

Table 3A. (Continued)

Test	Criteria for Performance of ESBL Test		ESBL Test		
Test method	Disk diffusion		Disk diffusion	Broth microdilution	
Inoculum	Standard disk diffusion procedure		Standard disk diffusion procedure	Standard broth dilution procedure	
Incubation conditions	35°C ± 2°C; ambient air		35°C ± 2°C; ambient air	35°C ± 2°C; ambient air	
Incubation length	16-18 h		16-18 h	16-20 h	
Results	For <i>K. pneumoniae</i> , <i>K. oxytoca</i> , and <i>E. coli</i> :		Growth at or above the concentrations listed may indicate ESBL production (ie, for <i>E. coli</i> , <i>K. pneumoniae</i> , and <i>K. oxytoca</i> , MIC ≥ 8 µg/mL for cefpodoxime or MIC ≥ 2 µg/mL for ceftazidime, aztreonam, cefotaxime, or ceftriaxone; and for <i>P. mirabilis</i> , MIC ≥ 2 µg/mL for cefpodoxime, ceftazidime, or cefotaxime).	A ≥ 5-mm increase in a zone diameter for either antimicrobial agent tested in combination with clavulanate vs the zone diameter of the agent when tested alone = ESBL (eg, ceftazidime zone = 16; ceftazidime-clavulanate zone = 21).	A ≥ 3 2-fold concentration decrease in an MIC for either antimicrobial agent tested in combination with clavulanate vs the MIC of the agent when tested alone = ESBL (eg, ceftazidime MIC = 8 µg/mL; ceftazidime-clavulanate MIC = 1 µg/mL).
	Cefpodoxime zone	≤ 17 mm			
	Ceftazidime zone	≤ 22 mm			
	Aztreonam zone	≤ 27 mm			
	Cefotaxime zone	≤ 27 mm			
	Ceftriaxone zone	≤ 25 mm			
	For <i>P. mirabilis</i> :				
	Cefpodoxime zone	≤ 22 mm			
	Ceftazidime zone	≤ 22 mm			
	Cefotaxime zone	≤ 27 mm			
Zones above may indicate ESBL production.					
Reporting			For all confirmed ESBL-producing strains: If laboratories use current cephalosporin and aztreonam breakpoints, test interpretations for these agents do not need to be changed from susceptible to resistant.		

Table 3A
Tests for ESBLs

Table 3A. (Continued)

Test	Criteria for Performance of ESBL Test				ESBL Test	
Test method	Disk diffusion		Broth microdilution		Disk diffusion	Broth microdilution
QC recommendations	When testing antimicrobial agents used for ESBL detection, <i>K. pneumoniae</i> ATCC ^{®b} 700603 is provided as a supplemental QC strain (eg, for training, competence assessment, or test evaluation). Either strain, <i>K. pneumoniae</i> ATCC [®] 700603 or <i>E. coli</i> ATCC [®] 25922, may then be used for routine QC (eg, daily or per IQCP).		When testing antimicrobial agents used for ESBL detection, <i>K. pneumoniae</i> ATCC [®] 700603 is provided as a supplemental QC strain (eg, for training, competence assessment, or test evaluation). Either strain, <i>K. pneumoniae</i> ATCC [®] 700603 or <i>E. coli</i> ATCC [®] 25922, may then be used for routine QC (eg, daily or per IQCP).		When performing the ESBL test, <i>K. pneumoniae</i> ATCC [®] 700603 and <i>E. coli</i> ATCC [®] 25922 should be used for routine QC (eg, daily or per IQCP).	When performing the ESBL test, <i>K. pneumoniae</i> ATCC [®] 700603 and <i>E. coli</i> ATCC [®] 25922 should be tested routinely (eg, daily or per IQCP).
	<i>E. coli</i> ATCC [®] 25922 (see acceptable QC ranges in Table 4A-1)		<i>E. coli</i> ATCC [®] 25922 = no growth (see acceptable QC ranges listed in Table 5A-1)		Acceptable QC: <i>E. coli</i> ATCC [®] 25922: ≤ 2-mm increase in zone diameter for antimicrobial agent tested in combination with clavulanate vs the zone diameter when tested alone.	Acceptable QC: <i>E. coli</i> ATCC [®] 25922: < 3 2-fold concentration decrease in MIC for antimicrobial agent tested in combination with clavulanate vs the MIC of the agent when tested alone.
	<i>K. pneumoniae</i> ATCC [®] 700603:		<i>K. pneumoniae</i> ATCC [®] 700603 = Growth:		<i>K. pneumoniae</i> ATCC [®] 700603: ≥ 5-mm increase in zone diameter of ceftazidime-clavulanate vs ceftazidime alone; ≥ 3-mm increase in zone diameter of cefotaxime-clavulanate vs cefotaxime alone.	<i>K. pneumoniae</i> ATCC [®] 700603: ≥ 3 2-fold concentration decrease in MIC for an antimicrobial agent tested in combination with clavulanate vs the MIC of the agent when tested alone.
	Cefpodoxime zone	9-16 mm	Cefpodoxime	MIC ≥ 8 µg/mL		
	Ceftazidime zone	10-18 mm	Ceftazidime	MIC ≥ 2 µg/mL		
	Aztreonam zone	10-16 mm	Aztreonam	MIC ≥ 2 µg/mL		
Cefotaxime zone	17-25 mm	Cefotaxime	MIC ≥ 2 µg/mL			
Ceftriaxone zone	16-24 mm	Ceftriaxone	MIC ≥ 2 µg/mL			

Abbreviations: ATCC[®], American Type Culture Collection; CAMHB, cation-adjusted Mueller-Hinton broth; ESBL, extended-spectrum β-lactamase; FDA, US Food and Drug Administration; h, hour(s); IQCP, individualized quality control plan; MHA, Mueller-Hinton agar; MIC, minimal inhibitory concentration; PK/PD, pharmacokinetic/pharmacodynamic; QC, quality control.

Table 3A. (Continued)**Footnotes**

- a. Preparation of ceftazidime-clavulanate (30 µg/10 µg) and cefotaxime-clavulanate (30 µg/10 µg) disks: Using a stock solution of clavulanate at 1000 µg/mL (either freshly prepared or taken from small aliquots that have been frozen at -70°C), add 10 µL of clavulanate to ceftazidime (30 µg) and cefotaxime (30 µg) disks. Use a micropipette to apply the 10 µL of stock solution to the ceftazidime and cefotaxime disks within 1 hour before they are applied to the plates, allowing about 30 minutes for the clavulanate to absorb and the disks to be dry enough for application. Use disks immediately after preparation or discard; do not store.
- b. ATCC® is a registered trademark of the American Type Culture Collection.