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Characterization of *Klebsiella* isolates by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) and determination of antimicrobial resistance with VITEK 2 advanced expert system (AES)

Alper KARAGÖZ[1], Sümeyra ACAR[1,*], Hanifi KÖRKOCA[2]
[1]Public Health Institution of Turkey, Molecular Microbiology Research and Application Laboratory, Ankara, Turkey
[2]Department of Nursing, School of Health, Muş Alparslan University, Muş, Turkey

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Background/aim: The purpose of the study was to evaluate the performance of the VITEK mass spectrometry (MS) (bioMérieux, France) system for the identification of *Klebsiella* spp. isolated from different sources. Moreover, while assessing the ability of the VITEK 2 automated expert system (AES) to recognize antimicrobial resistance patterns, the researchers have extended the study to compare VITEK 2 with the routine antimicrobial susceptibility testing method.

Materials and methods: This study tested 51 *Klebsiella* spp. isolates that were isolated from environmental examples and clinical examples. Results of conventional methods and the matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) MS were compared. Then, any differing results were compared against a reference 16S rRNA gene sequence, and when indicated, a recA sequencing analysis was done.

Results: VITEK MS correctly identified 100% of the *Klebsiella* spp. isolates. There were two *K. oxytoca* isolates incorrectly identified to the species level with conventional methods according to the 16S rRNA gene sequencing analysis. In addition, a VITEK 2 AST-N261 card was used for the detection of extended spectrum beta-lactamases (ESBL). Using the VITEK 2 AES, ESBL positivity was found at the rate of 16.3% whereas this rate was 4.08% using the disk diffusion method.

Conclusion: MALDI-TOF MS is a rapid and accurate method for the identification of *Klebsiella* spp. Moreover, the bioMérieux AES provides a useful laboratory tool for the interpretation of susceptibility results.

Key words: *Klebsiella* spp., MALDI-TOF MS, VITEK 2 AES

1. Introduction

Klebsiella spp. is a gram-negative bacillus that belongs to the family *Enterobacteriaceae* and causes severe nosocomial infections, including pneumonia and primary blood stream infections (1–3). *Klebsiella pneumoniae* and *Klebsiella oxytoca* are ubiquitous in nature and have two common habitats; one is the environment, including surface water, soils, sewage, and plants (4,5) and the other is humans, including the skin, bowels, bladder, and respiratory tract (3,6,7). It can easily survive in hospitals and is usually transmitted from patient to patient via the hands of health care personnel (3,8,9).

The identification and classification of *Klebsiella* spp. are traditionally based on morphological and physiological properties of the bacteria. Morphological identification of typical and atypical colonies on selective culture is typically followed, in particular, by biochemical assays (10).

In recent years, several reports have shown the feasibility of using matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectrometry (MS) to rapidly identify microorganisms (11).

Matrix-assisted laser desorption/ionization is a sensitive technique used in MS, allowing the analysis of biomolecules such as proteins, peptides, and sugars, as well as large organic molecules that tend to be fragile and fragment when ionized by more conventional ionization methods. The system employs laser light, and avoidance of damage to the biomolecule, vaporization, and ionization are ensured by the matrix. Spectra are used to identify the microorganism. A colony of the microorganism to be analyzed is spread on the target point and covered with the matrix. The formed mass spectra are analyzed by the appropriate software and compared with the registered profiles (12).

* Correspondence: sumeyrasavas@hotmail.com

Antibiotics can treat many infectious diseases in our day; however, treatment with many antibiotics that should be effective has failed due to the resistance developed by the microorganisms against the antibiotics (13).

The emergence of strains of multidrug resistant *Klebsiella pneumoniae* has been reported with increasing frequency in several countries worldwide, especially those producing extended spectrum beta-lactamases (ESBL) associated with the prior use of antibiotics, particularly cephalosporin (4).

ESBLs are enzymes that can hydrolyze oxyimino-cephalosporins and can be inhibited by clavulanic acid (14). There have been many methods established to identify ESBL-generating bacteria. The tests used in this regard include a double-disk synergy test, a three-dimensional test, an E-test, searching for ceftazidime resistance, using higher bacteria density, a combined disk method, and applying narrower resistance limits. There also other methods that are used less frequently such as disk diffusion in a clavulanic acid medium, minimal inhibitor concentration in combination with clavulanic acid, and the automated VITEK system. There are some studies investigating ESBL frequency in Turkey and around the world, but the VITEK method is not yet frequently used.

The VITEK 2 advanced expert system (AES) is an automated system that uses antimicrobial susceptibility data to suggest the phenotype of the tested isolate and thereby determine its susceptibility or resistance to antibiotics (15).

The VITEK 2 system provides two different identification tools. The first includes the analysis and interpretation of the minimal inhibitory concentration (MIC) of several beta lactams using the AES software. The second test is based on simultaneous detection and assessment of the antibacterial activity of cefepime, cefotaxime, and ceftazidime (16).

The present study aims to develop a MALDI-TOF MS method for the identification of the genus *Klebsiella* and species isolated from different sources, and assess the ability of the VITEK 2 AES to recognize resistance phenotypes. The researchers have extended the study to compare VITEK 2 with routine antimicrobial susceptibility testing methods.

2. Materials and methods

2.1. Bacterial strains

A total of 51 environmental and clinical isolates were included in the study. Clinical isolates were isolated from different polyclinic units; only strains st11 and st135 were isolated from the pediatric polyclinic 1 month apart. The strains isolated from potable water are the isolates that were isolated from the same center at different times. There was no epidemiological connection between the environmental samples and the clinical samples. Of the isolates, 46% were isolated from feces, 28% from urine, 22% from potable water, and 4% were isolated from animal stools. Detailed data on the isolates are presented in Table 1.

2.2. Biochemical identification of strains

For the human and animal samples collected with a stool collection container for isolation purposes, the cultures were prepared in a manner such that one colony was in EMG agar and incubated for 48 h at 37 °C. The suspected mucoid-like colonies growing on the EMG agar were moved to a TSA medium. The potable water samples were collected in sterile plastic bottles containing 300 mL of 6 mg thiosulfate. A 100 mL volume of each water sample was filtered through cellulose nitrate filters with a pore size of 0.45 µm by using the vacuum in a membrane filtration device (Sartorius, Gottingen, Germany). Then, the filters were placed into Endo's medium (Sartorius, Gottingen, Germany) and incubated for 48 h at 37 °C. The suspected colonies growing on the medium were moved to a TSA medium. In the study, the bacteria that were in the shape of gram-negative bacilli, oxidase-negative, catalase-positive, non-motile, capsular, DNase-negative, and fermentative among the isolates from the urine and feces of individuals with urinary tract infections and the potable water samples were identified as *Klebsiella* spp. Species-level identification was conducted using the IMVIC (indole, methyl red, Voges–Proskauer, and citrate), urease, lysine decarboxylase, and lactose fermentation tests.

2.3. MALDI-TOF mass spectrometry

Following the phenotype-based identification, all strains were also identified using the VITEK MS MALDI-TOF system. In the VITEK MS system, a small amount of bacteria taken from the isolated colonies in the medium plaques using a disposable 1-µL loop was inoculated as a thin layer into the separated areas on single-use target slides (bioMérieux, France). Then, 1 µL of α-cyano-4-hydroxy cinnamic acid (CHCA) matrix solution (bioMérieux, France) was added to the inoculated bacteria and air-dried. The inoculated yeast isolates were first covered with 0.5 µL of formic acid, air-dried, and then covered with 1 µL of matrix and air-dried again. The prepared slides were processed in the VITEK MS device and the obtained spectra were automatically analyzed via the MYLA software (bioMérieux, France). The strains with VITEK MS results compatible with the phenotype identification were considered to be correctly identified (17). For correct identification of incompatible results, a 16S rRNA sequence was used for the bacteria.

2.4. Data analysis

The VITEK MS results were compared with results obtained from amplification and sequencing of a 500-bp region of the 16S rRNA gene performed using an ABI Prism Big Dye Terminator and the *recA* sequencing was modified following Cesarini et al. (18).

Table 1. General information and biochemical test results for *Klebsiella* spp.

STRAIN NO.	MOTILITY	CAPSULAR	TSI	OXIDASE	INDOLE	GAS in TSI	H2S	MR	VP	UREASE	LYSINE	CITRATE	RESULT	Sources	Date
ST 1	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 2	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 3	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 4	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 5	–	+	s/s	–	–	–	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 6	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 7	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 8	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 9	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 10	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 11	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 12	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 13	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 14	–	+	s/s	–	+	+	–	–	+	+	+	+	*K. oxytoca*	FECES	2012
ST 15	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 16	–	+	s/s	–	+	+	–	–	+	–	+	+	*K. oxytoca*	FECES	2012
ST 17	–	+	s/s	–	–	+	–	+	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 18	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 19	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 20	–	+	s/s	–	–	+	–	–	+	+	–	+	*K. pneumoniae*	FECES	2012
ST 21	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 22	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 23	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 24	–	+	s/s	–	–	+	–	–	+	+	+		*K. pneumoniae*	FECES	2012
ST 25	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 26	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 27	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 28	–	+	s/s	–	–	–	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 29	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 30	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 31	–	+	s/s	–	–	–	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 32	–	+	s/s	–	+	+	–	–	+	+	+	+	*K. oxytoca*	WATER	2012
ST 33	–	+	s/s	–	+	–	–	–	+	+	+	+	*K. oxytoca*	WATER	2012
ST 34	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 35	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 36	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 37	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 38	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 39	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	FECES	2012
ST 40	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 4	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 42	–	+	s/s	–	–	–	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 43	–	+	s/s	–	–	–	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 44	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 45	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	URINE	2012
ST 46	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	Cattle farm	2012
ST 47	–	+	s/s	–	–	+	–	–	+	+	+	+	*K. pneumoniae*	Cattle farm	2012
ST 48	–	+	s/s	–	+	–	–	+	–	+	+	–	*K. pneumoniae*	FECES	2012
ST 49	–	+	s/s	–	–	+	–	+	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 50	–	+	s/s	–	–	+	–	+	+	+	+	+	*K. pneumoniae*	WATER	2012
ST 51	–	+	s/s	–	–	+	–	–	–	–	+	+	*K. pneumoniae*	WATER	2012

2.5. Disk diffusion testing

Antimicrobial susceptibilities for *Klebsiella* spp. isolates were performed using the standard disk diffusion method in Mueller–Hinton agar, in accordance with the procedures of the Clinical and Laboratory Standards Institute. All strains were tested for resistance to the following 17 antibiotics (Oxoid, England): aztreonam (ATM) (30 µg), cefepime (FEP) (30 µg), ceftazidime (CAZ) (30 µg), cefotaxime (CTX) (30 µg), imipenem (IPM) (10 µg), amikacin (AK) (30 µg), gentamicin (GN) (10 µg), ciprofloxacin (CIP) (5 µg), amoxicillin- clavulanic acid (AMC) (25 µg), cephalothin (KF) (30 µg), chloramphenicol (C) (30 µg), tetracycline (TE) (10 µg), sulfamethoxazole/trimethoprim (SXT) (25 µg), cefazolin (CEZ) (30 µg), tobramycin (NN) (10 µg), ofloxacin (OFX) (5 µg), and cefuroxime (CXM) (30 µg). Antibiotic susceptibility was determined according to the guidelines of the Clinical and Laboratory Standards Institute (19). *Escherichia coli* ATCC 25922 was used as the negative control strain.

2.6. VITEK 2

The isolates were tested on a VITEK 2 automated system using the card AST-N261. Strains were subcultured, and then grown for 18–24 h at 37 °C on a sheep blood agar. The bacterial isolate to be tested was diluted to a standard concentration (McFarland 0.5) in 0.45% saline and used to load the test cards for the VITEK 2, which was used according to the manufacturer's directions (bioMérieux, France). For the antibiotics on the antibiotic susceptibility test cards and in each antibiotic-containing test well, a signal was automatically measured every 15 min for 18 h. The resulting data were used to generate growth curves and, through comparison to a control, the MIC of each antibiotic was determined. The calculation was done with an algorithm specific for each antibiotic. *E. coli* ATCC 25922 was used as the negative control.

3. Results

3.1. Bacterial species

The study included 34 clinical isolates from different polyclinics of a single hospital, of which 24 were isolated from feces and 13 from urine, and 51 samples, of which 12 were isolated from potable water and 2 from animal stools. These materials were cultured in sheep blood agar and EMB agar, were identified through classical biochemical tests, and then confirmed via the MALDI-TOF method. As a result of the biochemical tests, all of the isolates were found to be H2S and oxidase negative, and citrate, lysine decarboxylation, and Voges–Proskauer positive. The tests showed that 1.3% of the feces samples and 1.8% of the water samples were indole-positive, and the presence of gas was detected in all samples except 3 urine samples, 1 water sample, and 3 feces samples. All samples, except 3 feces and 2 water samples, were found to be MR positive. All samples, except 1 feces and 1 water sample, were urease negative. All studied samples were non-motile gram-negative bacilli with a capsular structure. The results of all biochemical tests are presented in Table 1.

3.2. Bacterial species with MALDI-TOF MS

The test strains were analyzed by VITEK MS MALDI-TOF and the resulting spectra of their ionizable cell surface components were compared to the spectra in the release database provided with the MALDI-TOF biotyping software. The spectral peaks are shown in the Figure. The findings demonstrated that the VITEK MS system provided the correct results for a diverse group of gram-negative *Klebsiella* spp. bacteria.

Of the samples that were found to be members of the genus *Klebsiella* by the MALDI-TOF method, 2 samples of potable water (st32 and st33) were detected to be *K. oxytoca* with a similarity rate of 99%; furthermore, all isolates were found to be *K. pneumoniae* with a similarity

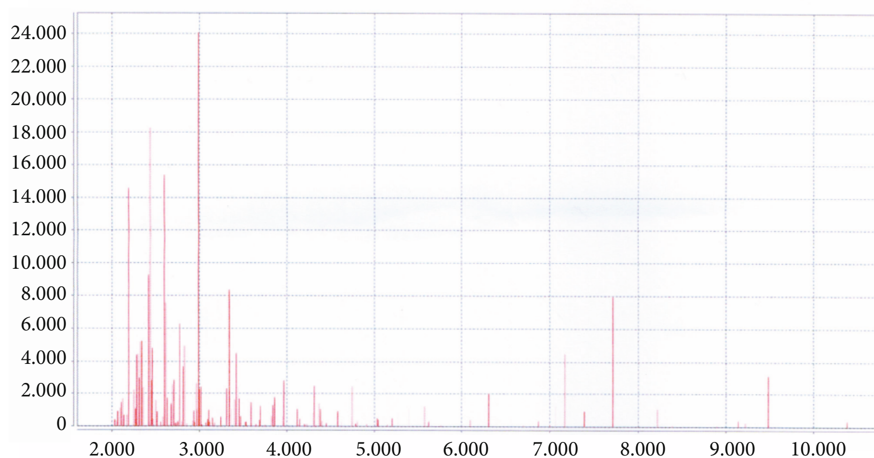

Figure. MALDI-TOF MS spectra of 99% *Klebsiella pneumoniae* (strain 20).

rate of 92.4%–99.9%. The 2 samples that were identified as *K. oxytoca* as a result of the biochemical tests (st14 and st16) were detected to be *K. pneumoniae* by the MALDI-TOF method with a similarity rate of 99.9%. The strains not matched with these two methods underwent 16S rRNA gene sequencing and when indicated, recA sequencing and phenotyping analysis. These samples were identified as *K. pneumoniae* at a rate of 99%. The results for MALDI-TOF identification of the isolates are presented in Table 2.

3.3. Antibiotic susceptibility testing

As shown in Table 3, all *K. pneumoniae* isolates were susceptible to imipenem (IPM) (100%), and amikacin (AK) (100%). Furthermore, a high rate of resistance was seen for *K. pneumoniae* against the following antibiotics: AMC, GN, and OFX (4.08%); FEP, CAZ, NN, C, and CIP (2.04%); KF (14.2%); CEZ, CXM, and ATM (6.1%); and CTX (8.1%). Additionally, *K. pneumoniae* isolates had intermediate levels of resistance to CAZ (6.01%); CIP, AMC, OFX, and ATM (4.08%); CXM, CEZ, FEP, NN, GN C, and TE (2.04%); and KF (26.5%). Finally, the *K. oxytoca* isolates were susceptible to all of the antibiotics.

3.4. Antibiotic susceptibility test with VITEK 2

According to the VITEK 2 AES antibiotic susceptibility results, *K. pneumoniae* isolates were susceptible to IPM, ertapenem (ETP), meropenem (MER), CIP, colistin (CL), and AK. While the samples were resistant to AMC (8.16%), piperacillin/tazobactam (PIP/TAZO) (6.12%), CXM (16.3%), cefuroxime/axetil (16.3%), CAZ (6.1%), ceftriaxone (CRO) (10.2%), FEP (2.04%), GN (4.08%), SXT (10.2%), and ampicillin (AMP) (100%), they were resistant to AMC (4.08%), PIP/TAZO, CAZ (6.12%), CXM, cefuroxime/axetil, cefoxitin (FOX), CRO, and FEP (2.04%) at an intermediate level. When the results of the disk diffusion tests were compared with those of the VITEK 2 AES automated system, consistent results for *K. pneumoniae* were obtained at a rate of 93.6%. With both methods, all samples were found sensitive to AK and IPM. ESBL positivity was found to be 16.3% with the VITEK 2 AES compared to 4.08% with the disk diffusion method. The difference between the two methods for CXM resistance was 16.3% whereas this rate was 8.1% for FEP and CAZ and it was 6.1% for SXT. Antibiotic susceptibility test results for VITEK 2 are given in Table 4.

4. Discussion

Protein profiling using MALDI-TOF MS was developed during the last decade as an important tool for the identification of microorganisms (20). Furthermore, it is a powerful method and is sufficiently reproducible and sensitive enough to rapidly survey the evolution of existing or emerging phenotypes with reduced financial and human costs (4). There are various studies regarding bacteria identification with MALDI-TOF. Wang et al. (10) demonstrated that MALDI-TOF MS identification for *Staphylococcus aureus* was highly correlated with biochemical and serological identification, with an accuracy of 97%. Also, Manji et al. (21) evaluated the performance of the VITEK MS system for the identification of the non-Enterobacteriaceae gram-negative bacilli and provided identification for 92.5% of the isolates; VITEK MS correctly identified 90.9%.

Richter et al. (22) provided identification accuracy of 97% for K. oxytoca in an identification conducted with the bioMérieux MALDI-TOF MS system for this non-Enterobacteriaceae gram-negative bacillus.

Saffert et al. (23) reported a lower percentage of *K. oxytoca* isolates identified to the genus or species level by the Bruker Biotyper (v2.0) software in comparison with the current VITEK MS study. The Bruker Biotyper had difficulty differentiating the genus *Klebsiella* from the closely related genus *Raoultella.*

In the current study, the bioMérieux VITEK MS system was able to identify the *Klebsiella* species with an accuracy of 100%, and the reliability of some results was confirmed by sequencing analysis.

Table 2. MALDI-TOF MS results for *Klebsiella* spp.

STRAIN NO.	Result of MALDI-TOF MS
ST1	99.8% *K. pneumoniae*
ST2–ST10, ST12–ST20, ST22–ST26, ST28–ST31, ST34–ST51	99.9% *K. pneumoniae*
ST 11	95.9% *K. pneumoniae*
ST 21	92.1% *K. pneumoniae*
ST 27	97.7% *K. pneumoniae*
ST 32	99.9% *K. oxytoca*
ST 33	99.9% *K. oxytoca*

Table 3. Antibiotic resistance profiles (disk diffusion testing). AMC = amoxicillin/clavulanic acid, CIP = ciprofloxacin, OFX = ofloxacin, CXM = cefuroxime, KF = cephalothin, CEZ = cefazolin, FEP = cefepime, CAZ = ceftazidime, CTZ = cefotaxime, AK = amikacin, NN = tobramycin, GN = gentamycin, IPM = imipenem, ATM = aztreonam, C = chloramphenicol, SXT = sulfomethaxazole/ trimethoprim (SXT), TE = tetracycline, R = resistant, I = resistant at an intermediate level, and S = susceptible.

STRAIN NO.	AMC	CIP	OFX	CXM	KF	CEZ	FEP	CAZ	CTX	AK	NN	GN	IPM	ATM	C	SXT	TE
ST 1	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 2	S	S	S	I	R	S	S	S	S	S	R	S	S	S	S	S	S
ST 3	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	R	R
ST 4	S	S	S	I	R	I	S	S	S	S	S	S	S	S	I	R	I
ST 5	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 6	S	S	S	S/I	S	S	S	S	S	S	S	S	S	S	S	R	R
ST 7	S	S	S	R	R	R	S	S	R	S	S	S	S	S	S	S	S
ST 8	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 9	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 10	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 11	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 12	S	I	R	R	R	S	S	I	S	S	S	S	S	S	S	R	S
ST 13	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 14	S	S	S	S	R	S	S	S	S	S	S	S	S	S	S	S	S
ST 15	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 16	R	S	S	R	R	R	S	S	S	S	S	S	S	S	S	S	I
ST 17	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 18	S	S	S	R	R	R	S	S	R	S	S	I	S	I	S	S	S
ST 19	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 20	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 21	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 22	I	S	S	R	R	R	R	R	R	S	S	S	S	R	S	R	S
ST 23	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 24	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 25	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 26	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 27	I	S	S	R	R	R	S	S	I	S	S	S	S	I	R	S	S
ST 28	S	S	S	S	R	S	S	S	S	S	S	S	S	R	S	S	S
ST 29	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 30	S	S	S	S	R	S	S	I	S	S	S	S	S	S	S	S	R
ST 31	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 32	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 33	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 34	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 35	S	S	S	S	R	S	S	S	I	S	S	R	S	I	S	S	S
ST 36	S	I	S	S	I	S	S	S	S	S	S	S	S	S	S	R	R
ST 37	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 38	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	R	S
ST 39	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 40	S	S	S	S	R	I	S	S	S	S	S	S	S	S	S	S	R
ST 41	I	S	S	R	R	R	I	I	R	S	I	R	S	R	S	R	S
ST 42	S	R	R	S	I	S	S	S	S	S	S	S	S	S	S	S	R
ST 43	I	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 44	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 45	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 46	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 47	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 48	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
ST 49	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 50	S	S	S	S	I	S	S	S	S	S	S	S	S	S	S	S	S
ST 51	R	S	S	I	R	R	S	S	S	S	S	S	S	S	S	S	S

Table 4. Antibiotic resistance profiles (VITEK AES). ESBL = extended spectrum beta lactamase, AMP = ampicillin, AMC = amoxicillin/clavulanic acid, PIP/TAZO = piperacillin, CXM = cefuroxime, CXM2 = cefuroxime/axetil, FOX = cefoxitin, CAZ = ceftazidime, CRO = ceftriaxone, FEP = cefepime, ETP = ertapenem, IPM = imipenem, MER = meropenem, AK = amikacin, GN = gentamycin, CIP = ciprofloxacin, R = resistant, I = resistant to an intermediate level, and S = susceptible.

STRAIN NO.	SUC NO.	ESBL	AMP	AMC	PIP/ TAZO	CXM	CXM2	FOX	CAZ	CRO	FEP	ETP	IPM	MER	AK	GN	CIP	CL	SXT
ST 1	4	NEG(–)	16(R)	4 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 2	5	NEG(–)	≥32 (R)	4 (S)	≤4(S)	4 (S)	4 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 3	6	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤320 (R)
ST 4	7	NEG(–)	16(R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 5	8	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	0.5 (S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 6	9	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤320 (R)
ST 7	11	POZ (+)	≥32 (R)	16 (I)	8 (S)	≥64 (R)	≥64 (R)	≤4 (S)	2 (I)	16 (R)	2 (I)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	40 (S)
ST 8	13	NEG(–)	≥32 (R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 9	16	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 10	17	NEG(–)	16(R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 11	19	NEG(–)	16(R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 12	20	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 13	21	NEG(–)	≥32 (R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 14	23	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 15	26	NEG(–)	16(R)	4 (S)	8 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 16	30	NEG(–)	16(R)	≤2 (S)	≤4(S)	4 (S)	4 (S)	8 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 17	30	NEG(–)	16(R)	≤2 (S)	≤4(S)	4 (S)	4 (S)	8 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 18	32	POZ (+)	≥32 (R)	4 (S)	≤4(S)	≥64 (R)	≥64 (R)	≤4 (S)	4 (I)	16 (R)	2 (I)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≥16 (R)	≤0.25 (S)	≤0.5(S)	40 (S)
ST 19	36	NEG(–)	16(R)	4 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	0.5 (S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 20	50	POZ (+)	≥32 (R)	≤2 (S)	16(S)	≥64 (R)	≥64 (R)	≤4 (S)	≥64 (R)	≥64 (R)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 21	73	NEG(–)	16(R)	4 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 22	75	POZ (+)	≥32 (R)	≥32 (R)	32 (I)	≥64 (R)	≥64 (R)	≤4 (S)	≥64 (R)	≥64 (R)	32 (R)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤320 (R)
ST 23	77	NEG(–)	16(R)	≤2 (S)	≤4 (S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 24	80	NEG(–)	16(R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 25	81	NEG(–)	16(R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 26	83	NEG(–)	16(R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)

Table 4. (Continued).

STRAIN NO.	SUC NO.	ESBL	AMP	AMC	PIP/ TAZO	CXM	CXM2	FOX	CAZ	CRO	FEP	ETP	IPM	MER	AK	GN	CIP	CL	SXT
ST 27	85	POZ (+)	16 (R)	4 (S)	≤4 (S)	32 (R)	32 (R)	8 (S)	2 (I)	2 (I)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 28	90	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 29	96	NEG(–)	8 (R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 30	99	NEG(–)	≥32 (R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 31	102	NEG(–)	16(R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 32	105	NEG(–)	16(R)	4 (S)	≤4(S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 33	112	NEG(–)	16(R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	0.5 (S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 34	113	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 35	115	NEG(–)	16(R)	4 (S)	8 (S)	≥64 (R)	≥64 (R)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 36	118	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	≥64 (R)	≥64 (R)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤1 (S)	≤0.5(S)	≤320 (R)
ST 37	119	NEG(–)	16(R)	4 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 38	124	NEG(–)	≥32 (R)	≤2 (S)	≤4(S)	2(S)	2(S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 39	128	NEG(–)	16(R)	≤2 (S)	≤4 (S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 40	134	POZ (+)	≥32 (R)	16 (I)	64 (I)	4 (S)	4 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 41	135	POZ (+)	≥32 (R)	≥32 (R)	≥128 (R)	≥64 (R)	≥64 (R)	≤4 (S)	16 (R)	≥64 (R)	2 (I)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≥16 (R)	≤0.25 (S)	≤0.5(S)	≥320 (R)
ST 42	136	NEG(–)	≥32 (R)	≥32 (R)	32 (I)	16 (I)	16 (I)	16 (I)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 43	138	POZ (+)	≥32 (R)	≥32 (R)	≥128 (R)	4 (S)	4 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 44	139	NEG(–)	≥32 (R)	≤2 (S)	≤4 (S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 45	140	NEG(–)	≥32 (R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 46	141	NEG(–)	16(R)	≤2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 47	144	NEG(–)	≥32 (R)	≤2 (S)	≤4 (S)	2 (S)	2 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 48	78	NEG(–)	16(R)	4 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 49	40	NEG(–)	16(R)	≤2 (S)	≤4(S)	4 (S)	4 (S)	8 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 50	41	NEG(–)	16(R)	≤2 (S)	≤4(S)	4 (S)	4 (S)	8 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)
ST 51	1	NEG(–)	16(R)	≤2 (S)	≤4(S)	≤1 (S)	≤1 (S)	≤4 (S)	≤1 (S)	≤1 (S)	≤1 (S)	≤0.5 (S)	≤0.25(S)	≤0.25(S)	≤2 (S)	≤1 (S)	≤0.25 (S)	≤0.5(S)	≤20 (S)

Interest in the VITEK 2 automated system has increased due to its minimization of errors, such as indicating false susceptibility for resistant microorganisms, the computer included in its system, its reduction of workload, and its objectivity in evaluation of the results. Its determination of the resistance profile based on MIC values is considered superior compared to the disk diffusion method (24).

In the current study, the results for the ESBL resistance profile showed a difference between VITEK 2 and the disk diffusion method; the consistency for the genus *Klebsiella* and species between the two methods was found to be 93.8%. The positivity was detected at a rate of 16.3% with VITEK AES as compared to 4.08% with the disk diffusion method. The VITEK 2 automated system has importance in determining the most proper antibiotic to be administered in treatment, thereby accelerating the treatment. Barry et al. (24) found VITEK 2 results to be consistent with routine resistance tests at a rate of 96.5%, 96.7%, and 95.9%, respectively, for staphylococci, enterococci, and gram-negative bacilli. Canton et al. (25) found the VITEK 2 method less reliable than the micro-diffusion method, whereas Leclercq et al. (26) stated that VITEK 2 was more reliable than the agar dilution method. According to Nyberg et al. (27), sensitivity and specificity vary in different studies, but high sensitivity and specificity have been achieved with VITEK 2 when testing *K. pneumoniae* isolates (15,27, 28). They also claimed that VITEK 2 AES was excellent compared to genotypic ESBL determination and it was very comparable to the agar dilution and diffusion methods.

Furthermore, Nyberg et al. reported that when comparing the results of ESBL positivity, the positive results from the VITEK and agar dilution methods were negative in the disc diffusion method. They found the ESBL test result accuracy of the VITEK 2 method to be 94.4% for *K. pneumoniae* and 84.0% for *K. oxytoca* (27).

Livermore et al. (29) demonstrated the capacity of VITEK 2 to detect and interpret resistance mechanisms with a high level of accuracy and standardization. According to Blondel-Hill et al. (30), the AES system is both adaptable and applicable in clinical microbiology laboratories to enhance the accuracy of susceptibility results and the clinical relevance of therapeutic recommendations.

Numanovic et al. (31) studied the VITEK 2 system on 55 isolates of ESBL-producing Enterobacteriaceae members, including 34 *Klebsiella pneumoniae* and 3 *K. oxytoca* isolates, and found that 48 of the 50 genotypically ESBL-producing microorganisms were ESBL positive. That study is consistent with the current study, and shows that it would be appropriate to support the conventional methods with the molecular methods and VITEK in ESBL identification for *K. oxytoca* isolates.

In conclusion, this study analyzes the different sources of *Klebsiella* spp. isolates in Turkey using the MALDI-TOF MS technique. We think that this methodology could be used routinely in clinical microbiology laboratories instead of conventional methods for the genus *Klebsiella*.

Furthermore, antibiotic susceptibility is difficult to detect through conventional methods during the identification of ESBLs, and the molecular methods are time-consuming and expensive for the routine clinical laboratories. The VITEK 2 AES was found to be a successful, reliable, and timesaving system for ESBL identification for the genus *Klebsiella*.

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