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Positive Extended-Spectrum-β-Lactamase (ESBL) Screening Results May Be Due to AmpC β-Lactamases More Often than to ESBLs[▽]

AmpC β-lactamases can interfere with extended-spectrum-β-lactamase (ESBL) confirmatory tests. Resulting failures to detect ESBLs can endanger patients because false susceptibility to cephalosporins may be reported (5). This problem occurs with CLSI and some other ESBL confirmatory tests, but there are tests that can be used to provide more accurate detection of ESBLs in AmpC-producing isolates (4, 5). While inaccurate susceptibility reports are never acceptable, it is of interest to know whether it is common for a lab to encounter ESBL-screen-positive, AmpC-producing isolates that yield negative CLSI confirmatory tests. That is, can it be common to encounter negative ESBL confirmatory test results of unknown accuracy? Apart from a small *Escherichia coli* study in which 20 of 26 cefpodoxime ESBL-screen-positive, cefoxitin-resistant *E. coli* isolates produced a plasmid-mediated AmpC β-lactamase (4), little is known about how often AmpC production creates uncertainty about the accuracy of CLSI ESBL confirmatory tests. Therefore, a study was conducted at a Louisville, KY, teaching hospital to determine how many ESBL-screen-positive isolates of *E. coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca*, and *Proteus mirabilis* were AmpC positive and ESBL confirmatory test negative by CLSI methodology.

During the period of 1 May 2006 to 28 February 2007, 952 isolates from patients at the University of Louisville Hospital, Louisville, KY, were analyzed. They comprised 682 *E. coli*, 152 *K. pneumoniae*, 44 *K. oxytoca*, and 70 *P. mirabilis* isolates. ESBL screening was performed by using CLSI microdilution methodology using cefotaxime, ceftriaxone, ceftazidime, cefpodoxime, and aztreonam with positive isolates then tested by the CLSI ESBL disk confirmatory method (2). Screen-positive isolates with negative ESBL confirmatory tests were investigated for AmpC production using a Tris-EDTA-based disk test (1) to determine if they were AmpC producers. If this test was positive, it meant that the negative ESBL confirmatory result was of unknown accuracy. Representative AmpC-positive isolates of *K. pneumoniae* were investigated for gene identification by PCR using primers specific for bla_{DHA}, bla_{FOX}, bla_{CMY}, bla_{ENT}, bla_{ACT}, and bla_{ACC} (3).

Only 13 of the 57 screen-positive isolates (22.8%) were ESBL producers by CLSI methodology, while 40 were AmpC producers (70.2%) (Table 1). Other resistance mechanisms were assumed to account for the other four positive screens. The screening-positive isolates comprised 37 *E. coli*, 16 *K. pneumoniae*, 3 *Klebsiella oxytoca*, and 1 *P. mirabilis*. Of these, 8 *E. coli* isolates (22% of screen-positive *E. coli*), 4 *K. pneumoniae* isolates (25% of screen-positive *K. pneumoniae*), and 1 *P. mirabilis* isolate were confirmed as ESBL producers. Of the isolates that gave an ESBL-negative result, 28 *E. coli* (76% of screen-positive isolates) and 12 *K. pneumoniae* (75% of screen-positive isolates) were AmpC positive. No *K. oxytoca* or *P. mirabilis* isolates were AmpC positive. The AmpC-positive *K. pneumoniae* isolates were assumed to produce a plasmid-mediated or imported AmpC β-lactamase because this organism's genome lacks an *ampC* gene. PCR testing of six representative *K. pneumoniae* isolates identified a FOX-like β-lactamase gene in each isolate. The AmpC-positive *E. coli* isolates were assumed to produce either a chromosomally mediated or plasmid-mediated AmpC β-lactamase.

TABLE 1. ESBL-screen-positive isolates that produced extended-spectrum or AmpC β-lactamases

Species	No. (%) of isolates with indicated test result		
	ESBL screening positive	ESBL positive[a]	AmpC positive
E. coli	37	8 (22)	28 (76)
K. pneumoniae	16	4 (25)	12 (75)
K. oxytoca	3	0	0
P. mirabilis	1	1	0

[a] AmpC status was not investigated if ESBL-positive isolates were confirmed because such isolates were not those in which AmpC production interfered with ESBL detection.

In conclusion, the aim of the study was not to reconfirm that AmpC β-lactamases may interfere with ESBL detection or to define methodology to overcome this problem (5), but rather to determine if it was common for AmpC production to necessitate the use of a more reliable ESBL confirmatory test. In this study, *E. coli* and *K. pneumoniae* isolates yielded positive ESBL screening results three times more often for AmpC-producing isolates that yielded negative CLSI ESBL confirmatory test results than for isolates with positive CLSI ESBL confirmatory test results. That is, ESBL confirmatory test results of unknown accuracy occurred often enough to warrant the need for an alternative ESBL confirmatory test of greater accuracy for AmpC-producing isolates. The results also indicated that the CLSI ESBL screening recommendations are useful to screen for AmpC β-lactamases.

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