# RESEARCH ARTICLE

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# Consistency analysis of the Sysmex UF-5000 and Atellica UAS 800 urine sedimentation analyzers

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# Abstract

**Objective:** To evaluate the consistency between the results of Sysmex UF-5000 system and Atellica® UAS 800 Urine Sediment Analyzer.

**Methods:** A total of 636 random urine samples were collected from inpatients and outpatients from March to September 2021. Urine was collected for analysis by the Sysmex UF-5000, Atellica UAS 800 systems, and manual microscopic examination. The results of manual microscopy as the gold standard, the coincidence rate and false-negative rate of Sysmex UF-5000 and Atellica UAS 800 systems in the detection of red blood cells, white blood cells, and casts were calculated.

**Results:** The coincidence rates of red blood cells, white blood cells, and cast, crystals, and other sediment components for the Sysmex UF-5000 system were 85.37%, 87.89%, 91.67%, 88.36%, and 71.86%. The false-negative rates were 28.47%, 3.75%, 68.97%, 37.25%, and 30.63%. The coincidence rates of red blood cells, white blood cells, and cast, crystals, and other sediment components for the Atellica UAS 800 system were 85.06%, 90.25%, 59.12%, 91.67%, and 67.45% and the false-negative rates were 60.42%, 21.25%, 36.21%, 19.64%, and 35.80%.

**Conclusion:** Two instruments are superior in the detection of red blood cells and white blood cells. The Atellica UAS 800 system with image review has a good coincidence rate in the identification of crystals and casts. The identification of various sediment components in urine by both instruments meets the laboratory requirements. Two instruments with different methodologies have their own characteristics, and we should reasonably use them according to the conditions of the laboratory.

# **KEYWORDS**

Atellica UAS 800, consistency, Sysmex UF-5000, urine sediment analyzer

# 1 | INTRODUCTION

The detection of sediment components in urine is an important indicator for the diagnosis, treatment, and monitoring of kidney

diseases, urinary system diseases,<sup>1</sup> and systemic diseases, and an important basis for diagnosis and differential diagnosis. With the clinical application of automated urine sediment analyzers, the problem of standardizing urinary sediment detection has been solved and

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workflow efficiency has been greatly improved. However, manual microscopic examination is still the gold standard for urine examination.<sup>2</sup> In this experiment, the results of two automated urine analyzers, the Atellica UAS 800 Urine Sediment Analyzer (digital imaging method) and Sysmex UF-5000 system (flow cytometry), were compared with those of manual urine sediment microscopic examination to verify the performance and characteristics of these instruments employing different methodologies, so as to provide a more reliable detection method for clinical application.

# 2 | MATERIALS AND METHODS

# 2.1 | General information

Source of specimens: 636 urine specimens randomly selected from inpatients and outpatients in the West China Second University Hospital from September to December 2021 were collected, and the middle 30ml of clean urine was retained in a sterile urine tube. The collection of each urine sample was conducted in strict accordance with standard operating procedures. After the urine was mixed, 10 ml of urine was collected into a graduated tube for analysis by the Sysmex UF-5000 system, Atellica UAS 800 system, and manual microscope examination. All samples were tested within 2 h.

Instruments and reagents: Atellica UAS 800 Urine Sediment Analyzer and reagents from Siemens Co., Ltd. with Bio-Rad controls (positive control lot no. 67222 and negative control lot no. 67221) and calibrators, etc.; Sysmex UF-5000 automated analyzer and reagents, controls (positive control lot no. UK0107 and negative control lot no. UK0107), and calibrators (lot no. UA0089), etc.; LEICA DM500 optical microscope, glass slides, cover glasses; KDC-2046 low-speed refrigerated centrifuge and urine sediment detection centrifuge tubes; sterile urine cups, disposable urine sediment counting plates; disposable pipettes, etc.

# 2.2 | Methods

# 2.2.1 | Detection methods of the Sysmex UF-5000 system and Atellica UAS 800 Urine Sediment Analyzer

The Sysmex UF-5000 system uses the flow cytometry method employing a semiconductor laser. By detecting the forward-scattered light, side-scattered light, and side fluorescence signals generated by the sediment components of urine, the type and quantity of sediments can be measured.

The conversion relationship between the flow cytometry method and microscopic examination according to the manufacturer's instructions is:

 $0.18 \, \text{p} / \mu \text{L} = 1 \, \text{p} / \text{HPF}$ 

 $2.9 \, \text{p} / \mu \text{L} = 1 \, \text{p} / \text{LPF}$ 

The Atellica UAS 800 analyzer uses a digital image-based system, which provides a complete field of view, similar to the manual microscope. The conversion relationship between the sediment image of the digital imaging method and microscopic examination according to the manufacturer's instructions is:

6.6 p / image = 
$$1p / \mu l$$
  
4.4 p /  $\mu L$  =  $1p / HPF$   
10.7 p /  $\mu L$  =  $1p / LPF$ 

Prior to this experiment, instrument performance, including within-lot precision, between-lot precision, linearity range, coincidence rate, false-negative rate, and carryover rate, was verified according to CLSI guidelines.<sup>3-5</sup> In this experiment, manual microscopic examination was used as the gold standard to compare the coincidence rate and false-negative results of 636 urine samples.

# 2.2.2 | Manual microscope examination

According to CLSI guidelines GP16-A3,<sup>6</sup> standard KOVA cell counting plates were used for manual microscopy. The volume of the large grid of the counting plate is 0.9  $\mu$ l (3 mm \* 3 mm \* 0.1 mm), each large grid is composed of 9 middle grids with a volume of 0.1  $\mu$ l, and each middle grid is composed of 9 small grids with a volume of  $0.0111 \,\mu$ l. 10ml of each portion of the mixed urine was centrifuged for 5 min under a centrifugal force of 400g. After 9.8 ml of the supernatant was absorbed through a tube, the remaining 0.2 ml of urine sediment was fully mixed, and 20µl was removed using a 1 ml pipette and dropped onto the counting plate. The plate stood for 5 min after full expansion. We observed the plate with  $10 \times 10$  power, counted each cell component in 10 large grids with 10×40 power, and recorded the results. All the samples were tested within 2 h. The particle counting result was  $p/\mu I = n/(N^*50)$  \*90 (n = number of cell component particles, N = sum of small grids, 50 = times) concentrated from 10-0.2 ml, 90 = 1/(0.0111\*1). Microscopic examination was performed twice by two experienced technicians using the double-blind method. The positive criteria were red blood cells >3/HPF, white blood cells >5/HPF, and casts >1/LPF. The consistency of microscopic examination results of the two groups was evaluated by Kappa test in SPSS 25.0 statistical software. When the consistency is high, the mean value of the microscopic examination results of the two groups can be taken as the judgment standard.

According to the instructions for the KOVA cell counting plate, the conversion relationship between microscopic examination and the KOVA cell counting plate is:

 $p / HPF = 1.6^* p / \mu L(KOVA)$  $p / LPF = 5.76^* p / \mu L(KOVA)$ 

# 2.3 | Statistical analysis

All test data were processed qualitatively and analyzed statistically, and manual microscopic examination was used as the gold standard. Positive criteria were RBC >3 p/HPF, WBC >5 p/HPF, and cast >1 p/LPF.<sup>7,8</sup> Therefore, the positive criteria after Sysmex UF-5000 system conversion were RBC >0.54 p/µl, WBC >0.9 p/µl, and cast >2.9 p/µl. The positive criteria after Atellica UAS 800 analyzer conversion were RBC >13.2 p/µl, WBC >22 p/µl, and cast >10.7 p/µl. The qualitative results of the sediment components in the selected urine samples were compared by the coincidence rate and the false-negative rate. Coincidence rate =  $100\% \times [(a+d)/n]$ , false-negative rate =  $100\% \times [c/(a+c)]$ . Kappa value was calculated by Kappa test in SPSS 25.0 statistical software for the evaluation of the consistency of the results of manual microscopy. Kappa scale values ranges were 0.41–0.60, moderate consistency; 0.61–0.80, high consistency; and 0.81–1, complete consistency.

# 3 | RESULTS

# 3.1 | Evaluation of manual microscope examination technology

Kappa values for red blood cells, white blood cells, and casts in 636 samples examined by the two experienced technicians were calculated to be 0.72, 0.78, and 0.54, respectively. The results showed that the red blood cell microscopic examination results were highly consistent, the white blood cell microscopic examination results were highly consistent, and the cast microscopic examination results were moderately consistent. The mean value of the two microscopic examination results could be taken as the judgment standard.

# 3.2 | Result comparison of the Sysmex UF-5000 and Atellica UAS 800 urine sediment analyzers

## 3.2.1 | Coincidence rates

The performance verification of each instrument met the standard. Compared with the microscopic examination results, the coincidence rates for red blood cells, white blood cells, casts, crystals, and other sediment components for the Sysmex UF-5000 system were 85.37%, 87.89%, 91.67%, 88.36%, and 71.86%, respectively. The coincidence rates for red blood cells, white blood cells, casts, crystals, and other sediment components for the Atellica UAS 800 system were 85.06%, 90.25%, 59.12%, 91.67%, and 67.45%, respectively. The two instruments met the requirements of the CLSI guide-lines. See Table 1.

# 3.2.2 | False-negative rates

The false-negative rates for red blood cells, white blood cells, casts, crystals, and other sediment components for the Sysmex UF-5000 system were 28.47%, 3.75%, 68.97%, 37.25%, and 30.63%, respectively, compared to the microscopic examination results. The false-negative rates for red blood cells, white blood cells, casts, crystals, and other sediment components for the Atellica UAS 800 system were 60.42%, 21.25%, 36.21%, 19.64%, and 35.80%, respectively. The two instruments met the requirements of the CLSI guidelines.

# 3.2.3 | Detection rates for pathological casts

Six hundred thirty-sixth fresh urine samples with positive pathological casts by microscopic examination were analyzed by the two automated analyzers. The Sysmex UF-5000 system's detection rates for red blood cell, white blood cell, granular, tubular, waxy, mixed, and fat casts were 11.11%, 0%, 28.57%, 0%, 50%, 46.15%, and 25%, respectively. The Atellica UAS 800 system's detection rates for erythrocyte, leukocyte, granular, renal tubular, waxy, mixed, and fat casts were 77.77%, 100%, 71.42%, 0%, 8.93%, 46.15%, and 25%, respectively. See Table 2.

# 3.2.4 | Detection rates for urine sediment

The detection rates for epithelial cells, crystals, yeast-like fungus spores, sperm, and mucus for the Sysmex UF-5000 system were 90.74%, 36.78%, 37.5%, 0%, and 91.75%, respectively. The detection

TABLE 1	Comparison of the S	ysmex UF-5000 and Atellica	UAS 800 system results	s with microscop	oic examination (	n = 636
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		Sysmex UF-5000 system		Atellica UAS 800 system	
		Negative	Positive	Negative	Positive
Microscopic red blood cells	Negative	440	52	484	8
	Positive	41	103	87	57
Microscopic white blood cells	Negative	405	71	484	28
	Positive	6	154	34	126
Microscopic casts	Negative	565	13	339	239
	Positive	40	18	21	37
Microscopic crystals	Negative	530	19	538	11
	Positive	55	32	42	45
Other sediment components <sup>a</sup>	Negative	235	98	212	121
	Positives	81	222	86	217

<sup>a</sup>Other sediment components include epithelium, yeast-like bacteria spores, sperm, and mucus.

TABLE 2 Detection rates for pathological casts by the Sysmex UF-5000 system, Atellica UAS 800 system, and microscopy

	Pathological casts							
	Red blood cell casts ( $n = 9$ )	Leukocyte casts (n = 2)	Granular casts (n = 21)	Tubular casts (n = 1)	Waxy casts (n = 8)	Mixed casts (n = 13)	Fat casts (n = 4)	Total (n = 58)
Sysmex UF-5000 system	1	0	6	0	4	6	1	18
Atellica UAS 800 system	7	2	15	0	6	6	1	37

*Note: n* is the number of positive cases of pathological casts detected by manual microscope examination.

TABLE 3 Detection rates for urine sediment for the Sysmex UF-5000 system, Atellica UAS 800 system, and microscope examination

	Urine sediment						
	Epithelial cells (n = 108)	Crystals (n = 87)	Yeast-like spores (n = 8)	Sperm ( <i>n</i> = 3)	Mucus (n = 97)	Total (n = 303)	
Sysmex UF-5000 system	98	32	3	0	89	222	
Atellica UAS 800 system	89	45	5	1	77	217	

*Note: n* is the number of positive cases detected by the manual microscope examination.

TABLE 4 Detection rates for crystals for the Sysmex UF-5000 system, Atellica UAS 800 system, and microscopic examination

	Crystals					
	Triphosphate crystals (n = 55)	Uric acid crystals (n = 15)	Calcium oxalate crystals (n = 16)	Other crystals (n = 1)	Total (n = 87)	
Sysmex UF-5000 system	16	4	12	0	32	
Atellica UAS 800 system	27	10	8	0	45	

*Note*: *n* is the number of positive crystals detected by manual microscope examination. Other crystals include cholesterol, leucine, and cystine crystals.

rates for epithelial cells, crystals, yeast-like bacteria spores, sperm, and mucus for the Atellica UAS 800 system were 82.4%, 51.72%, 62.5%, 33.33%, and 79.38%, respectively. See Table 3.

#### 3.2.5 | Detection rates for crystals

The detection rates for triphosphate, uric acid, calcium oxalate, and other crystals for the Sysmex UF-5000 system were 29.09%, 26.66%, 75%, and 0%, respectively. The detection rates for the Atellica UAS 800 system for triphosphate, uric acid, calcium oxalate, and other crystals were 49.09%, 66.66%, 50%, and 0%, respectively. See Table 4.

# 3.3 | Comparison of sediment components

The two instruments employ different detection principles and report different sediment components. Types of sediment detected by the Sysmex UF-5000 system and Atellica UAS 800 analyzer are shown in Table 5.

# 4 | DISCUSSION

The Sysmex UF-5000 system employs flow cytometry using a blue semiconductor laser as its detection principle.<sup>9-11</sup> The stained sample is introduced into a FLOWCELL flow chamber to form a sheath flow. A laser beam detects the forward-scattered light, side-scattered light, and side fluorescence signals generated by the flow of urine components. The detected signal is transmitted to a wave-form processing unit that extracts waveform height, pulse width, and other parameters. Based on the parameters, a scattergram is calculated that classifies the urine components according to where they were generated.

In contrast, the Atellica UAS 800 system uses a digital imaging method as its basic detection principle.<sup>11,12</sup> The urine sample, wrapped in a sheath fluid, flows through a digital camera that takes

		Atellica UAS 800		Sysmex UF-5000		
	Type of Sediment	Quantitative parameter	Qualitative parameters	Quantitative parameter	Qualitative parameters	
RBC	Red blood cells	$\checkmark$		$\checkmark$		
	Homogeneous RBC		$\checkmark$		$\checkmark$	
	Heterogeneous RBC		$\checkmark$		$\checkmark$	
	Hybrid RBC				$\checkmark$	
WBC	White blood cells	$\checkmark$		$\checkmark$		
WBCc	Pus cells		$\checkmark$	$\checkmark$		
NEC	Non-squamous epithelial cells	$\checkmark$		$\checkmark$		
	Renal tubular epithelial cells			$\checkmark$		
	Transitional epithelial cells			$\checkmark$		
	Superficial transitional epithelial cells					
	Bottom transitional epithelial cells					
EPI	Squamous epithelial cells	$\checkmark$		$\checkmark$		
CRY	Crystals	$\checkmark$		$\checkmark$		
	Calcium oxalate crystals	$\checkmark$				
	Uric acid crystals	$\checkmark$				
	Cholesterol crystals	$\checkmark$				
	Triphosphate crystals	$\checkmark$				
	Calcium phosphate crystals	$\checkmark$				
	Cystine crystals	$\checkmark$				
	Leucine crystals	$\checkmark$				
	Tyrosine crystals	$\checkmark$				
HYA	Transparent casts	$\checkmark$		$\checkmark$		
PAT	Pathological casts		$\checkmark$	$\checkmark$		
	Granular casts		$\checkmark$			
	Tubular casts		$\checkmark$			
	Red blood cell casts		$\checkmark$			
	Leucocyte casts		$\checkmark$			
	Fat casts		$\checkmark$			
	Waxy casts		$\checkmark$			
	Mixed casts		$\checkmark$			
YEA	Yeast		$\checkmark$	$\checkmark$		
BAC	Germ	$\checkmark$		$\checkmark$		
	Bacillus	$\checkmark$				
	Coccus	$\checkmark$				
	Gram-negative bacteria				$\checkmark$	
	Gram-positive bacteria				$\checkmark$	
MUC	Mucus		$\checkmark$	$\checkmark$		
SPRM	Sperm		$\checkmark$	$\checkmark$		
TRV	Trichomonad		$\checkmark$			

digital images of the sample. To obtain the images, urine flows through counting plates of various specifications and precipitates after centrifugation, or it is maintained in a static state without centrifugation. Finally, urine components are classified and counted by the instrument's image recognition software. In this study, we evaluated the diagnostic performance of the Sysmex UF-5000 and Atellica UAS 800 systems. The red blood cell coincidence rates were 85.37% and 85.06%, respectively, but there was still some recognition bias. The reasons for these deviations may be related to common issues with manual microscopy,<sup>11.13</sup> such

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as inaccurate counting, problems in sample processing, and the destruction and loss of red blood cells during centrifugation. The laboratory can minimize these deviations by establishing standardized manual procedures.<sup>14</sup> In addition, errors can occur in red blood cell recognition caused by the presence of deformed, ghost, or budding red blood cells or confusion in the classification of calcium oxalate crystals and yeast-like fungus spores.<sup>10,11</sup> Other possible error sources include differences in identification between digital imaging and manual microscopy,<sup>15</sup> the difference between manually counting only 9 cells versus counting all the cells, and errors in conversion between manually counted particles/µl and instrument-derived particles/HPF. Compared with red blood cells and white blood cells, cast identification is more difficult. The coincidence rates for the Atellica UAS 800 system for granular, white blood cell, red blood cell, and waxy casts are 71.42%, 100%, 77.77%, and 8.93%, respectively.

As a specialized hospital for women and children, the urine samples of pregnant women account for about half of all urine samples. The anatomical structure and physiological changes of pregnant women caused by pregnancy complicate the composition of urine samples. Increased amounts of mucus, clustered pus cells, bacteria, and vaginal secretions easily interfere with the casts. The image analysis employed by the Atellica UAS 800 analyzer improves the sensitivity of cast identification, but Sysmex UF-5000 system does not have this function. Although the sensitivity of the Sysmex UF-5000 system to cast is lower than that of the Atellica UAS 800 system, the cutoff value of the Sysmex UF-5000 system for detection of RBC, WBC, and cast can be established through the receiver operating characteristic curve (ROC) analysis to evaluate sensitivity, specificity, coincidence rate, and other performance characteristics. The laboratory's own judgment threshold can be established to improve the sensitivity of detection.<sup>16</sup>

In addition to red blood cells, white blood cells, and casts, we evaluated the consistency of qualitative results for the two urine sediment analyzers, including crystals, yeast-like spores, sperm, and mucus, compared to results from manual microscopic examinations. Although these deposits are less clinically important than erythrocytes, leukocytes, and casts, they are still valuable in a number of specialized areas. For example, kidney calculi are relatively common for adults and have relatively complete diagnosis and treatment guidelines. The incidence rate of kidney calculi in children is lower than in adults but has been increasing in recent years.<sup>17</sup> Because children are still in the growth and development stage, their physiology and anatomy are different from those of adults. For example, the presence of calcium oxalate and calcium carbonate crystals, which is often ignored in adults, is a high-risk factor for childrens' kidney calculi.

The sensitivity of the two urine sediment analyzers to each type of crystal was different, and the Sysmex UF-5000 system's detection rate for crystals was relatively low. Calcium oxalate crystal detection was highest at 75%, and uric acid crystal detection was lowest at 26.6%. This might be related to the fact that some of the crystals were dissolved in advance by the reagent used on the Sysmex UF-5000 system. In contrast, the Atellica UAS 800 system showed high sensitivity for crystals. At present, domestic and foreign laboratories do not use a single method (urine sediment analyzer) for urine detection, but usually employ a urine sediment analyzer together with a urine dry chemistry analyzer for joint detection. Therefore, when a flag message appears on the urine sediment analyzer, technicians must combine the results of the urine dry chemistry analyzer and/or the clinical history of the patient to comprehensively judge whether manual microscopic examination is required.

For medical labs that need to judge clinical efficacy based on the types of casts and crystals in urine, having an analyzer with high sensitivity to casts and crystals should be an advantage. However, it also requires technicians to be more skilled in image recognition or manual microscopy to filter out false-positive samples and confirm true-positive samples.

In this study, the Atellica UAS 800 system also identified a case of trichomonas vaginalis. The sample was obtained in our hospital's Department of Reproductive Andrology from a mawith azoospermia. This result was greatly significant for clinical diagnosis and treatment, and it reminds us that, in addition to excrement and blood samples,<sup>18</sup> the examination of parasites in urine samples should not be ignored.<sup>19,20</sup>

Urine culture is the gold standard for the diagnosis of urinary tract infection. Although the analysis of bacterial parameters was not included in this study, the flow cytometry technology employed by the Sysmex UF-5000 system was able to perform fluorescence staining on bacterial nucleic acids, and it could distinguish gram-positive and gram-negative bacteria, with better performance than that of the Atellica UAS 800 system. The urine culture cycle is long. If a urine sediment analyzer can provide faster diagnostic information, it can not only reduce patients' treatment costs, but also avoid the abuse of clinical antibiotics to a certain extent.

In summary, the two urine sediment analyzers are both excellent for red blood cells and white blood cells and can replace manual microscopy for these applications. The digital imaging technology of the Atellica UAS 800 system showed high sensitivity to casts and crystals but also produced a high false-positive rate. When using the Sysmex UF-5000 system, the lab should set its own cutoff values for the detection of various particle components. By understanding the different characteristics of these urine sediment analyzers and optimizing the laboratory workflow accordingly, labs can better serve the needs of clinical testing.

#### AUTHOR CONTRIBUTIONS

HL performed the statistical analysis and prepared the manuscript. QL and YDZ collected the data and prepared the manuscript. FY and DYH were responsible for the study design and coordination, guided the statistical analysis, and revised the manuscript critically. All authors reviewed and edited the manuscript and approved the final version of the manuscript.

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# CONFLICT OF INTEREST

Hai Liu, Qing Li, Yiduo Zhang, Dongyue Huang, and Fan Yu declare that they have no conflict of interest.

## DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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