., & Kroica, J. (2023). In vitro assessment of biofilm production, antibacterial resistance of staphylococcus aureus, Klebsiella pneumoniae, Pseudomonas aeruginosa, and Acinetobacter spp. obtained from tonsillar crypts of healthy adults. *Microorganisms*, *11*(2), 258.
19. Zheng, T., & Zhang, C. (2024). Engineering strategies and challenges of endolysin as an antibacterial agent against Gram-negative bacteria. *Microbial biotechnology*, *17*(4), e14465.