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Article

Comparative study on the accuracy of SysmexUF5000 and UF1000i automatic urine sediment analyzer for urine specimen detection

Chunhua Luo*, Yucheng Luo, Qianyuan Li, Jun Luo, Peng Wang

Department of Clinical Laboratory, Yichang Central People's Hospital, 183 Yiling Avenue, Yichang 443003, China
*** Corresponding author:** Chunhua Luo, 18292050142@stu.suse.edu.cn

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Abstract: Background: Urinalysis includes the physical examination of urine and the examination of formation scores, and the microscopic evaluation of the visible components in urine is a time-consuming process with high labor intensity and requires solid morphological expertise. **Objective:** To assess the Sysmex UF5000 automatic urine sediment analyzer's diagnostic capability and contrast its accuracy with UF1000i urine samples. **Methods:** The precision, contamination rate, linear range, and reportable range of UF5000 were evaluated according to relevant regulations and verification methods provided by manufacturers. Urine samples were detected by UF5000 and UF1000 sediment analyzers, respectively. The accuracy of the false positive rate, false negative rate, sensitivity, and specificity of urine samples detected by the two instruments were compared using the results of centrifugal microscopy as the gold standard. **Results:** Red blood cells (RBC), White blood cells (WBC), and Epithelial cells (m) were detected by the Sysmex UF5000 urinary sediment analyzer. The intra-batch precision and inter-batch precision of EC), tubular type (CAST) and bacteria (BACT) were all within the required range. The contamination rate and linear range of RBC, WBC, and BACT met the requirements and could be reported widely. The false positive rate of UF5000 RBC was significantly lower than UF1000i, while the false positive rate of CAST was higher than UF1000i, the difference was statistically significant. In UF5000, the false negative rate of MALE WBC was significantly lower than that of females, and the difference was statistically significant. There was no difference in the specificity of white blood cells detected by the two instruments. When compared to UF1000i, UF5000 had much higher specificity for erythrocyte and tube types, and the sensitivity of three commonly used parameters of UF5000 was significantly higher than that of UF1000i, with statistical significance ($P < 0.05$). **Conclusion: Compared** with the UF1000i, the UF5000 showed similar or better diagnostic performance on most parameters.

Keywords: automatic urine sediment analyzer; SysmexUF5000; UF1000i; urine specimen

1. Background

One of the three standard laboratory tests, after blood counts and biochemical analyses, is urinalysis, which is used for screening, diagnosis, and monitoring of urinary tract diseases [1]. Urinalysis includes the physical examination of urine and the examination of formation scores, and the microscopic evaluation of the visible components in urine is a time-consuming process with high labor intensity and requires solid morphological expertise. In addition, due to the existence of subjectivity, there are certain differences between observers, with high imprecision and inaccuracy [2]. The automatic urine sediment analyzer has overcome the above limitations and ameliorated the speed and consistency of urine detection. Sysmex UF5000 Urinary Sediment Analyzer (hereinafter referred to as UF5000) has been

gradually popularized and applied in clinical laboratories. The detection principle and SysmexUF1000i (hereinafter referred to as UF1000i) are both flow cytometry methods. Through the red semiconductor laser beam irradiation, nucleic acid fluorescence staining, and the sheath flow sample formed in the sheath flow analysis cell, the photoelectric signal converted from the pre-scattered light, side-scattered light and side-fluorescence signal generated from each particle is analyzed, and the identification particle is determined. UF1000i urine sediment analyzer can ameliorate the speed of urine detection and provide the basis for the diagnosis of renal diseases. The method is fast and convenient, but it is also easily affected by many factors. UF5000 has been greatly ameliorated in the channel for detecting the visible components of urine, which is combined with cytochemical staining technology. By examining forward scattered light, lateral scattered light, lateral fluorescence, and depolarized lateral scattered light, the visible components of urine were discovered and categorized [3–5].

The major test for diagnosis that is frequently executed by clinical laboratories is the routine urinalysis. It is essential for the diagnosis and assessment of illnesses of the urinary system, the extra treatment of metabolic diseases, and the assessment of the side effects of medication therapy [6]. Urine dry chemical analysis findings are frequently utilized by therapists to predict the possibility of an infection in the urinary tract. Whereas it is related to UTI screening, Sysmex technologies devices like the UF5000 and UF1000i have demonstrated superior performance [7]. The identical technology like UF5000 and UF1000i, fluorescent flow cytometry is the basis for UF5000. The fluorescent source has been enhanced, and the detecting channel and chemicals have received significant modifications [8]. Improved technologies in computerized urine particle analysts have decreased the quantity of microscopic testing, improved processing times, enhanced accuracy and dependability, and decreased expenses [9]. Suspected UTIs, stones in the kidneys, infected and non-infectious renal disorders, pregnancy, diabetes mellitus, acidosis, or alkalosis are among the most prevalent illnesses that require a urinalysis. Several laboratories initially employed standard, non-standardized urine sediment analysis [10]. With the lack of qualified staff or an excessive quantity of samples, manual examination of urine particles is difficult and costly. Urine analysis can be performed more rapidly and easily considering the growing use of computerization in labs [11]. As the symptoms, including a high body temperature, vomiting, nausea, and lack of sleep, are non-specific and frequently overlap with symptoms of other infections, diagnosing a UTI can be difficult, and quantitative urine culture is still considered as the “gold standard” is an essential component of laboratory diagnostics [12]. Rarely occurring with lesser frequency than upper respiratory tract infections, UTIs are among the most prevalent bacterial illnesses. The most accurate method for diagnosis the UTI is the quantitative cultures of urine samples and the process requires 18 to 24 hours to complete [13]. Reproductive or urological issues are frequently characterized by microscopic hematuria. Glomerular hematuria (GH) or non-glomerular hematuria (NGH) is the two categories into that it occurs. A technique that consistently separates GH from NGH is very valuable for the first triage of patients with microscopic hematuria, as it enables clinical treatment and care more efficient and effective [14]. This study evaluates the main performance of UF5000 according to relevant

regulations and verification methods provided by manufacturers. Furthermore, fresh urine samples for daily examination were selected and tested on UF5000 and UF1000i at the same time. Microscopic examination results were used as the gold standard to compare the accuracy of urine samples. The reports are as follows.

2. Related works

In the diagnosis [15] of many diseases, the routine urine test is a common biochemical test in clinical practice. Based on the change in urine composition and content caused by diseases is of great significance for the initial judgment of diseases, subsequent accurate diagnosis and treatment, and assessment of disease outcome. Therefore, the accuracy and reliability of urine routine examinations are required to be higher and higher in the clinic. Routine urine examination involves many examination items, mainly including leukocyte examination, and red blood cell examination. Article [16] utilized different examination techniques and methods to clarify the level of various indexes and provide the basis for disease judgment. Microscopic detection is a traditional method for routine urine examination provided in the study [17], which has high accuracy in the detection of salt crystal quantity, morphology, urine sediment cast, and cells in urine samples. The development of modern medical technology, not only promotes the application of more testing instruments and reagents in urine routine examination but also promotes the diversified development of urine routine examination methods. Under such circumstances, how to better adapt to the quality of clinical urine routine examination in the future has become a problem that cannot be ignored in the medical field.

Recently, research [18] presented the automatic analyzer of urine sediment, which has gradually been applied in clinical examination, which provides more perfect method support for routine urine examination. Different urine routine detection techniques have their advantages and disadvantages, and there are also some differences in the test results. For example, urine RBC and WBC are the test items of the automatic urine sediment analyzer, but they are easily confused with cells such as casts in the detection application and form interference, resulting in different degrees of false positive and false negative results, which must be confirmed by manual microscopic examination to ensure the authenticity and reliability of the results. Therefore, comparing the application effects of different testing techniques can provide a guarantee for the selection and application of high-quality and high-efficiency urine routine testing techniques. The precision, carrying contamination rate, linear range, and reportable range of UF5000 were evaluated in our laboratory by using original quality control materials and fresh urine samples tested routinely in clinical practice [19]. The experimental results show that the intra-batch precision and inter-batch precision of RBC, WBC, EC, CAST, and BACT, as well as the carrying pollution rate and linear range of RBC, WBC, and BACT in the UF5000 test items meet the standards provided by the manufacturer, which indicates that the UF5000 instrument has good test performance, accurate and reliable results, and the reportable range of RBC, WBC and BACT is wide, which can satisfy the demand of routine testing work.

In this experiment [20], they selected fresh urine samples for daily examination

and studied the accuracy of UF5000 in detecting RBC, WBC, and CAST. The results showed that the false negative rate of UF5000 in urine samples met the relevant requirements. The urine samples were tested simultaneously on UF1000i, and the accuracy of RBC, WBC, and CAST was compared. The outcomes of the analysis demonstrated the false positive rate of RBC in UF5000 was lower than that in UF1000i, and the false positive rate in CAST was higher than that in UF1000i, the difference was statistically significant in the study [21]. It can be seen that although the false positive of CAST in UF5000 is slightly higher, UF5000 and UF1000i still have similar detection performance in detecting WBC and CAST in the visible components of urine. Since EH-2090 has not been the subject of any research as of yet, we compared it to manual microscopy and assessed its analytical and clinical capabilities to ensure it can handle routine clinical tasks [22]. However, UF5000 is superior to UF1000i in RBC detection performance, especially the false positive rate. Authors of [23] determined the presence of a small number of morphologically altered red blood cells in a large number of samples or the misclassification of components with similar volume and chromaticity, such as yeast or crystals, or the high density of the sample causing changes in the shape and volume of red blood cells, resulting in false positive results. The new SF channel of UF5000 [24] can combine the pulse width of scattered light, the waveform area of the scattered light signal, and the waveform area of the fluorescence signal to detect RBC in urine, and the depolarized lateral scattered light can capture birefringent crystals, separate red blood cells and crystals with higher accuracy, and reduce the interference of crystals on red blood cells, thus reducing the false positive rate of RBC detection. This makes the performance of UF5000 in detecting RBC more excellent [25]. In the study [26], it is believed that lymphuria will occur in the urine, or the subject will ingest a large amount of antibacterial drugs, and the urine and urine sediment will be affected by the environment of high concentration of antibiotics, resulting in inactivation of leukocyte specific enzymes, resulting in false negative results. The study [27] determined the accuracy of WBC, CAST, and UF1000i has not been significantly ameliorated, and the false-negative rate of WBC, CAST, and UF1000i is in line with relevant approval regulations, indicating that the performance and accuracy of UF5000 in detecting WBC and CAST are also in line with the standard requirements when comparing with microscopic examination results. The analysis [28] showed that the false-negative rate of WBC in men in UF5000 was significantly lower than that in women, which can be related to the difference in sexual physiology between men and women. UF5000 meets the relevant approved requirements in the performance of the instrument and the comparison with the microscopic examination results. There was no difference in the specificity of leukocytes detected by the two instruments in the article [29]. The specificity of RBC and casts of UF5000 has improved than the UF1000i, and the sensitivity of the three commonly used parameters of UF5000 is most effective compared with UF1000i. statistically significant variations in the research [30], suggested that when patients were carrying out routine urine examinations, the urine count level of the automatic urine sediment analyzer was selected for detection, which was more accurate and could also provide a reliable basis for making a reasonable and effective treatment plan. According to the analysis [31], UF5000 uses its built-in chip to detect the cell content in urine samples, generates relevant data according to some specific indicators,

uses the signal waveform information of tangible components, and then uses the light splitting system to process the difference of light to promote the light source point to emit a spot beam energy super excellent cup, and then selects the characteristics that can match some components in blood to check the wavelength, and then obtains the corresponding substance content.

3. Material and methods

3.1. General material

From May 2021 to October 2021, urine samples from outpatient, inpatient, and health check-ups were collected randomly. The selected sample covered the linear range of all parameters as far as possible, with high concentration and normal urine samples for performance evaluation of the instrument and the remaining samples for accuracy comparison of UF5000 and UF1000i. There were 103 males and 86 females, aged 18–74 years, with a mean age of (51.7 ± 6.7) years.

Inclusion criteria: (1) Patients who have undergone routine urine examination; (2) With no infectious diseases; (3) Aged ≥ 18 years; (4) Patients were told about the trial and gave their consent.

Exclusion criteria: (1) patients who suffer from significant organ dysfunction; (2) With malignant tumor disease; (3) With mental illness and cognitive impairment.

3.2. Instruments and reagents

UF5000 and UF1000i are both provided by Sysmex Company of Japan. The instrument reagents, related quality control products, and calibrators used for testing are all matched by the original factory. All reagents are within the validity period. Both instruments have been regularly calibrated, and quality control tests have been carried out before daily specimen testing, and the results meet the manufacturer's requirements. The centrifuge is a TD4N low-speed urine centrifuge of Shanghai Lu Xiangyi Centrifugal Instrument Co., Ltd. The microscope adopts a CX23 (upgraded version) binocular microscope produced by Olympus Optical Industry Co., Ltd.

3.3 Methods

3.3.1. Performance evaluation of UF5000

The performance of the instrument was evaluated in terms of precision, carrying contamination rate, linear range, and reportable range according to the CNASCL02-A002:2018 Instructions for the Application of Quality and Competence Accreditation Criteria for Medical Laboratories in the Field of Humoral Examination.

Precision

According to the requirements of CLSI(EP15-A2), the quality control products with high and low concentrations of the original factory are used and then mixed according to the instructions for determination. The quality control products with each concentration level are continuously measured 3 times a day for a total of 5 days. Intra- and inter-assay precision was calculated according to the CLSI(EP15-A2) formula for the counts of Red blood cells (RBC), white White blood cells (WBC), epithelial cells (EC), casts (CAST), and bacteria (BACT). Intra-batch precision should meet the

following requirements: erythrocyte ≤ 10.00%, leukocyte ≤ 10.00%, EC ≤ 30.00%, CAST ≤ 40.00%, BACT ≤ 20.00%, and inter-batch precision should be the same in-batch precision.

Carrying contamination rate

According to the standard provided by the manufacturer, this experiment only does the carrying contamination rate of RBC, WBC, and BACT. High-concentration urine samples (about 10000/μL) were mixed and measured three times, with values of H1, H2, and H3 respectively. Then concentration urine samples (< 10/μL) were taken three times, with values of L1, L2, and L3. The rate of carrying contamination was calculated. Carrying contamination rate=| L1-L3 |/(H3-L3) × 100%. The pollution rate required by the manufacturer shall meet the following requirements: RBC ≤ 0.10%, leukocyte ≤ 0.10%, BACT ≤ 0.05%.

Linear range

According to the standard provided by the manufacturer, this experiment only does the linear range of RBC, WBC, and BACT. Take the high-concentration urine sample (about 10000/μL) and use the normal urine sample supernatant as 1:2, 1:4, 1:16, 1:64, 1:256... series of equal specific dilutions, the original dilution result of the sample as the expected value, the test result obtained by the instrument after the sample dilution is the measured value, the expected value is the abscissa (x), and the measured value is the ordinate (y). A linear regression analysis was performed (equation: Y = aX + b) and the correlation coefficient (r) was calculated. The following requirements shall be met: a is within the range of 0.95~1.05, r ≥ 0.975.

Reportable range

Based on the linear range results, the theoretical and actual CV values of RBC, WBC, and BACT at each dilution ratio are calculated, and the reportable ranges are calculated according to the manufacturer's allowable CV values: RBC ≤ 10.00%, WBC ≤ 10.00%, BACT ≤ 20.00%. Reportable range = original multiple values × maximum dilution of allowable CV value.

3.3.2. Comparison of the performance of Uf5000 and Uf1000i

Specimen collection

Samples were collected according to standard operating procedures, and relevant precautions for routine urine examination were introduced to the subjects to ensure that they fully mastered urine collection skills. 10 ml of midstream urine was collected in a sterile leak-proof container in the morning, and properly stored for inspection, and the collected urine samples were divided into 3 equal parts and marked respectively, namely No. 1, No. 2, and No. 3, for UF5000, UF1000i and microscope detection respectively.

Individuals having clinically relevant urine specimens were included in the participant selection criteria, irrespective of age or illness. To ensure that the sample was accurate, both men and women were included. To maintain the homogeneity of the sample, people with renal illnesses, urinary tract infections, or any other condition that can change the composition of urine sediment were excluded. With 103 men and 86 women, the final distribution probably represents the gender distribution in the population under examination, providing an equal sample for a detailed examination.

Instrumental testing

The urine sample was centrifuged at 1500 r/min for 20 min, the supernatant was discarded, and 0.5 ml sediment was taken on the analyzer and analyzed sequentially by automatic injection mode, i.e. after a test needle was inserted into the urine sediment sample, the device automatically detected and displayed the test result. After the test result was judged to reach a stable state, the second analysis was carried out in the reverse order to minimize the influence of residue and drift on the repeated average value in strict accordance with the instrument instruction manual, record the detection values of RBC, WBC, CAST, etc. of each specimen of the two instruments. If there is an obvious counting difference between one or more parameters of the two analyses (CV of the two counts is > 10%), repeat the analysis for the third time, select the calculated average value of the two counts in which CV is < 10% as the final measurement value, and the original record is archived and backed up.

Microscopic examination

Microscopic examination was performed by two experienced examiners through blind experiments. After testing by the instrument, the specimens in the other test tube were centrifuged with a low-speed centrifuge, centrifuged at 1500 r/min for 5 min, the centrifuge tube was inclined at 45°–50°, the supernatant was discarded, the remaining 0.2 mL sediment was mixed evenly, and 20 μL of the sediment was aspirated into the counting plate of abalone cattle for microscopic examination. RBC, WBC, CAST, and other urine visible components were calculated under the microscope, the average value was taken as the microscopic counting result, and the microscopic examination method was used as the gold standard. Two inspectors have carried out personnel comparisons according to ISO15189 in advance, and the comparison results meet the requirements. To ensure the detection result, the microscopic examination should be completed within 2 hours after the collection of the specimen.

3.3.3. Criteria for outcome judgments

In this experiment, the accuracy of RBC, WBC, and CAST were compared. Positive criteria for UF5000 were: RBC > 18 μL, WBC > 13 μL, CAST > 1 μL for men; RBC > 33pL, WBC > 34 μL, CAST > 1 μL for women; positive criteria for UF1000i were RBC > 15 pL, WBC > 18 μL, CAST > 2.5 pL for men; RBC > 18 pL, WBC > 23 μL, and Microscopic positive results (10 visual fields observed under high magnification): RBC 0~3/HP, WBC > 3/HP (male) or > 5 /HP (female), transparent tube type CAST 0~1/LP or visible other pathological casts. False positive (FN) = (amount of false positives (FP) instances /amount of negative instances on microscopic examination); FN = (amount of FN instances/number of positive instances on microscopic examination); sensitivity = (amount of true positive (TP) instances on instrument/number of positive instances on microscopic examination); specificity (amount of true negative (TN) instances on instrument /number of negative instances on microscopic examination), among them, the FP rate and the FN rate of < 5% at the same time are acceptable (the FN rate of < 5% must be verified by referring to the reexamination rules for urinary sediment analysis in the CNAS application guidelines)

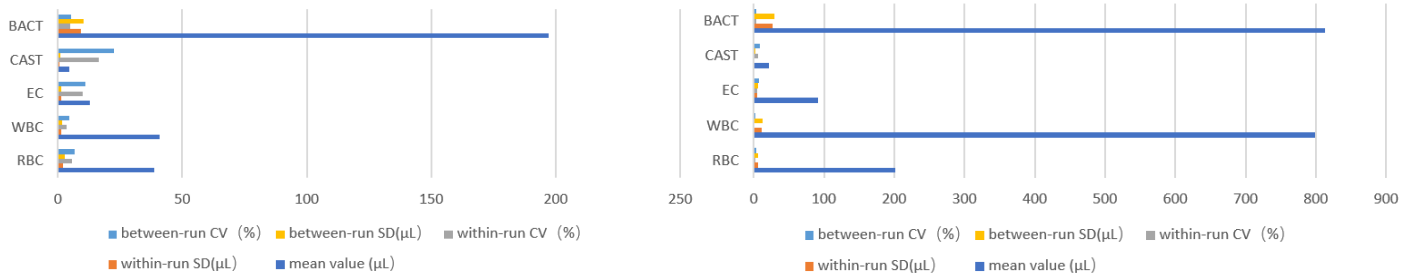
3.4. Statistical methods

SPSS 23.0 was used to process and analyze the data. Using microscope results as the gold standard, the coincidence rate of UF5000 and UF1000i for RBC, WBC, and CAST was calculated. χ^2 test was used to compare the accuracy, false positive rate, and false negative rate of different test items of different genders and between two instruments in UF5000. The variance is statistically significant ($P < 0.05$).

4. Results

4.1. Horizontal quality control

The within-run and between-run precision was tested to meet the quality control of low and high levels, which were shown in **Figure 1**. Variations between between-run and within-run CV and SD are shown through the evaluation of the blood component quality. WBC has the lowest within-run and between-run CVs of all the components, indicating reliable observations. BACT and RBC show an appropriate level of quality. CAST shows increased variability in batches exhibited by higher between-run CVs as compared to within-run CVs. EC have an average CV value and exhibit constant quality. These outcomes demonstrate various levels of efficiency for various blood components.

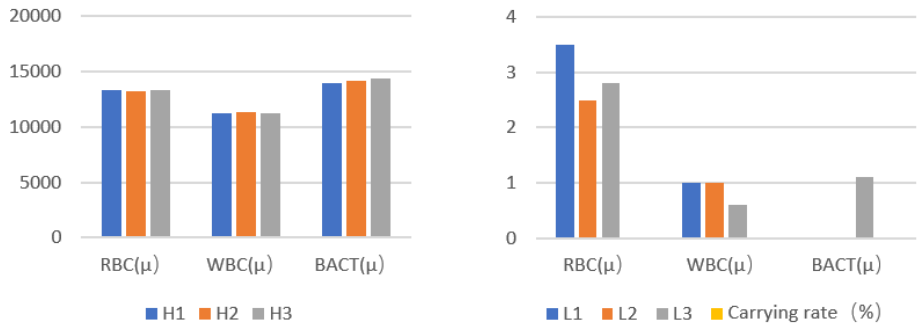


(a) **(b)**

Figure 1. Within-run and Between run precision: **(a)** Low level quality control; **(b)** High level quality control.

4.2. Carrying contamination rate

RBC carrying pollution rate is 0.01%, WBC is 0.01%, and BACT is 0.01%, which meets the manufacturer's requirements. The results are shown in **Figure 2**.



(a) **(b)**

Figure 2. Total amount: **(a)** RBC carrying contamination rate; **(b)** RBC carrying pollution rate.

4.3. Linear range and reportable range

The correlation coefficients of RBC, WBC, and BACT met the requirements. According to the maximum allowable CV value, the reportable ranges are RBC: 13253.60 × 2 μL, WBC: 12489.60 × 8 μL, and BACT: 12809.44 × 127 μL respectively, as shown in **Table 1**.

Table 1. Linear range and reportable range.

	Correlation coefficient	Linear range (/μL)	Maximum dilution ratio of CV value allowed	Reportable range (/μL)
RBC	0.9963	0~13253.60	2	0~13253.60 × 2
WBC	0.9994	0~12489.60	8	0~12489.60 × 8
BACK	0.9997	0~12809.44	127	0~12809.44 × 127

4.4. Evaluation of the diagnostic performance of instruments by gender

To compare the diagnostic performance of UF5000 and UF1000i instruments with a microscope as the gold standard, the analysis showed that the RBC false positive rate of UF5000 was significantly lower than that of UF1000i, while the false positive rate of CAST is more efficient than the UF1000i with significant variance. Further comparing the accuracy of different test items in UF5000 for men and women, the false negative rate of WBC in men was significantly lower than that in women. The specificity of leukocytes detected by the two instruments has no variance. The specificity of erythrocytes and casts of UF5000 was more improved contrasted with UF1000i, and the sensitivity of the three commonly used parameters of UF5000 is higher than the other one, the difference was statistically significant ($P < 0.05$), as shown in **Figure 3**.

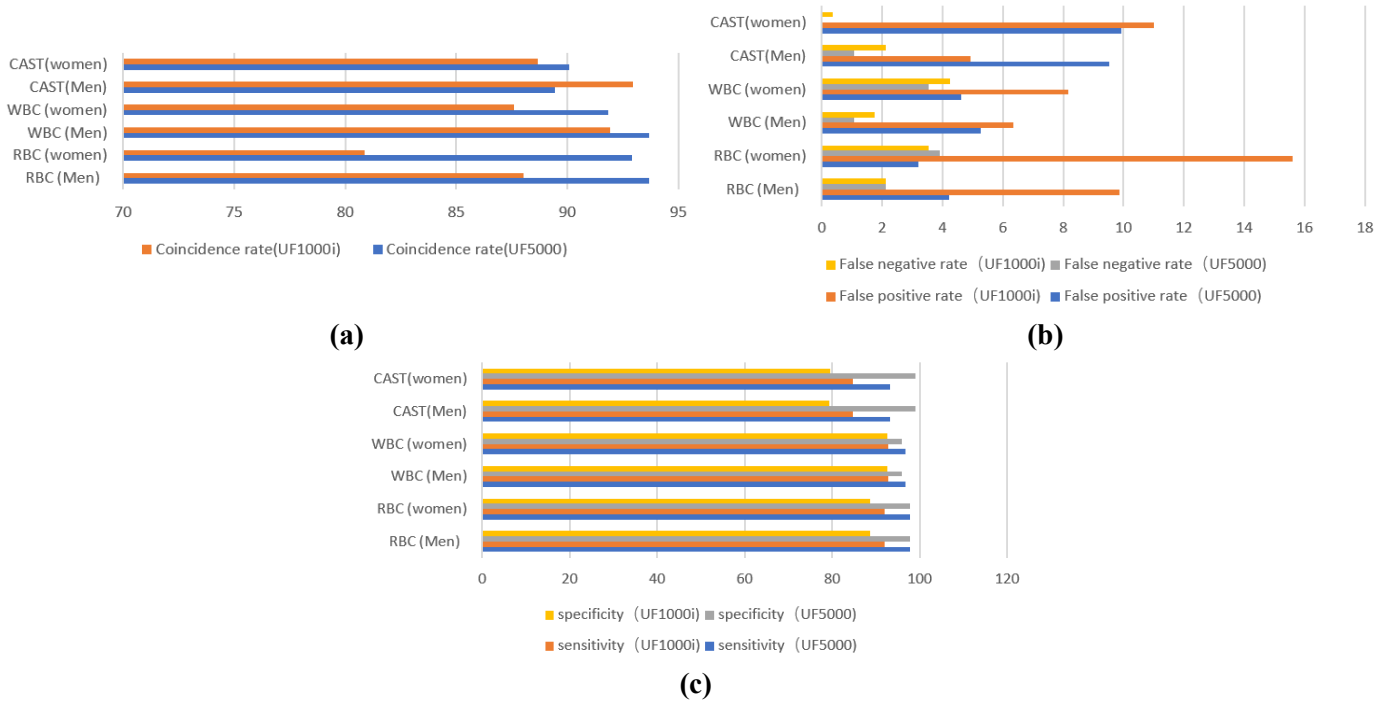


Figure 3. Exact value: **(a)** Coincidence rate of UF1000i and UF5000; **(b)** False negative and positive rate of UF1000i and UF5000; **(c)** Sensitivity and specificity of UF1000i and UF5000.

5. Discussion

A vital component of the basic healthcare system is the laboratory-based treatment for UTI diagnosis. It helps specialists make medical choices, maintains a focus on conditions that involve renal and urinary tract issues significantly lowers total medical expenses over time, and can prevent the development of resistance to antibiotics. Significant differences in performance were identified between the UF5000 and UF1000i devices and were analyzed using microscopy as an instance of comparison. In comparison with UF1000i, UF5000 showed improved specificity for erythrocytes and castings and decreased RBC false positive rates. UF5000 showed greater sensitivity throughout an extensive variety of frequently evaluated parameters. Men have lower WBC negative error rates demonstrated by an interesting gender-based investigation. The RBC, WBC, and BACT contamination rates and linear range exceeded the specifications and can be made available. A statistically significant difference was identified between the rate of false positives of UF5000 RBC and UF1000i, which was significantly reduced, and the incorrect positive rate of CAST, which was higher than UF1000i. These outcomes emphasize the increased accuracy and specificity of UF5000, particularly for identifying erythrocytes and casts, providing improved diagnostic potential in urine sediment analysis.

6. Conclusion

When compared to the UF1000i, the UF5000 offers significantly improved accuracy, high precision, a low contamination rate, a wide linear range, and a reportable range. It can exceed the laboratory's daily urine detection requirements. Whereas considering urine has a complex composition and wide range of morphological variations, and the number of variables, including sample gathering and storage conditions can affect the outcomes, false positives and false negatives are prevalent, which leads to clinical practice inaccuracies. To improve the accuracy of illness assessment, it consequently needs to determine an appropriate choice based on the real condition of the patients, requiring more investigation and analysis.

Author contributions: Conceptualization, CL and YL; methodology, JL; software, PW; validation, CL, QL and JL; formal analysis, QL; investigation, CL; resources, QL; data curation, PW; writing—original draft preparation, CL; writing—review and editing, CL; visualization, QL; supervision, PW; project administration, YL; funding acquisition, JL. All authors have read and agreed to the published version of the manuscript.

Ethical approval: Not applicable.

Conflict of interest: The authors declare no conflict of interest.

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