

## Matrix-Assisted Laser Desorption Ionization Time-of-Flight Mass Spectrometry for Identification of Microorganisms in Clinical Urine Specimens after Two Pretreatments

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### Abstract

Rapid identification of microorganisms in urine is essential for patients with urinary tract infections (UTIs). Matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS) has been proposed as a method for the direct identification of urinary pathogens. Our purpose was to compare centrifugation-based MALDI-TOF MS and short-term culture combined with MALDI-TOF MS for the direct identification of pathogens in urine specimens. We collected 965 urine specimens from patients with suspected UTIs, 211/965 isolates were identified as positive by conventional urine culture. Compared with the conventional method, the results of centrifugation-based MALDI-TOF MS were consistent in 159/211 cases (75.4%), of which 135/159 (84.9%) had scores  $\geq 2.00$ ; 182/211 cases (86.3%) were detected using short-term culture combined with MALDI-TOF MS, of which 153/182 (84.1%) had scores  $\geq 2.00$ . There were no apparent differences among the three methods ( $p = 0.135$ ). MALDI-TOF MS appears to accelerate the microbial identification speed in urine and saves at least 24 to 48 hours compared with the routine urine culture. Centrifugation-based MALDI-TOF MS is characterized by faster identification speed; however, it is substantially affected by the number of bacterial colonies. In contrast, short-term culture combined with MALDI-TOF MS has a higher detection rate but a relatively slow identification speed. Combining these characteristics, the two methods may be effective and reliable alternatives to traditional urine culture.

**Key words:** Matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS), rapid identification, urinary tract infection

### Introduction

Urinary tract infections (UTIs) are one of the most common infections (Klein and Hultgren 2020). Although mild UTIs do not cause severe organ damage, pathogenic bacteria can lead to adverse consequences such as pyelonephritis, kidney abscess formation, and acute kidney injury through the urethra, bladder, ureter, and other ways, causing septicemia and even death (Korbel et al. 2017; Hsu and Melzer 2018). Fast and reliable microbial identification is essential for the diagnosis and treatment of UTIs. The current diagnosis of UTIs relies on routine urine culture identification, a process that requires 48 hours or longer (de Cueto et al. 2017). Although 16S rRNA gene sequencing, multiplex PCR, and fluorescence *in situ* hybridization can quickly detect pathogens in urine cultures, only 20%

to 30% of clinical urine samples show significant bacterial growth (Manickam et al. 2013). These expensive and cumbersome methods are not suitable for clinical practice (Akoachere et al. 2012).

Matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS) is a new method for rapid microbial identification without the need for target amplification (Nomura 2015). It can replace methods such as 16S rRNA gene sequencing (Vincent et al. 2013). It has a great potential in identifying bacteria directly from urine samples and culture-positive blood samples (Sauget et al. 2017; Nomura et al. 2020). Nevertheless, this novel method has not been widely used in clinical practice. Therefore, in this study, we compared a centrifugation-based MALDI-TOF MS, a short-term culture with MALDI-TOF MS, and the conventional diagnostics of UTI in a clinical practice setting.

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## Experimental

### Materials and Methods

**Sample collection.** From May to August 2020, we collected 965 urine specimens from patients with suspected UTIs from the Second Affiliated Hospital of Wenzhou Medical University (a teaching hospital with 2,667 beds in Wenzhou, Zhejiang, China) and excluded duplicate specimens from the same patient. Samples were collected in 30-ml sterile containers, processed immediately, or stored at 4°C for no more than 8 hours. Each sample was divided into three aliquots: the first aliquot was identified using conventional methods; the second was centrifuged and identified directly using MALDI-TOF MS, and the third was cultured for 5 hours and then identified by MALDI-TOF MS.

**Routine microbiological processing.** After mixing, 10 µl of urine samples were inoculated on a Columbia blood plate (Bio T.K., Zhejiang, China) and incubated at 35°C for 18–48 h in aerobic conditions (Sumsung Laboratory Instrument Co. Ltd., Shanghai, China). We observed bacterial growth and counted colonies. If no bacteria were found after 48 hours, the sample was considered negative. VITEK 2 Compact Automatic bacterial identification and the drug sensitivity analysis system (Bio Mériex, Lyon, France) were used for strain identification.

### The centrifugation-based MALDI-TOF MS

**Sample preparation for MALDI-TOF MS.** Procedures were as follows: (i) 3 ml of urine was placed in a sterile centrifuge tube (Gongdong Medical Technology Co. Ltd. Zhejiang, China) and centrifuged at 715 × g for 10 minutes to remove the cells (USTC ZONKIA Scientific Instruments Co. Ltd., Anhui, China); (ii) supernatants were placed in another 1.5 ml tube; (iii) this was followed by centrifugation at 15,000 × g for 5 minutes to collect the bacteria (Beckman Coulter, Inc, Brea, California, USA); (iv) the supernatants were discarded, and the pellets were mixed with 1.5 ml sterile water; (v) we repeated steps iii and iv; (vi) this was followed by centrifugation at 15,000 × g for 5 minutes; (vi) supernatants were discarded, and the pellets were retained for the subsequent step (Zboromyrska et al. 2016).

**MALDI-TOF MS.** One microliter of the pellet was applied to a clean MALDI target plate (Bruker Daltonik GmbH, Bremen, Germany) and air-dried. Next, it was covered with 1 µl of 70% of formic acid (AIKEDA Chemical Reagent Co. Ltd., Chengdu, China) and left to dry, then covered with 1 µl of matrix solution (α-cyano-4-hydroxy-cinnamic acid solution in 50% acetonitrile and 2.5% trifluoroacetic acid) (Bruker Daltonics) and air-dried. Spectrum acquisitions were obtained using

the default setting through the MALDI Biotyper system (Bruker Daltonics). The final data analysis and bacterial identification scores were achieved using the MALDI Biotyper software (Bruker Daltonics). Each sample was analyzed in duplicate, and the higher score obtained from the two points was recorded as the final score. *Escherichia coli* ATCC 8739 was used as a quality control strain. Based on the manufacturer's instructions, an identification score < 1.70 indicated no identification, an identification score between 1.70 and 2.00 indicated genus identification, and an identification score ≥ 2.00 indicated species identification.

### Short-term culture combined with MALDI-TOF MS

**Urine specimen processing.** After mixing, 10 µl urine samples were inoculated on a Columbia blood plate and incubated at 35°C for 5 h in aerobic conditions.

**MALDI-TOF MS.** The pathogen was identified using MALDI-TOF MS after short-term culture. The bacteria were uniformly coated on the target plate and covered with 1 µl of 70% formic acid. After drying, covered with 1 µl of matrix solution and air-dried. The acquisition and interpretation of the identification results were the same as in the centrifugation-based MALDI-TOF MS.

**Statistical analyses.** The  $\chi^2$  test was used to compare the differences in the pathogen identification among the three methods, and the Student's *t*-test was used to compare the score between the two MALDI-TOF MS methods. Differences were considered statistically significant when  $p < 0.05$ .

## Results

Among the 965 patients with suspected UTIs, 425 were male (44.0%), and 540 were female (56.0%). Because our institution is a teaching hospital and a children's hospital, 374 patients (38.8%) were under 13 years of age, and 202 (20.9%) were under 1 year of age. The mean age was 38.5 (patients under 1 year were counted as 1 year old), the median age was 45 years (IQR 3–68 years). The sources of the samples were as follows: 814 inpatients (84.4%); 149 outpatients (15.5%); one unknown (0.1%).

**Conventional urine culture and two MALDI-TOF MS methods.** Among 965 urine specimens, 211 positive specimens were identified by conventional urine culture, including Gram-negative bacteria 170/211 (80.6%) and Gram-positive bacteria 41/211 (19.4%). The most common microorganisms were *E. coli* in 100 cases (44.2%), followed by *Klebsiella pneumoniae* in 28 cases (12.4%) and *Enterococcus faecalis* in 15 cases (6.6%) (Table I).

Table I  
Identification results by conventional culture and two MALDI-TOF MS methods.

Conventional urine culture (No. of cases)	Centrifugation-based MALDI-TOF MS			Short-term culture combined with MALDI-TOF MS		
	Score > 1.7 (No. of cases)	Score > 2.0 (No. of cases)	Mean Score $\pm$ SD	Score > 1.7 (No. of cases)	Score > 2.0 (No. of cases)	Mean Score $\pm$ SD
<i>Escherichia coli</i> (100)	84	75	2.22 $\pm$ 0.16	95	87	2.17 $\pm$ 0.08
<i>Klebsiella pneumoniae</i> (28)	23	18	2.16 $\pm$ 0.21	26	22	2.18 $\pm$ 0.18
<i>Proteus mirabilis</i> (10)	10	8	2.18 $\pm$ 0.19	9	6	2.04 $\pm$ 0.15
<i>Enterobacter cloacae</i> (9)	8	7	2.14 $\pm$ 0.19	8	6	2.09 $\pm$ 0.18
<i>Pseudomonas aeruginosa</i> (7)	3	2	1.95 $\pm$ 0.13	5	5	2.24 $\pm$ 0.09
<i>Enterobacter aerogenes</i> (4)	3	3	2.27 $\pm$ 0.10	4	4	2.26 $\pm$ 0.10
<i>Citrobacter freundii</i> (4)	4	4	2.22 $\pm$ 0.12	4	4	2.23 $\pm$ 0.11
<i>Acinetobacter baumannii</i> (3)	2	1	2.14 $\pm$ 0.19	2	1	2.03 $\pm$ 0.18
<i>Stenotrophomonas maltophilia</i> (3)	1	1	2.09	0	0	N/A
<i>Morganella morganii</i> (2)	2	2	2.18 $\pm$ 0.16	2	1	2.22 $\pm$ 0.28
<i>Enterococcus faecalis</i> (15)	7	5	2.15 $\pm$ 0.19	10	6	2.03 $\pm$ 0.20
<i>Enterococcus faecium</i> (14)	8	6	2.08 $\pm$ 0.17	12	7	2.04 $\pm$ 0.24
<i>Staphylococcus aureus</i> (2)	1	0	2.20	2	1	1.96 $\pm$ 0.23
<i>Staphylococcus haemolyticus</i> (2)	0	0	N/A	1	1	2.22
<i>Staphylococcus epidermidis</i> (2)	0	0	N/A	1	0	1.76
<i>Streptococcus agalactiae</i> (1)	1	1	2.09	1	1	2.15
<i>Streptococcus gallicans</i> (1)	1	1	2.20	0	N/A	N/A
<i>Streptococcus anginae</i> (1)	1	0	1.75	0	N/A	N/A
<i>Staphylococcus saprophyticus</i> (1)	0	0	N/A	0	0	N/A
<i>Enterococcus avium</i> (1)	0	0	N/A	0	0	N/A
<i>Lactobacillus crispatus</i> (1)	0	0	N/A	0	0	N/A
Total (211)	159	135		182	153	

159/211 (75.4%) positive specimens were detected by the centrifugation-based MALDI-TOF MS. The corresponding rates with routine culture were Gram-negative bacteria: 140/170 (82.4%), Gram-positive bacteria: 19/41 (46.3%) ( $p=0.053$ ). The top three detected bacteria were *E. coli* ( $n=84$ , detection rate: 84.0%), *K. pneumoniae* ( $n=23$ , 82.1%), and *Proteus mirabilis* ( $n=10$ , 100%). Short-term culture combined with MALDI-TOF MS detected 182/211 (86.3%) positive specimens. The corresponding rates with routine culture were Gram-negative bacteria: 155/170 (91.2%), and Gram-positive bacteria: 27/41 (65.9%) ( $p=0.230$ ). The top three detected bacteria were *E. coli* ( $n=95$ ,

detection rate: 95.0%), *K. pneumoniae* ( $n=26$ , 92.9%), *Enterococcus faecium* ( $n=12$ , 85.7%) (Table I). There was no significant difference between the three methods ( $p=0.135$ ) (Table II).

**Comparison between the two MALDI-TOF MS methods.** The centrifugation-based MALDI-TOF MS identified 159 cases with a score  $\geq 1.70$ , and 135/159 cases (84.9%) were identified with scores  $\geq 2.00$ , including Gram-negative bacteria 121/140 (86.4%) and Gram-positive bacteria 14/19 (73.7%). The mean score was  $2.17 \pm 0.24$ . Short-term culture combined with MALDI-TOF MS identified 182 cases with scores  $\geq 1.70$ , and 153/182 cases (84.1%) were identified by

Table II  
Comparison of three methods for identification of Gram-negative bacteria and Gram-positive bacteria.

Method	Identification results		Total	$p$ -value
	Gram-negative bacteria	Gram-positive bacteria		
Conventional urine culture	170	41	211	$p=0.135$
Centrifugation-based MALDI-TOF MS	140	19	159	
Short-term culture combined with MALDI-TOF MS	155	27	182	

Table III  
Comparison of MALDI scores between the two methods.

Germ	Centrifugation-based MALDI-TOF MS		Short-term culture combined with MALDI-TOF MS		<i>p</i> -value
	score $\pm$ SD	RSD	score $\pm$ SD	RSD	
Gram-negative bacteria	2.18 $\pm$ 0.25	11.4%	2.14 $\pm$ 0.18	8.3%	<i>p</i> = 0.113
Gram-positive bacteria	2.10 $\pm$ 0.19	9.1%	2.03 $\pm$ 0.23	11.2%	<i>p</i> = 0.282
Total	2.17 $\pm$ 0.24	11.2%	2.09 $\pm$ 0.20	9.5%	<i>p</i> = 0.003

scores  $\geq 2.00$ , with Gram-negative bacteria 137/155 (88.4%) and Gram-positive bacteria 16/27 (59.3%). The mean score was  $2.09 \pm 0.20$ , slightly lower than centrifugation-based MALDI-TOF MS. Statistical analysis showed that, although the mean MALDI score of the centrifugation-based MALDI-TOF MS was higher than that of short-term culture combined with MALDI-TOF MS in the identification of Gram-negative and Gram-positive bacteria, the difference was insignificant. Nevertheless, in terms of overall identification, the difference in identification score was statistically significant ( $p = 0.003$ , Table III).

To determine correlations between colony counts and the detection rates of MALDI-TOF MS, we compared the identification results in various colony ranges using two MALDI-TOF MS methods. The statistical results showed that there was no difference between the two MALDI-TOF MS methods for the identification of Gram-positive bacteria ( $p = 0.075$ ). In contrast, when identifying Gram-negative bacteria, the difference was significant ( $p = 0.016$ ). Further analysis showed that, when the colony count was more than  $1 \times 10^5$  CFU/ml, the centrifugation-based MALDI-TOF MS and short-term culture combined with MALDI-TOF MS detected 133/147 (90.5%) and 137/147 (93.2%) of Gram-negative bacteria, respectively; there were no significant differences ( $p = 0.394$ ). When counts were between  $1 \times 10^4$  and  $1 \times 10^5$  CFU/ml, however, the detection rate of Gram-negative bacteria in the centrifugation-based MALDI-TOF MS was significantly lower than that in short-term culture combined with MALDI-TOF MS, which were 5/15 (33.3%) and 12/15 (80.0%), respectively; the difference was significant ( $p = 0.025$ ). Moreover, when the count was less than  $1 \times 10^4$  CFU/ml, Gram-negative bacteria were detected in 2/8 (25.0%) and 6/8 (75.0%) by the two methods, respectively. The detection rate decreased significantly with decreased colony count, especially for the centrifugation-based MALDI-TOF MS (Table IV).

We found that, although the identification score of short-term culture combined with MALDI-TOF MS was slightly lower than that of the centrifugation-based MALDI-TOF MS, the detection ability of the short-

term culture combined with MALDI-TOF MS was significantly higher than that of the centrifugation-based MALDI-TOF MS, especially when the colony count was low.

## Discussion

Rapid identification of urinary microorganisms and timely application of antibiotics can significantly reduce the length of hospital stay and costs (Sood et al. 2015). MALDI-TOF MS is a clinical bacterial identification method that can provide microbial identification results within 15 minutes; it is simple to operate and moderately priced ( $< 1$  USD/sample). It is suitable for the microbiological identification of urine specimens from patients with UTIs (Dierig et al. 2015; Sauget et al. 2017).

The primary pathogens of UTIs are Gram-negative bacteria, followed by Gram-positive bacteria, and fungi are in the minority (Flores-Mireles et al. 2015). The detection rates of Gram-negative and Gram-positive bacteria by the centrifugation-based MALDI-TOF MS were 82.3% and 46.3%, respectively. In contrast, the detection rates of Gram-negative and Gram-positive bacteria by short-term culture combined with MALDI-TOF MS were higher, reaching 91.2% and 65.9%. Although there was no statistically significant difference in the overall identification rate between the two MALDI-TOF MS methods ( $p = 0.437$ ), there was a significant difference in the detection results of Gram-negative bacteria when the colony count was between  $1 \times 10^4$  and  $1 \times 10^5$  CFU/ml ( $p = 0.027$ ). The reason may be that the decrease in urine colony counts had a significantly more significant impact on the centrifugation-based MALDI-TOF MS than on short-term culture combined with MALDI-TOF MS. When the colony count was higher than  $1 \times 10^5$  CFU/ml, the bacterial density could meet the requirements for direct identification by MALDI-TOF MS after centrifugation and washing. However, when the bacterial colony number decreased to  $1 \times 10^4$  and  $1 \times 10^5$  CFU/ml, the detection rate of Gram-negative bacteria by the centrifugation-based MALDI-TOF MS dropped signi-

Table IV  
Correlation between colony count and the MALDI-TOF MS identification.

Germ	Centrifugation-based MALDI-TOF MS No. of cases (%)		Short-term culture combined with MALDI-TOF MS No. of cases (%)		<i>p</i> -value
	Detected		Detected		
	Yes	No	Yes	No	
<b>Gram-negative bacteria</b>					
≥ 10 <sup>5</sup> CFU/ml	133/147 (90.5%)	14/147 (9.5%)	137/147 (93.2%)	10/147 (6.8%)	<i>p</i> = 0.394
10 <sup>4</sup> ~ 10 <sup>5</sup> CFU/ml	5/15 (33.3%)	10/15 (66.7%)	12/15 (80.0%)	3/15 (20.0%)	<i>p</i> = 0.027
≤ 10 <sup>4</sup> CFU/ml	2/8 (25.0%)	6/8 (75.0%)	6/8 (75.0%)	2/8 (25.0%)	<i>p</i> = 0.132
Total	140/170 (82.4%)	30/170 (17.6%)	155/170 (91.2%)	15/170 (8.8%)	<i>p</i> = 0.016
<b>Gram-positive bacteria</b>					
≥ 10 <sup>5</sup> CFU/ml	15/25 (60.0%)	10/25 (40.0%)	18/25 (72.0%)	7/25 (28.0%)	
10 <sup>4</sup> ~ 10 <sup>5</sup> CFU/ml	3/12 (25.0%)	9/12 (75.0%)	8/12 (66.7%)	4/12 (33.3%)	
≤ 10 <sup>4</sup> CFU/ml	1/4 (25.0%)	3/4 (75.0%)	1/4 (25.0%)	3/4 (75.0%)	
Total	19/41 (46.3%)	22/41 (53.7%)	27/41 (65.9%)	14/41 (34.1%)	<i>p</i> = 0.075
<b><i>Escherichia coli</i></b>					
≥ 10 <sup>5</sup> CFU/ml	81/87 (93.1%)	6/87 (6.9%)	84/87 (96.6%)	3/87 (3.4%)	<i>p</i> = 0.494
10 <sup>4</sup> ~ 10 <sup>5</sup> CFU/ml	2/11 (18.2%)	9/11 (81.8%)	9/11 (81.8%)	2/11 (18.2%)	<i>p</i> = 0.009
≤ 10 <sup>4</sup> CFU/ml	1/2 (50.0%)	1/2 (50.0%)	2/2 (100.0%)	0/2 (0.0%)	<i>p</i> = 1.000
Total	84/100 (84.0%)	16/100 (16.0%)	95/100 (95.0%)	5/100 (5.0%)	<i>p</i> = 0.011
<b><i>Enterococcus faecalis</i></b>					
≥ 10 <sup>5</sup> CFU/ml	5/8 (62.5%)	3/8 (37.5%)	6/8 (75.0%)	2/8 (25.0%)	
10 <sup>4</sup> ~ 10 <sup>5</sup> CFU/ml	2/5 (40.0%)	3/5 (60.0%)	3/5 (60.0%)	2/5 (40.0%)	
≤ 10 <sup>4</sup> CFU/ml	0/2 (0.0%)	2/2 (100.0%)	1/2 (50.0%)	1/2 (50.0%)	
Total	7/15 (46.7%)	8/15 (53.3%)	10/15 (66.7%)	5/15 (33.3%)	<i>p</i> = 0.462

ificantly from 90.5% to 33.3%, compared with that by short-term culture combined with MALDI-TOF MS, which only decreased from 93.2% to 80.0%, the difference was statistically significant (*p* = 0.027). When the colony number was lower than 1 × 10<sup>4</sup> CFU/ml, the number of positive samples was too low, and the statistical results were not reliable. Among all Gram-negative bacteria, *E. coli* was the most common: 81/87 cases (93.1%) were detected by the centrifugation-based MALDI-TOF MS when the colony count was ≥ 1 × 10<sup>5</sup> CFU/ml, but when the colony count was between 1 × 10<sup>4</sup> and 1 × 10<sup>5</sup> CFU/ml, the detection rate decreased sharply to 2/11 (18.2%). Although the detection rate of *E. coli* by short-term culture combined with MALDI-TOF MS was also affected by the decrease in the bacterial count, it was much less affected than the centrifugation-based MALDI-TOF MS, with the detection rate reduced from 96.6% to 81.8%. Therefore, it is necessary to pay attention to the possible false-negative specimens in clinical practice.

Although the detection rate of bacteria by short-term culture combined with MALDI-TOF MS was higher than that by centrifugation-based MALDI-TOF MS, its MALDI score was slightly lower. The reason

may be that even after 5 hours of short-term culture, the colony number was still low, and it was easy to scrape the medium agar when applied to the target plate, which leads to too few bacteria scraped or failure to scrape bacteria, ultimately affecting the identification results. In the experiment, we could find that the MALDI spectrogram of the centrifugation-based MALDI-TOF MS was cleaner than that of short-term culture combined with MALDI-TOF MS. Nevertheless, in the experiment, we found another interesting phenomenon: when too many colonies were collected, the MALDI score could not be improved, and it is directly affected the identification results. It is probably because the bacteria were so clustered that it was not easy to spread evenly on a coated target board. There was a specimen with apparent bacterial growth, but MALDI-TOF MS did not detect it. Under the microscope, we found the bacteria gathered together. After a second test, *Proteus mirabilis* was identified. A similar phenomenon had been observed in other Gram-negative bacteria, such as *K. pneumoniae*. Therefore, in the clinical work, the appropriate bacterial density and the uniform coating on the target plate could guarantee their effective identification by MALDI-TOF MS.

Short-term culture combined with MALDI-TOF MS can eliminate the interference of proteins in MALDI-TOF MS. After short-term culture, it had higher detection ability for specimens with fewer colonies. The centrifugation-based MALDI-TOF MS could directly identify pathogens in clinical urine specimens within 1 hour and showed good identification ability on urine samples when colony counts was more than  $1 \times 10^5$  CFU/ml. However, it was affected by colony counts. To reduce the influence of small bacterial numbers on the detection results and improve the identification efficiency, we will introduce urine flow cytometry in subsequent experiments. As a new urine tangible component analysis technology, it can accurately provide the bacterial counts in urine samples (Wang et al. 2013; Sun et al. 2020). We plan first to count the number of urine bacteria using the urine flow cytometer. Urine samples with colony count higher than  $1 \times 10^5$  CFU/ml will be identified using the centrifugation-based MALDI-TOF MS, and if less than  $1 \times 10^5$  CFU/ml by short-term culture combined with MALDI-TOF MS.

Although the two MALDI-TOF MS methods have many advantages in clinical urine bacteria identification, there are some limitations. First, during the identification of Gram-positive bacteria by short-term culture combined with MALDI-TOF MS, the bacterial growth was not significant even after 5 hours of culture. Second, due to their thick and highly anionic cell walls, even the formic acid was added in the process, the detection of Gram-positive bacteria by MALDI-TOF MS was not optimal. It is necessary to improve the identification of Gram-positive bacteria. We tried to mix the specimens with formic acid and centrifuged at high speed; however, this did not work. Third, the study did not include antibiotic sensitivity tests and could not distinguish among antibiotic-resistant strains. Oviaño successfully used MALDI-TOF MS to rapidly identify the carbapenemase-producing *Enterobacteriaceae* in urine samples and detect their drug resistance (Oviaño et al. 2017). We will try to co-culture urine sediments and drugs on MALDI steel plates for short periods and rapidly detect the antimicrobial resistance of bacteria based on the mass spectrum peak change.

## Conclusions

MALDI-TOF MS can accelerate the bacteriological identification of organisms causing UTIs. The centrifugation-based MALDI-TOF MS warrants fast identification. However, it is greatly affected by the number of bacterial colonies, while short-term culture combined with MALDI-TOF MS has a higher detection rate but a relatively slow identification speed. The screening the urine colony count first, then assigning to the two

MALDI-TOF MS methods for identification, may be an effective and reliable alternative to the traditional urine culture, and it has great potential in measuring antibiotic susceptibility (Bizzini et al. 2011; Croxatto et al. 2012; Oviaño et al. 2017).

## Conflict of interest

The authors do not report any financial or personal connections with other persons or organizations, which might negatively affect the contents of this publication and/or claim authorship rights to this publication.

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