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Evaluation of Procalcitonin and Interleukin-6 as a Marker of Bacterial Urinary Tract Infection

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ABSTRACT

Urinary tract infection (UTI) is a major clinical problem in a wide age range that is associated with a high morbidity rate. Due to issues such as low specificity and the inability to differentiate between different types of infection in current diagnostic methods, there is a need to introduce novel UTI markers. The present study was conducted to evaluate the utility of Procalcitonin (PCT) and interleukin-6 (IL-6) as a marker of bacterial urinary tract infection. For this purpose, a cross-sectional study was conducted between November 2020 and February 2021 among 125 patients and 60 healthy volunteers (control) in Erbil Teaching Hospital. The concentration of PCT and IL-6 was quantified using the ELISA cloud immunoassay test. Between-group comparisons were assessed for the variables with analysis of variance. The results revealed that there was a significant difference between PCT levels in UTI patients (104.6±6.07) and control groups (54±2.24) ($p < 0.0001$). The differences in IL-6 concentration in UTI patients (55.74±4.2) and control groups (24.56±2.4) were also significant ($p < 0.0001$), implying that the level of both PCT and IL-6 increased due to bacterial infection in the urinary tract. As a whole, the findings of this study provide evidence supporting that PCT and IL-6 can be used as a marker of UTI in both children and adults.

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Introduction

Urinary tract infection (UTI) represents a major microbial disease that, in addition to clinical challenges, imposes a heavy financial burden on society. It is also one of the most common causes of hospitalizations for infections among elderly people and the most common indication for antibiotic prescriptions in primary care. Diagnosis and management of UTI is a challenging issue in clinical practice due to the high prevalence and recurrence of the disease, and the worldwide increase of antibiotic resistance. The clinical symptoms of UTI are often uncharacteristic or asymptomatic (1). Escherichia coli is the main cause of UTI, which binds to epithelial cells and urinary mucosal tissue in humans with its adhesion ability endowed by both fimbriae and non-fimbriae agents, and causes the symptoms of the disease. Other bacterial species that cause UTIs include *Enterococcus* species (spp.), *Enterobacter* spp., *Pseudomonas aeruginosa*, *Kelbsiella pneumoniae*, *Proteus mirabilis*, and *Staphylococcus* spp. (2).

Due to the high prevalence of UTI and the global increase in antibiotic resistance, the diagnosis and treatment of urinary tract infections has become a major clinical challenge. On the other hand, the clinical symptoms of UTI are not well characterized. Considering the long-term negative consequences as well as the risk of septicemia, early diagnosis and treatment of UTI represent a critical issue in clinical practices. Although UTI occurrence can be determined through laboratory tests, accurate diagnosis of this infection requires careful consideration of the patient's symptoms along with test results (3).

It is crucial to understand the pathogenesis of UTIs and the host defense mechanisms against such an infection. The inflammatory response of the host involves the release of a set of proinflammatory cytokines that enhance the inflammatory process (4). Inflammation is the immune system's response to

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harmful stimuli, such as pathogens, damaged cells, toxic compounds, or irradiation. Inflammation is therefore a defense mechanism that plays a vital role in disease treatment (5). Usually, during acute inflammatory responses, cellular and molecular events and interactions efficiently minimize impending injury or infection. Since urinary tract infection is caused by microorganisms invading the urethra and bladder, this infection causes inflammatory responses (6). One of the most important components of the inflammatory response is the secretion of interleukin 6 (IL-6), which is secreted by urothelial cells upon exposure to UTI-causing agents; therefore, it can be used as a biomarker for early UTI diagnosis (7).

IL-6 is a multifunctional cytokine with pro-inflammatory and immune regulatory functions such as the acute phase response, inflammation and organ development (8,9). This spectrum of biological activities fits well with the findings in patients with systemic UTI, who have elevated temperature and circulating acute phase reactants such as C-reactive protein (CRP). Based on this, IL-6 from the infected urinary tract may be a key mediator of these responses (10,11).

Considering E. coli as the major cause of UTI, and given the role of IL-6 as part of the response to E. coli infection, an inverse relationship can be inferred between the severity of Uropathogenic E. coli (UPEC) infection and IL-6 secretion (12). This proposition is supported by the studies in children (4,13) and model animals (14,15) which show that IL-6 production is positively correlated with UTI. This empirical evidence and the immune mechanism of exposure of the body to E. coli infection and other UTI-causing factors indicate IL-6 involvement in UTI; therefore, it is postulated that IL-6 can be used as a marker to detect UTI.

Procalcitonin (PCT), a sepsis-related protein, was first found to be elevated in patients with sepsis in 1993 (16). Today, this peptide precursor is widely used as a biomarker for bacterial infections. Activated adherent macrocytes and various tissues produce PCT during the inflammation process, especially during the response to bacterial infection (17,18). In contrast to bacterial infections, viral infections such as viral pneumonia usually induce modest PCT production because interferon- inhibits adipocytes from producing PCT (19). PCT levels in normal individuals are very low (less than 0.1 ng/ml), which can be elevated as slightly as 1.5 ng/ml in inflammatory conditions and viral infections; however, in bacterial infections, PCT levels increase sharply to as high as 100 ng/ml (20). PCT is a precursor of the hormone calcitonin, which is secreted by C-cells from the thyroid gland, and during severe bacterial infection, it is produced by the monocyte-macrophage system (21). Procalcitonin is the most reliable index in the diagnosis and separation of bacterial and viral infections in patients admitted to hospitals for acute reasons such as burns, inflammation, surgery, respiratory problems, and neonatal infections (22). Experimental studies also support this theorem. For example, Xu et al. (2014) reported that PCT has remarkable sensitivity and specificity in the early diagnosis of UTI (23). Similarly, Prat et al. (24) also found superiority of PCT over C-reactive protein in the prediction of UTI.

Due to the high prevalence of UTI, which in addition to clinical problems, causes antibiotic resistance, and considering the low specificity of some diagnostic methods, the present study was conducted to evaluate the effectiveness of Procalcitonin IL-6, and CRP as a marker of bacterial urinary tract infection. Given the urgent need for new and reliable markers for UTI diagnosis, the present study could have potentially useful clinical implications. In addition, the results of this study pave the road for further research into the effectiveness of PCT and IL-6 as a marker of UTI diagnosis.

Material and methods

Study subjects

A cross-sectional study was conducted between November 2020 and February 2021 and the samples were collected from Erbil Teaching Hospital. 125 patients (18-75 years old) were selected who were clinically suspected of having UTI, signing informed consent, not being diagnosed with any disease, not taking antibiotics within <24 hours prior to collection of specimens. Sixty healthy volunteers based on the results of a medical examination by an internal medicine physician, with negative urine culture results, were included as a control group. Participants were asked to sign a consent form provided in the questionnaire and approval was obtained from the ethics committee of the Department of Biology,

College of Education, Salahuddin University-Erbil, Kurdistan Region, Iraq.

Urine culture

Midstream urine sample was taken aseptically into a sterile container and homogenized urine specimens were inoculated on Nutrient agar medium and MacConkey agar medium. The culture media was then incubated for 24 hours at 37°C. The number of bacterial colonies grown was counted with colony counter and the results were multiplied by 102. Specimens without bacterial growth or specimens with bacterial growth ≤105 CFU/mL were considered negative, while those with >105 CFU/mL were considered positive (25). Identification of Gram-negative and Gram-positive bacteria was performed using VITEX 2 automated microbiology system analyzer.

Blood samples

Venous blood samples were collected aseptically from UTI patients and control group using sterile disposable syringes (about 5 ml). Blood samples were divided into two parts, 1-2ml of blood was transferred into anticoagulant ethylene diamine tetraacetic acid (EDTA) containing tubes for estimation of white blood cells (WBCs) and absolute leukocyte counts, and 3ml of blood was poured into plain universal tubes allowed to clot and then centrifuged at 2500-3000 rpm for 15 min. to separate the serum and dispensed into sterile Eppendorf tubes, stored at –20°C and used for the following tests. The serum used for the determination of IL-6 was assessed with Elisa Cloud Immunoassay with sensitivity levels of 7.81 pg/ml, respectively, following the manufacturer's instructions. The serum concentration of Procalcitonin was determined with a commercially available Elisa Cloud Immunoassay test provided by Cloud-Clone Corp. The test sensitivity was 31.2 pg/ml for Procalcitonin, and serum IL-6 was quantified by an automated Cobas E411 analyser.

Statistical Analysis

All statistical analyses were performed using SPSS 19.0 (SPSS Inc., Chicago, IL, USA) statistical package. Normally distributed variables were expressed as mean ± SD as appropriate. A p-value<0.05 was considered to be statistically significant. Between-group comparisons were assessed for categorical variables with the (ANOVA) test.

Results and discussion

First, the number of Gram-negative and Gram-positive bacteria in the test subjects was investigated. As shown in Table 1, the number of gram-negative bacteria was higher in women; whereas Gram-positive bacteria were predominant in male subjects.

Next, the bacterial species in the samples were examined (Table 2). As can be seen, *Enterococcus faecalis*, *Escherichia coli* and *Staphylococcus aureus* were the most common bacterial species in the samples, respectively. The presence of two Gram-positive bacteria among the three dominant bacterial species in the samples was a notable observation in this phase.

Then, the antibiotic sensitivity pattern of both Gram-positive and Gram-negative bacteria in UTI patients was studied. Among gram-positive species, *S. aureus, S. epidermis, E. faecalis* and *S. agalacticae* showed full resistance against Cefoxitin, Benzylpencillin, Rifampicin and Oxacillin. Full resistance against Gentamicin was also observed for *S. aureus, E. faecalis* and *S. agalacticae*. In general, Teicoplanin, Vancomycin, Tetracycline and Tigecycline showed high inhibitory impact against gram-positive bacteria. Regarding gram-negative bacteria, *E. coli, K. pneumoniae* and *S. paucimobilis* exhibited full resistance against Ampicillin/sulbactam, Pipercellin/tazobactam and Cefazolin antibiotics. *P. aeruginosa* showed full resistance against Ampicillin/sulbactam, Cefazolin, Ceftazidime, Cefipime, Ertapenem, Imipenem and Trimethoprim/sulfamethoxazole. High resistance of *S. paucimobilis* was observed in this test; that is the species was fully resistant against Ampicillin/sulbactam, Pipercellin/tazobactam, Cefazolin, Ceftazidime, Ceftriaxone, Cefipime and Aztreonam. Full descriptions of bacterial resistance to antibiotics are presented in Tables 3 and 4.

Table 1. Distribution of bacterial growth in UTI patients according to gender

Gender	Number	Growth status		
		No growth	Growth	
			G+ bacteria	G-bacteria
Male	61	31	21	14
Female	64	23	16	20
Total number	125	54	37	34

Table 2. Frequency (no.) of isolated bacteria from positive urine samples according to gender

Bacteria group	**Bacterial species**	**No.**	**Gender**			
			Male		**Female**	
			n.	**%**	**n.**	**%**
Gram+ bacteria	*Staphylococcus aureus*	13	9	12.50	4	5.56
	Staphylococcus epidermis	2	2	2.78	0	0.00
	Staphylococcus lentus	1	1	1.39	0	0.00
	Enterococcus faecalis	15	4	5.56	11	15.28
	Micrococcus lentus	2	1	1.39	1	1.39
	Streptococcus agalacticae	4	4	5.56	0	0.00
Gram- bacteria	*Pseudomonas aeruginosa*	8	4	5.56	4	5.56
	Escherichia coli	14	6	8.33	8	11.11
	Klebsiella pneumoniae	7	2	2.78	5	6.94
	Enterobacter aerogenes	2	2	2.78	0	0.00
	Serratia fonticola	1	0	0.00	1	1.39
	Burkholderia cepacia	1	0	0.00	1	1.39
	Sphingomonas paucimobilbis	1	0	0.00	1	1.39
	Total	71	35	49.296	36	50.704

Following serological studies, the concentrations of urea, creatine and WBC in UTI and control blood samples were compared, the results of which are presented in Tables 5 and 6. As expected, white blood cell count, creatine and urea levels in UTI samples were significantly higher than those of the control group. Urea concentration in the UTI group was 171.9±5 mg.dl-1 compared to 37.8 ± 2.73 mg.dl-1 in the control group, which shows a significant difference ($p<0.0001$). Creatine levels in the UTI group were more than eight times higher than that of the control group (Table 6). The core phase of this study was to evaluate the levels of PCT and IL-6 in UTI blood samples and compare them with the control group to determine whether the levels of these factors change due to urinary tract infection. The results of this test are presented in Table 7. The values of PCT, IL-6 and CRP in the UTI group were 104.6±6.07, 55.74±4.2, and 4.43±1.05; respectively; while the

counterpart values in the control group were 54±2.24, 24.56±2.4, and 0.17±0.05.

Table 3. Antibiotic sensitivity pattern of Gram-positive bacteria in UTI patients

No.	Antimicrobial	Number (%) of Resistance susceptibility of Gram-positive bacteria					
		S. aureus (13)	*S. epidermis* (2)	*S. lentus* (1)	*E. faecalis* (15)	*M. lentus* (2)	*S. agalacticae* (4)
1	Cefoxitin	13 (100)	2 (100)	-	15 (100)	2(100)	4 (100)
2	Benzylpencillin	13 (100)	2 (100)	1(100)	15 (100)	0(0)	4 (100)
3	Oxacillin	13 (100)	2 (100)	1(100)	15 (100)	0(0)	4 (100)
4	Gentamicin	13 (100)	1 (50)	1(100)	13 (86.66)	0(0)	4 (100)
5	Tobramycin	0 (0)	1 (50)	1(100)	6 (40)	0(0)	4 (100)
6	Levofloxacin	13 (100)	2 (100)	0(0)	9 (60)	1(50)	2 (50)
7	Moxifloxacin	13 (100)	2 (100)	-	15 (100)	0(0)	2 (50)
8	Inducible clindamycin resistance	0 (0)	0 (0)	-	12 (80)	0(0)	2 (50)
9	Erythromycin	0 (0)	0 (0)	0(0)	0 (0)	0(0)	0 (0)
10	Clindamycin	13 (100)	0 (0)	-	0 (0)	0(0)	2 (50)
11	Linezolid	0 (0)	2 (100)	1(100)	0 (0)	0(0)	2 (50)
12	Teicoplanin	0 (0)	0 (0)	1(100)	0 (0)	1(50)	0 (0)
13	Vancomycin	0 (0)	0 (0)	0(0)	15 (100)	0(0)	0 (0)
14	Tetracycline	0 (0)	0 (0)	-	0 (0)	2(100)	0 (0)
15	Tigecycline	0 (0)	0 (0)	-	0 (0)	0(0)	0 (0)
16	Fosfomycin	13 (100)	1 (50)	-	6 (40)	0(0)	0 (0)
17	Nitrofurantoin	0 (0)	0 (0)	1(100)	15 (100)	0(0)	0 (0)
18	Fusidic acid	13 (100)	0 (0)	1(100)	15 (100)	0(0)	0 (0)
19	Mupirocin	13 (100)	1 (50)	-	6 (40)	2(100)	4 (100)
20	Rifampicin	13 (100)	2 (100)	1(100)	15 (100)	0(0)	4 (100)
21	Trimethoprim/sulfamethoxazole	0 (0)	0 (0)	1(100)	15 (100)	0(0)	4 (100)

Table 4. Antibiotic sensitivity pattern of Gram-negative bacteria in UTI patients

No.	Antimicrobial	Number (%) of Resistance susceptibility of Gram-negative bacteria					
		P. aeruginosa (8)	*E. coli* (14)	*K. pneumoniae* (7)	*S. paucimobilis* (1)	*B. cepacia* (1)	*S. fonticola* (2)
1	Ampicillin/sulbactam	8 (100)	14 (100)	7 (100)	1 (100)	0(0)	0(0)
2	Pipercellin/tazobactam	2 (25)	14 (100)	7 (100)	1 (100)	0(0)	0(0)
3	Cefazolin	8 (100)	14 (100)	7 (100)	1 (100)	1(100)	1(50)
4	Ceftazidime	8 (100)	3 (21.42)	3 (42.85)	1 (100)	1(100)	1(50)
5	Ceftriaxone	2 (25)	14 (100)	3 (42.85)	1 (100)	1(100)	1(50)
6	Cefipime	8 (100)	12 (85.71)	0 (0)	1 (100)	0(0)	1(50)
7	Aztreonam	2 (25)	14 (100)	3 (42.85)	1 (100)	0(0)	2(100)
8	Ertapenem	8 (100)	0 (0)	7 (100)	0 (0)	0(0)	0(0)
9	Imipenem	8 (100)	0 (0)	3 (42.85)	0 (0)	0(0)	2(100)
10	Meropenem	4 (50)	0 (0)	0 (0)	0 (0)	0(0)	2(100)
11	Amikacin	0 (0)	0 (0)	0 (0)	0 (0)	0(0)	1(50)
12	Gentamicin	0 (0)	6 (42.85)	7 (100)	0 (0)	0(0)	1(50)
13	Tobramycin	0 (0)	0 (0)	0 (0)	0 (0)	1(100)	2(100)
14	Ciprofloxacin	0 (0)	6 (42.85)	7 (100)	0 (0)	0(0)	2(100)
15	Levofloxacin	8 (100)	0 (0)	0 (0)	0 (0)	1(100)	0(0)
16	Tigecycline	4 (50)	0 (0)	0 (0)	0 (0)	0(0)	0(0)
17	Trimethoprim/sulfamethoxazole	8 (100)	3 (21.42)	3 (42.85)	1 (100)	1(100)	2(100)

Table 5. Total WBC counts in UTI patients and control

Parameters	UTI	Control	*p*-value
White blood cells	9.29±0.3	7.38±0.15	0.95
Lymphocytes	1.36±0.06	1.51±0.07	0.99
Monocytes	0.87±0.08	2.27±0.61	0.95
Granulocytes	7.05±0.26	4.34±0.13	0.90

Table 6. Urea and Creatine serum level in UTI and control

Parameters	UTI	Control	*p*-value
Urea mg/dl	171.9±5 mg/dl	37.08±2.73	<0.0001
Creatine mg/dl	6.87±0.24	0.84±0.04	0.009

Table 7. Procalcitonin, IL-6 and CRP serum level in UTI and control

Parameters	**UTI**	**Control**	***p*-value**
Procalcitonin	104.6±6.07	54±2.24	<0.0001
IL-6	55.74±4.2	24.56±2.4	<0.0001
CRP	4.43±1.05	0.17±0.05	0.0002

Table 8. Procalcitonin and IL-6 serum level gram-positive and gram-negative bacterial infection in UTI patients.

Parameters	G+VE	G-VE	*p*-value
Procalcitonin	109.2±8.05	78.46±4.64	0.01
IL-6	77.05±6.1	35.6±3.96	<0.0001

As seen, a significant difference was observed between PCT concentration in UTI and control groups ($p < 0.0001$). The observed differences in IL-6 concentration in UTI and control groups were also significant ($p < 0.0001$), implying that the secretion of both PCT and IL-6 increased due to bacterial infection in the urinary tract. Moreover, increased concentrations of PCT and IL-6 as a result of Gram-positive and Gram-negative bacterial infections were compared (Table 8). According to the Table, the concentrations of PCT and IL-6 due to infection with Gram-positive bacteria are significantly higher than the infection caused by Gram-negative bacteria. Regarding PCT, the concentration of this factor increased from 78.46 ± 4.64 in Gram-negative bacterial infection to 109.2±8.05 which was statistically significant ($p < 0.01$). Regarding IL-6, the level of t this factor increased from 35.6 ± 3.96 in Gram-negative infection to 77.05 ± 6.1 in Gram-positive bacterial infection, which is also significant ($p < 0.0001$).

Counting the number of white blood cells is one of the current methods of diagnosing UTI. Since the aim of this study was to evaluate the efficacy of Procalcitonin and IL-6 as diagnostic markers of UTI, in the last phase, the correlation between WBC and the two proposed markers (PCT and IL-6) was investigated. The results of this statistical analysis revealed that there is a positive correlation between WBC and PCT as well as between WBC and IL-6.

This study aimed to investigate the possibility of using PCT and IL-6 as markers for the early diagnosis of UTI. Results showed that UTI infection samples gave about 71 positive bacterial cultures and about 54 samples registered as negative bacterial cultures. The lack of growth in negative samples of bacterial culture may be attributed to the fact that the infection causes may be either viral or anaerobic bacteria that cannot be isolated by the usual culture method used in this study. Alternatively, this may be because of antibiotic administration in patients or even inappropriate use of the antibiotics that cause the disappearance of bacteria that cause urinary tract infection. In this regard, first, its frequency between males and females and the type of bacterial species and gram typology were investigated. In terms of gender distribution, it was found that the frequency of gram-negative and gram-positive bacteria is higher among women and men, respectively. Furthermore, the number of gram-positive bacteria was more than gram-negative species. Regarding the type of causative pathogens, it was revealed that both gram-positive and gram-negative bacteria are involved in the development of UTI. While in most studies *E. coli* has been identified as the most common causative agent of UTI infection (26–28), in the present study *Enterococcus faecalis* was the dominant species followed by *E. coli* and *S. aureus*. This finding indicates that in addition to *E. coli*, the potential involvement of other bacterial species should also be considered as causative agents of UTI; especially *E. faecalis* which is an opportunistic pathogen that with several adhesion factors is also involved in various infections including urinary tract infections. Other authors have also

mentioned the involvement of *E. faecalis* in UTI (29,30).

The main focus of the present study was to investigate the possibility of using PCT and IL-6 as markers for the early diagnosis of UTI. Our main motivation for conducting this research is the problems and shortcomings of current methods of UTI diagnosing, which is due to the complex nature of the disease. On one hand, determining the severity and extent of UTIs is essential to determining subsequent management of the disease; because depending on the type of infection (whether lower UTI or pyelonephritis is the case) both the duration of treatment and the type of prescription antibiotics vary (31). On the other hand, failure in early diagnosis and treatment of UTI leads to further consequences such as sepsis and renal abscess; this highlights the importance of early diagnosis of UTI in the prevention of subsequent complications (21). The clinical signs of UTI alone are not sufficient to diagnose the infection. Therefore, finding effective markers for early diagnosis has become one of the important areas of UTI research. Factors such as white blood count, erythrocyte sedimentation rate and C-reactive protein have been reported as potent markers of UTI. These indicators, despite their advantages, suffer from some limitations such as low specificity. So the quest to find more accurate and reliable markers continues.

Urinary tract cytokine responses are triggered when bacteria reach the mucosal surface. Attachment to epithelial cells activates a first cytokine cascade that includes IL-6, IL-1, IL8, and other chemokines (32,33). The magnitude and repertoire of cytokines is influenced by the virulence of the infecting bacteria, including the fimbriae. Epithelial cell activation is followed by the recruitment of neutrophils and other inflammatory cells to the local site by the second round of cytokine responses (34).

Recent original studies and meta-analyses highlighted the effectiveness of PCT protocols in the early diagnosis of bacterial infection and further in assisting in the initiation and termination of antibiotic treatment (35,36). Levin *et al.* (20) examined the correlation between serum PCT and UTI and reported that PCT level <0.25ng / ml excludes the presence of UTI and thus helps to reduce antibiotic use in the suspected patients. In another study, Luo *et al.* (37) identified the procalcitonin/albumin ratio as a marker for early diagnosis of febrile urinary tract infection. Gervaix *et al.* (38) also reported the utility of procalcitonin-based diagnosis for the management of children with urinary tract infections. The authors maintained that PCT is a stronger predictor than CRP . The results obtained in the present study connote that PCT is a potentially useful marker for the rapid and timely diagnosis of UTI, which gives physicians better decision-making power when dealing with urinary tract infections. According to these findings, it can be concluded that the use of PCT not only improves the treatment of UTI patients but also reduces the use of antibiotics and thus (in addition to reducing the cost of antibiotic use) plays an important role in counteracting antibiotic resistance.

Recent studies suggested that a highly elevated blood PCT level is associated with Gram-negative infection. In healthy volunteers, PCT was found to increase within 4 hours after the injection of endotoxin, a specific pathogenic factor of Gram-negative bacteria, and fall rapidly during recovery. This feature makes PCT an ideal candidate for early identification of GNBSI with further potential in guiding antibiotic treatment (39,40). Different pathogens are believed to induce various levels of PCT as they activate different Toll-like receptor signaling pathways. In healthy individuals, PCT found in the circulation would be ≤0.1 ng/mL (41, 42). Normal or slightly elevated PCT level in bacterial infection and septic patients was more likely to be a result of viral infection or systemic inflammatory response of noninfectious origin rather than bacteremia (including both Gram-negative and Gram-positive infection) or fungemia (43,44). In a meta-analysis, the mean concentration of PCT was found to be around 6 ng/mL in patients with Gram-positive and/or fungal infections, which is significantly higher than in healthy controls. In Gram-negative infections, the PCT level was found to be even higher with its value being around 13 ng/mL, denoting that the level of induced PCT concentration varies among pathogens even in bacteremia (45).

IL-6 was another factor examined in the present study as a potential UTI marker. Based on the results, the level of IL-6 in patients with UTI (55.74 ± 4.2) was significantly higher than its value in the control group (24.56 ± 2.4) ($p < 0.0001$). Interleukins are part of the urinary cytokine response to bacterial

infections. In response to a urinary tract infection, the urothelium rapidly secretes chemokines and cytokines, making interleukin concentrations much higher in UTI patients than in healthy individuals (46). IL-6 is a multifunctional cytokine that plays a key role in activating an acute-phase response. The wide extent of IL-6 bioactivity is consistent with the immune symptoms of patients with systemic UTIs (47). Given the role of IL-6 in the treatment of bacterial infections, its elevated value in the serum of UTI patients can be justified. In fact, many studies support the usefulness of IL-6 as a marker in the diagnosis of various types of UTI. In a study conducted among 81 patients with febrile UTI, Otto *et al.* (48) showed that IL-6 secreted by the urinary tract triggers the host systemic response to bacterial infection. The findings of Jantausch *et al*. (49) show that IL-6 and IL-8 are part of the host defense system whose concentrations increase significantly in children with UTI; therefore, they are a strong predictor of UTI. Another study found that IL-6 levels in the serum and urine of children with acute pyelonephritis were significantly higher than lower UTIs; which highlights the discrimination ability of IL-6 in different types of urinary tract infections (50). Otukesh *et al.* (51) also concluded that urinary and serum concentrations of IL-6 have a high diagnostic value in differentiating pyelonephritis from cystitis and can be an important indicator in the early diagnosis of upper and lower urinary tract infections in the acute phase of the disease in children. Ching *et al.* (52) reported that Interleukin-6/Stat3 signaling plays a key role in the host's immune response during bacterial urinary tract infection; implying that disruption of this signaling pathway increases the accumulation of bacteria. In general, the literature supports our findings on the utility of factor IL-6 as a marker for UTI diagnosis.

CRP levels increase in the first hours of tissue damage or at the onset of a bacterial infection; therefore, it can potentially be used as a predictor of UTI. In healthy people, the CRP level is very low (less than 6 mg / l); however, due to infection, its value increases significantly (53). Various studies show that in UTI patients, CRP values increase significantly compared to normal individuals. Changes in CRP levels are most often seen in UTIs caused by *Escherichia coli*, *Proteus* spp., *Klebsiella pneumoniae*, and *Staphylococcus aureus* (54). In addition, CRP levels are used as a marker for treatment progression (55). However, it should be noted that increased CRP levels are not unique to UTI and elevated CRP is one of the predictors of inflammation in patients with various infections such as neonatal sepsis, fungal infections and pelvic inflammatory diseases (53).

Finally, it should be noted that our findings were obtained in a sample with a very wide age range (18-75 years old) that improves the generalizability and reliability of the results. Given that much previous research has been done on UTI biomarkers in children, our results could add to the existing literature on the utility of PCT and IL-6 as UTI markers. For example, Nanda *et al.* (21) stated that due to the current focus of UTI research on pediatric groups, it is better to do further studies on UTI markers in age classes other than the pediatric age group.

Conclusions

Urinary tract infection is a major clinical problem in various age groups with high levels of morbidity. Drawing upon previous studies highlighting the increased level of PCT and IL-6 during UTI, the present study was carried out to investigate the utility of these factors as a reliable marker for UTI diagnosis. The results of this study showed that PCT and IL-6 values in patients with UTI are significantly higher than those in healthy individuals; suggesting that PCT and IL-6 could potentially be used as diagnostic markers of UTI. Considering the body's defense mechanism against bacterial infection and its underlying mechanisms well supports this finding; because PCT and IL-6 play an effective role in the host's immune response to bacterial infection. Evidence from various clinical studies also confirms the usefulness of PCT and IL-6 as a marker of UTI. Considering the clinical significance of UTI and the shortcomings in the diagnosis of this infection, the findings of the present study can have important implications for clinical settings. Current diagnostic methods typically rely on urine culture and analysis, which is a costly and time-consuming method; while using PCT and IL-6 as a marker not only speeds up the detection process, it can also save costs. On the other hand, while many previous studies have been

performed in children, this study has examined a wide range of age groups, which can increase the reliability of our findings. In other words, our results show that PCT and IL-6 have the predictive power of UTI in a wide range of age classes. Overall, our findings contribute to the existing literature by providing further evidence of the utility of PCT and IL-6 as an efficient UTI marker.

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None

Interest conflict

The authors declare no conflict of interest.

Author's contribution

Suhaila N. Darogha presented the idea, conceived the design. Sarhang H. Azeez laboratory work and statistical analysis of data. Zhian G. Abdullah wrote the manuscript.

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