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Evaluation of a Leukocyte Dip-Stick Test Used for Screening Urine Cultures

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A 2-min leukocyte esterase dip-stick test for pyuria was used to screen 409 consecutive clinical specimens received for culture. The leukocyte esterase dip-stick test proved to be a sensitive and inexpensive means of screening urine specimens for the absence of leukocytes and, by inference, for the absence of significant bacteruria.

More than half of all urine specimens received for routine culture in community hospital settings will not contain culturable pathogenic bacteria, whereas a small but significant portion of the culture-positive specimens will contain various levels of "contaminating" organisms assumed to have originated not from the urinary tract but from skin, vaginal, or perianal sources (4, 5). By definition, urine specimens having bacteria or yeasts derived solely from contaminating sources would not contain leukocytes or other evidence of ongoing urinary tract infection and could reasonably be exempted from the normal bacteriological work-up. The advantages of a screening procedure that would reliably and economically separate specimens which contained evidence of clinical infection from those which did not are thus obvious. The introduction of Chemstrip (Bio-Dynamics, Indianapolis, Ind.) urine dip-sticks, which includes the leukocyte esterase test (LET), appears to offer precisely this possibility through biochemical detection of lysed leukocytes in the voided specimen.

The Chemstrip 9 version was selected for evaluation, and a total of 412 consecutive urine specimens from all sources were processed. Three specimens (<1%) were grossly bloody and not suitable for dip-stick testing. Gross specimen color and clarity were recorded immediately upon receipt for the remaining 409 specimens, as were the dip-stick "9" results for LET, blood, protein, and nitrite (glucose, ketones, urobilinogen, bilirubin, and pH are also determined on dip-stick "9"). Dip-stick test reactions were observed and recorded at the time intervals specified by the package insert except that a final LET reading was not made until a full 2 min had elapsed. This 2-min distinction is important for increased LET sensitivity; earlier package inserts had recommended a 1-min wait, and most current literature reflects the prior instruction.

Gram stains were performed on all specimens by allowing 20 μl of uncentrifuged urine to dry on a slide without spreading. This procedure is traditionally assumed to produce ≥1 bacterium and ≥1 leukocyte per oil immersion field when the count is $\geq 10^5$ CFU/ml and pyruia is present (1). Although still controversial, an acceptable normal level of leukocytes in urine is ≤5 per mm^3; excretion at levels of ≥20 cells per mm^3 is assumed by most physicians to have clinical significance (2, 3). The level of reliable detection of leukocytes by the LET has been reported to be ≥20 cells per mm^3 (3).

Routine aerobic 24-h quantitative cultures were made by spreading 1.0 μl of uncentrifuged urine onto one 5% sheep blood tryptic soy agar plate and one eosin methylene blue plate. Cultures were typically evaluated after 24 h of incubation at 35°C, but 48 h was allowed when there was an obvious discrepancy between Gram stain and culture results. A significant positive culture was defined as one having $\geq 5 \times 10^4$ CFU/ml (voided) or $\geq 1 \times 10^3$ CFU/ml (any catheterized specimen) for less than three species of bacteria or $\geq 1 \times 10^3$ CFU of yeast per ml (any specimen).

Of 409 urine specimens screened, 63 were judged not to have clinical significance: 4 with questionable Gram stains (rare leukocytes) but negative culture and LET results; 11 with charted evidence of bedpan collection or vaginal contamination; and 48 which contained viable bacteria but fell within our existing in-house criteria for contamination (Table 1). Although not universally standardized, these criteria appeared to reflect the general clinical laboratory approach to contaminated urine specimens (1, 2, 5, 6). There were no voided specimens containing fewer than 5×10^4 CFU of

TABLE 1. LET and Gram stain results for 63 contaminated specimens

Contamination	LET positive/ Gram stain positive	LET positive/ Gram stain negative	LET negative/ Gram stain positive	LET negative/ Gram stain negative
Scanty[a]	4	13	2	16
Gross[b]	9	3	0	1
Charted evidence of bedpan collection or vaginal contamination			6[c]	9

[a] Scanty contamination, Slight growth of skin or vaginal flora.
[b] Gross contamination, $>10^5$ CFU/ml of >2 organisms.
[c] Includes four questionable Gram stains (rare leukocytes) from patients with no evidence of infection.

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TABLE 2. LET, Gram stain for leukocytes, and culture results for 346 urine specimens[a]

LET result	Culture positive/ Gram stain positive	Culture positive/ Gram stain negative	Culture negative/ Gram stain positive	Culture negative/ Gram stain negative
Negative	3 (2)	6 (5)	0 (0)	179 (166)
Positive	80	16	8	54

[a] A positive culture was defined as $\geq 5 \times 10^4$ (voided) or $\geq 1 \times 10^3$ CFU/ml any catheterized specimen) of ≤ 2 species of bacteria or $\geq 1 \times 10^3$ CFU/ml of yeast. The numbers in parentheses indicate the number of specimens also having a negative nitrite result.

crease in sensitivity could be effected by incorporating protein or blood dip-stick results or both, whereas inclusion of any combination of positive results for these tests would have greatly increased the number of nonproductive cultures. Inclusion of a gross appearance criterion such that only lightly colored and clear specimens would be subject to LET screening would have eliminated at least four of the false-negative LETs (Table 3), but the probable concomitant increase in nonproductive cultures was not evaluated.

In summation, of the 346 noncontaminated specimens, 52% (179/346) were correctly identified as culture negative

TABLE 3. Laboratory data for specimens having a false-negative LET result

Specimen	Specimen clarity	Specimen color	Gram stain for leukocytes	Nitrite test	Culture result	
					CFU/ml	Organism
825	Cloudy	Amber	Negative	Negative	5×10^4	*Pseudomonas aeruginosa*
836	Cloudy	Yellow	Negative	Negative	5×10^4	*Pseudomonas aeruginosa*
921	Cloudy	Amber	Negative	Negative	$>10^5$	*Klebsiella pneumoniae*
661	Cloudy	Orange	Positive	Negative	$>10^5$	*Klebsiella oxytoca*
712	Slightly cloudy	Yellow	Negative	Negative	1×10^4	*Candida albicans*
715	Slightly cloudy	Yellow	Positive	Negative	$>10^5$	*Escherichia coli*
622	Clear	Yellow	Negative	Negative	1×10^5	*Candida tropicalis*
601	NR[a]	NR	Positive	Positive	$>10^5$	*Escherichia coli*
497	NR	NR	Negative	Positive	$>10^5$	*Klebsiella pneumoniae*

[a] NR, Not recorded.

bacteria per ml in which physicians felt the isolate had clinical significance.

Of the 346 noncontaminated specimens, 179 (52%) had negative LET, culture, and Gram stain results (Table 2). An additional nine specimens with negative LETs were culture positive: three were positive for leukocytes by Gram stain, whereas six were negative (five of the latter patients were immunocompetent, and the sixth was possibly immunocompromised by prednisone therapy for advanced rheumatoid disease). For the purposes of this investigation, a false-negative LET was defined as a negative LET result from specimens with a significant positive culture. The predictive value of a negative LET (for a negative culture) was therefore 95% (179/188).

Quantitative cultures were positive for 96 of 158 urine specimens with a positive LET. The predictive value of a positive LET (for a positive culture) was thus only 61% (96/158), and no further attempt was made to correlate nitrite or other test results with specimens with a positive LET. Since 8 of the 62 culture-negative specimens revealed leukocytes by Gram stain, however, an unidentifiable portion of the 39% apparent false-positive results might be attributable to viral, chlamydial, or anaerobic pathogens.

Our data indicate that the LET, if used as the sole criterion for not culturing urine specimens, had a sensitivity of 91.4% (96/105). Two of the nine specimens with false-negative LETs had positive nitrite tests (Table 3). If negative results for both the LET and the nitrite test had been made the criteria for not culturing, the accuracy would have increased to 93.3% (98/105), and the number of nonproductive cultures would have increased by 13 (Table 2). No consistent inby a negative LET, whereas 4.8% (9/188) were falsely negative. Inclusion of a negative nitrite test into screening criteria would have lowered the false-negative rate to 3.7% (7/188). Although any false-negative rate may be unacceptable to some clinicians, the advantages of reduced costs and immediate turnaround for greater than 50% of all urine specimens should be carefully weighed in comparison.

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