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Research Article

Performance of a multi-criteria algorithm based on urinalysis in predicting negative urine culture results

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Abstract

Objectives: Urinalysis by automated urine analyzer, including microscopy and dipstick test, is easy, rapid, and inexpensive. Among these parameters, leukocyte count (WBC) or leukocyte esterase (LE) has relatively sufficient sensitivity in urinary tract infection (UTI) detection. Urine culture is accepted as the gold standard for the diagnosis of UTI. Unnecessary culture requests constitute the majority of the microbiology laboratory workload. This study aimed to evaluate the performance of a multi-criteria algorithm by using urinalysis in predicting negative urine cultures.

Methods: During a 2-week period, randomly selected 600 urine samples that reached the microbiology laboratory for urine culture were subjected to chemical and microscopic urinalysis without waiting. Urinalysis was performed using Iris iQ200 (Iris Diagnostics, Chatsworth, USA). LE >25 μL^{-1}, nitrite positive, and WBC >5 per HPF were accepted as abnormal/positive. The sensitivity and specificity were also calculated by Receiver operating characteristic analysis at alternative threshold values for all small particles. A multi-criteria algorithm based on these urine parameters was developed, and its performance was evaluated by determining specificity, sensitivity, negative predictive value (NPV), and positive predictive value.

Results: Multi-criteria algorithm based on urinalysis gave false-negative results for only 3 (0.5%) samples. This algorithm had 98.9% NPV and eliminated 47.4% of urine samples from the culture workflow.

Conclusion: This algorithm may reduce unnecessary culture requests and more effective use of time and resources.

Keywords: Urinalysis, urine culture, urinary tract infection

Urinary tract infection (UTI) is the most common bacterial infection in both inpatients and outpatients [1]. *Escherichia coli* is the most commonly isolated bacterium [2]. Urine culture, which is accepted as the gold standard for diagnosing UTI, is expensive and includes a time-consuming procedure. The absence of growth in the majority of patient samples indicates that unnecessary culture requests constitute most of the microbiology laboratory workload [3, 4].

Urinalysis by automated urine analyzer, including microscopy and dipstick test, is an easy, rapid, and inexpensive diagnostic tool for the early diagnosis of UTI [5]. Studies showing the performance of urinalysis in excluding UTI have reported that leukocyte count (WBC) or leukocyte esterase (LE) has higher sensitivity, but lower specificity than nitrite [6-8]. Detection of nitrite positivity indicates the presence of reductase-producing bacteria that convert nitrate to nitrite. This test may cause false-negative results because this conversion requires time, and not all microorganisms have the ability to reduce nitrate to nitrite [4]. In the presence of trauma or catheterization, pyuria may occur; therefore, WBC or LE is not specific [2].

Automated urine microscopy analyzer Iris iQ200 detects a different parameter called "all small particles" (ASP), which consists of unclassified particles of 3 µm, to improve the sensitivity of the assay about the presence of a potential infection [9].

The sensitivity of urinalysis in detecting urine samples with negative culture results may differ according to the method used for urine microscopy, patient characteristics (age, gender, underlying diseases), and defined threshold values for urinalysis parameters. Therefore, a single parameter is not enough in predicting urine culture results [4, 6, 10]. Multi-criteria al-

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gorithms may have better performance in excluding UTI and eliminating urine samples with negative culture results [11]. The best combination of them is still unclear. Moreover, in many hospitals, urinalysis and urine culture are requested together despite the lack of suitable indication.

The objective of this study was to evaluate the performance of a multi-criteria algorithm by using urinalysis (LE, WBC, nitrite, and ASP) in predicting negative urine culture results.

Materials and Methods

Study design

The present study was conducted using 600 urine specimens in the microbiology laboratories of Erciyes University Hospital for urine culture. Approval for the study was granted by the Ethics Committee of Erciyes University Faculty of Medicine.

Patients and samples

During a 2-week period, randomly selected 600 urine samples that were submitted to the microbiology laboratory for urine culture were subjected to chemical and microscopic urinalysis without waiting. Insufficient samples for urinalysis were excluded from the study. As polymicrobial growth (contaminated) was detected in 20 samples, culture results of 580 urine samples were compared with urinalysis.

The demographic information of patients and distribution of clinical settings requesting urine culture were investigated by scanning electronic medical records.

Urinalysis

Urinalysis was performed using Iris iQ200 (Iris Diagnostics, Chatsworth, USA), an automated digital imaging-based system that uses flow cytometry and iChemVelocity system (Iris Diagnostics, USA). Urinalysis was completed within 1 h after taking samples. The parameters included nitrite, LE, WBC, and ASP. Particles smaller than 3 µm, such as cocci, some other bacteria, crystals, and other formed elements, were classified by the system as ASP [9]. LE >25 μL^{-1}, nitrite positive, and WBC >5 per HPF were accepted as abnormal/positive. Different threshold values were investigated for ASP and 5155 μL^{-1} value, which has lower sensitivity and a negative predictive value (NPV), was accepted as the threshold value. The results obtained from urinalysis were compared with urine cultures. WBC, LE, nitrite, and ASP were added step by step to develop a multi-criteria algorithm, and its performance was evaluated in predicting samples with negative culture results.

Urine culture

A quantity of 10 µL of urine specimens was cultured on 5% sheep blood and MacConkey/eosin methylene blue agar (Oxoid, UK). After incubation for 18-24 h at 35±2°C, the growth of ≤2 microorganisms, each ≥10 000 CFU/mL, was considered positive [2]. The Phoenix automated system (Becton Dickinson, USA) was used to identify the bacteria, supported by conventional tests when needed.

The specimen was accepted contaminated (mixed growth) whenever three or more various colonies with no dominant type had grown (mixed flora) and were refused.

Statistical analysis

Receiver operating characteristic (ROC) curves, sensitivity, specificity, NPV, and positive predictive value (PPV) were calculated for all criteria with different threshold values. All statistical analysis was carried out using MedCalc Ver. 15.2. Different thresholds were investigated to obtain the most optimal result. We calculated the area under the ROC curve (AUC) with 95% confidence interval (CI). A $p<0.05$ was accepted as statistically significant.

Results

The distribution of the patients according to age, gender, and clinical settings is shown in Table 1. Approximately 38% of patients were males and 62% were females. Of the total patients, 48% of them were pediatric patients.

Table 1. Distribution of the patients according to age, gender, and clinical settings

Patient characteristics	n=580	%
Age		
<18	27	48
18-65	219	38
>65	83	14
Gender		
Male	221	38
Female	359	62
Clinical settings		
Pediatrics	256	44
Urology	154	27
Obstetrics	41	7
Others	129	22
Outpatients/inpatients	535/45	92/8

Table 2. Identified pathogens in urine culture

Pathogens	n	%
Escherichia coli	58	72
Klebsiella spp.	6	7
Enterococcus spp.	6	7
Proteus spp.	4	5
Pseudomonas spp.	3	4
Streptococcus spp.	2	3
Staphylococcus spp.	1	1
Enterobacter spp.	1	1
Total	81	100

Table 3. Performance characteristics of urine parameters at different threshold values

Urine parameters	Threshold	Sensitivity	Specificity	PPV	NPV
Leukocyte esterase	>0	81.4	64.7	27.2	95.5
	>25	75.3	73.7	68.2	94.8
White blood cell	>3	79	67.5	28.3	95.2
	>5	69.1	76.9	32.7	93.8
Nitrite	Positive	61.7	99.8	98	94.1
All small particles	>5155	88.8	78.9	40.6	97.7
	>13 060	5.3	95.0	70.9	95.9

PPV: Positive predictive value; NPV: Negative predictive value.

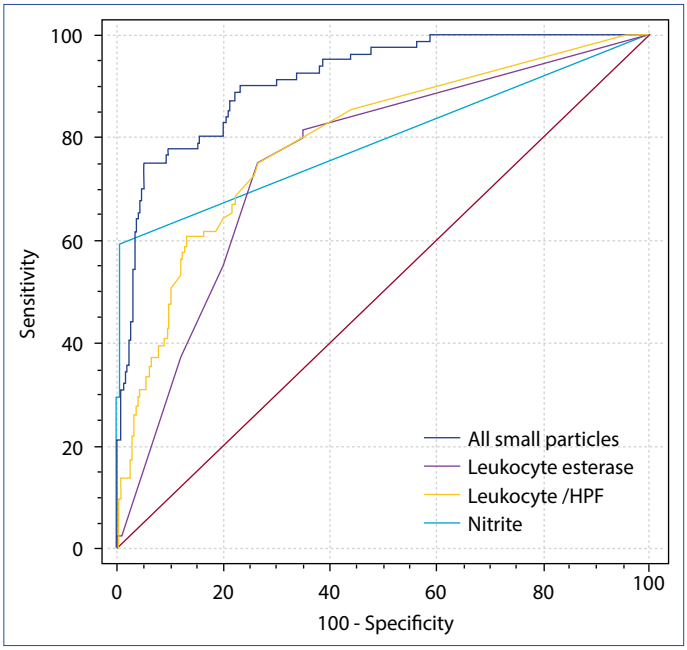

Figure 1. ROC curves for leukocyte esterase (>25, AUC 0.757, 95% CI=0.720 to 0.791, p<0.001), nitrite (AUC 0.795 p<0.001), WBC (>3, AUC 0.795, 95% CI=0.760 to 0.827, p<0.001) and all small particles (>13060, AUC 0.916, 95% CI=0.890 to 0.937, p<0.001).

ROC: Receiver operating characteristic; AUC: Area under the curve; CI: Confidence interval; WBC: White blood cell; HPF: High power field.

In total, 81 (13.9%) patients had positive urine culture. Among the 81 positive samples, a single microorganism was identified in 80 samples and two pathogens were identified in 1 sample. The most frequently determined microorganism in urine culture was *E. coli*. Other pathogens are shown in Table 2.

The performance of urinalysis parameters is given in Table 3. Nitrite had the highest specificity and PPV, but it had the lowest sensitivity. LE, WBC, and ASP had higher sensitivity than nitrite.

The ROC curves are plotted according to the optimal threshold value for each urine parameter in Figure 1. The AUC for WBC, nitrite, and ASP were 0.795, 0.795, and 0.916, respectively. Urine culture was accepted as a reference method for ROC analysis.

Although ASP had the largest area under the curve, the algorithm was generated to obtain the most effective criteria in predicted samples with negative urine cultures, using the nitrite, LE, WBC, and ASP values. With this algorithm, it was tried to obtain the lowest false-negative results without increasing false positives as much as possible. Only three outpatient female patients were evaluated as false-negative, as shown in Figure 2.

This algorithm excluded 275 (47.4%) of 580 specimens from the culture workflow (Table 4).

Discussion

Among the important findings of the present study are that 86% of 580 urine samples had negative culture results, a multi-criteria algorithm based on urinalysis gave false-negative results for only 3 samples, and this algorithm was able to eliminate 47.4% of urine samples from culture workflow.

Previous studies reported >75% unnecessary culture requests [7, 12-14]. Researchers found the sensitivity and specificity for LE to be 58.8%-85% and 61%-75% and those for nitrite to be 22%-51.4% and 94%-99%, respectively [7, 15-17]. It has been reported that nitrite had the highest specificity and PPV, while LE had higher sensitivity and NPV [7, 17]. Additionally, AUC was 0.818 for WBC and 0.774 for LE [7]. These findings were similar to the present study.

Another study showed that bacterial counts measured by urine flow cytometry provided better diagnostic accuracy than WBC or nitrate and LE [8]. Kayalp et al. [12] reported that bacteriuria had a specificity of 97.8% and a sensitivity of 78.8%. Bacteriuria was determined with digital imaging of sediment by automated microscopy. When bacteriuria and nitrite combination was used, NPV of 99.5% could be achieved. Öztürk et al. [7] found a sensitivity and specificity of 70% and 83%, respectively, for bacteriuria. They detected bacteriuria with digital flow microscopy and automatic particle identification. AUC was 0.798 for bacteria, and the false-negative rate was reported to be 30%. Another study reported a false-negative rate of 21.2% for bacteriuria [12].

It has been suggested that multi-criteria algorithms are more successful in estimating patients with negative urine culture results. In one study, an algorithm based on nitrite and WBC negative and ASP <2022 was able to identify 26.5%

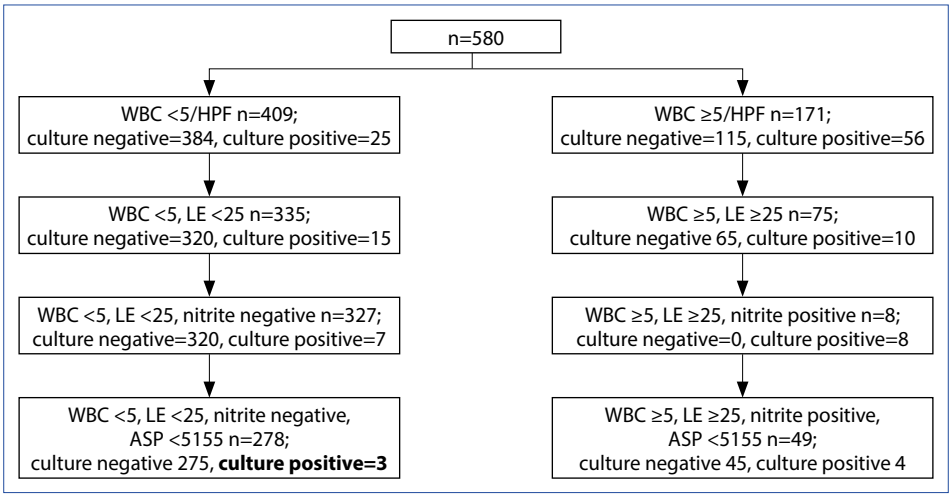

Figure 1. Most effective criteria for elimination of negative urine culture results.
WBC: White blood cell; HPF: High power field; LE: Leukocyte esterase; ASP: All small particles.

Table 4. Performance of a multi-criteria algorithm

Algorithm	Urine culture (negative)	Urine culture (positive)	Total
Culture is necessary	224	78	302
Culture is not necessary	275	3	278
Total	499	81	580

Sensitivity=96.3%; specificity=55.1%; PPV=25.8%; NPV=98.9%.

of samples with a negative urine culture. With this algorithm, 98.9% sensitivity and 32.2% specificity were obtained. Moreover, an NPV of 100% could be achieved using a multi-criteria algorithm. However, the false-positivity rate increase does not provide the expected decrease in the number of samples sent to urine cultures. They were able to achieve a 15.7% reduction in the culture workup [11]. Lynch et al. [18] found that urine reflex culturing resulted in a nearly 40% decrease in cultures.

Previous investigators reported 94.2% NPV and a culture saving rate of 48.9% [7]. Stürenburg et al. [19] found a sensitivity of 95%, specificity of 61%, and a culture saving rate of 35% under the combinations of urine parameters. Another study with the iQ200 system exhibited an acceptable NPV of 97.7% and approximately 50% reduction of urine culture when using WBC ≥ 4 per HPF as a threshold value in predicting urine culture results, but the PPV was only 24.5% in the same study [20].

The limitations of the present study had a low number of patients. In addition, we could not access clinical information of patients such as symptomatic/asymptomatic.

The present study showed that the number of unnecessary urine cultures could have been reduced by about 50% if this algorithm had been used. This finding also means significant savings in time and costs can be achieved. However, false-negative results might occur when the most effective criteria are used. For this reason, laboratory results should be evaluated together with the patient's clinical information and examination findings. The false-negative rate was only 0.5% in the present study. In addition, blood tests such as C-reactive protein or complete blood count may be used at this stage.

Conflict of Interest: The authors declare that there is no conflict of interest.

Ethics Committee Approval: The study was approved by The Erciyes University Faculty of Medicine Clinical Research Ethics Committee (No: 2022/124, Date: 09/02/2022).

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