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Comparative study of urine erythrocyte detection by urine dry chemical analyzer, urine sediment analyzer and microscope

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Abstract: 200 urine samples were measured by Sysmex UN9000 automatic urine analysis line(consists of UF5000 automatic urine formed element analyzer and UC3500 automatic urine analyzer) respectively for the urine dry chemical analysis, urinary sediment analysis and artificial phase contrast examination, to explore the reliability of urine dry chemical analyzer and urine sediment analyzer to test the red blood cells in the urine by comparing the results of urine red blood cells with urine dry chemistry analyzer, urine sediment analyzer and microscope. The sensitivity of urinary sediment analyzer detection was 95.2% and the specificity was 93.8%; urine dry chemistry analyzer detection sensitivity was 97.1% and the specificity was 86.0%; the urine erythrocyte morphology coincidence rate of urine sediment analyzer was 88.5%. Therefore, the combination of dry chemical analyzer and urinary sediment analyzer can obviously improve the detection rate and provide a reliable basis for clinical diagnosis.

Urine routine is mainly used in the diagnosis and treatment of urological, endocrine and other diseases, nephrology, especially rely on, is one of the three routine clinical tests, the diagnosis and treatment of disease and prognosis of judgment is of great significance. Urine routine includes urine sediment analysis and urine dry chemical analysis. Urine sediment analysis test can make the patient's condition and the type of disease more clearly expressed by using various methods to analyze the precipitate in the urine and analyze the many types of organic components (bacteria, parasites, crystals, etc.). Traditional urine sediment examination methods can be divided into stained urine sediment microscopy method, non-stained urine sediment microscopy method and urine sediment quantitative method[1] . With the development of science and technology, many hospitals have chosen to use automatic urine analyzers for routine urinalysis in order to improve detection efficiency and accuracy. Urine dry chemistry analyzer can detect various indicators in urine (protein, ketone bodies, white blood cells, occult blood, glucose, bilirubin, urine specific gravity, PH). Urine dry chemistry analyzer testing specimens in smaller quantities, the relative accuracy is higher, and contains a number of items of testing, for high-volume specimen screening has a certain positive significance, but its red blood cells, crystals, etc. may cause misdiagnosis or omission. Both tests

have their own advantages and limitations, and sometimes the results may even be contradictory, increasing the difficulty of clinical diagnosis. Manual microscopy with phase contrast mirror is required when necessary. Based on this, the reliability of the three assays for the detection of RBCs and their morphology in urine is analyzed and compared.

1. Materials and Methods

1.1 Reagents and instruments (Including routine detection and morphological detection)

Sysmex UN9000 (UF5000 automatic urinary component analyzer + UC3500 automatic urine analyzer, made in Japan) and its quality control fluid and the original supporting reagents; Nissan Olympus CX41RF phase contrast microscope (made in Japan); Baiyang BY-320A medical centrifuge (made in China).

1.2 Specimen source (Inclusion criteria)

200 fresh morning urine specimens from outpatients and inpatients were obtained from the Affiliated Hospital of Hebei University from December 2021 to March 2022 (the specimens were occult blood positive specimens and all of them were subjected to manual phase contrast microscopy).

1.3 Collection Methods (To ensure freshness)

Mid-morning urine was collected in a clean container, and the specimen was thoroughly mixed and divided into three portions for use in three different assays, which were operated by the teacher of the body fluids group within the Laboratory Department within half an hour.

1.4 Urine dry chemical analysis (Screening test)

Before each day's use, the UC3500 automatic urine analyzer was firstly analyzed for quality control, and the test process ensured that the results were all under control, and then each specimen was standardized for measurement.

1.5 Urinary Sediment Analysis (Flow cytometry)

The UF5000 Automatic Urine Organic Fraction Analyzer was first analyzed for quality control before use each day, and the results were guaranteed to be under control during the testing process. According to the operating instructions for each mixed specimen for determination.

1.6 Phase contrast microscopy (Observe the morphology of the urinary erythrocytes)

Specification of each specimen was determined by mixing 10 ml of fresh sterile morning urine in a centrifuge tube, and then placing it in a centrifuge and centrifuging it at a speed of 1500 r/min for 10 min[2] . The supernatant was poured off, mixed, and then microscopically examined on a urinary sediment-pair counting plate and slide and observed the morphology of the urinary erythrocytes.

1.7 Judgment method (From expert consensus and textbook)

The erythrocyte information displayed by the urine sediment test analyzer is 1.0 for homogeneity, mainly normal red blood cells (abnormal pattern red blood cells <32%); the erythrocyte information

displayed by the urine sediment test analyzer is 2.0 for non-homogeneity, mainly abnormal pattern red blood cells (abnormal pattern red blood cells ≥72%); the erythrocyte information displayed by the urine sediment test analyzer is 3.0 for mixed (32% ≤ abnormal pattern red blood cells <72%). as mixed (32% ≤ abnormal pattern erythrocytes < 72%)[3-5] .

1.8 Statistical methods (Using SPSS 19)

The chi-square test in medical statistics was applied to analyze the data of the information, and $P < 0.05$ indicated that there was a significant difference between the two.

2. Results

2.1 Analysis results of erythrocyte detection rate (Comparison of instrument and manual method)

Taking the results of manual phase contrast microscopy microscopy as the gold standard, the detection rate specificity of erythrocytes by urine sediment detection analyzer was 93.8%, and the sensitivity of the detection rate was 95.2%, and the detection rate specificity of erythrocytes by urine dry chemical detection analyzer was 86.0%, and the sensitivity of the detection rate was 97.1 %, and the urine sediment detection analyzer and microscopic detection were done with the x^2 test, and *the results indicated that there was statistical significance; and the results indicated that there was statistical significance; and the results indicated that there was statistical significance with the urine dry chemical detection analyzer P<0.05 (see Tables 1, 2). lt;*0.05, the result indicated statistical significance; P<0.05 for microscopy and urine dry chemical detection analyzer, the result indicated statistical significance (see Table 1, 2).

Table 1: Results of erythrocytes detected by urine sediment test analyzer and microscopy

microscopic examination	Urine Sediment Analyzer Test		add up the total
	examine and discover	not detected	
erythrocyte detection	100	5	105
Erythrocytes not detected	5	90	95
add up the total	105	95	200

Table 2: Urine dry chemistry analyzer test and microscopy results for red blood cells

microscopic examination	Urine dry chemistry analyzer test		add up the total
	examine and discover	not detected	
erythrocyte detection	102	3	105
Erythrocytes not detected	77	18	95
add up the total	179	21	200

2.2 Analysis results of erythrocyte morphology compliance rate (Comparison of instrument and manual method)

Taking the results of manual phase contrast microscopy as the gold standard, when the erythrocyte information detected by the urine sediment test analyzer was homogeneous (normal

erythrocytes >72%), the compliance rate was 71.9%; when the erythrocyte information was non-homogeneous (normal erythrocytes ≦32%), the compliance rate was 94.8%; when the erythrocyte information was mixed (normal erythrocytes between 32%-72%), the compliance rate was 50.0%; urine sediment detection analyzer and manual phase contrast microscopy microscopy erythrocyte morphology compliance rate (the ratio of the sum of the fully compliant specimens in all phases to the total number of specimens) was 88.5%, urine sediment detection analyzer detection and microscopic erythrocyte morphology analysis to do the chi-square test, $P<0.05$, the results indicated statistically significant (see Table 3).

Table 3: The results of urine sediment test analyzer and manual phase contrast microscopy match

Artificial phase contrast mirror detection	Urinary Sediment Test Analyzer Test			add up the total
	1.0 △	2.0	3.0	
Alien red blood cells <32%	23	7	4	34
Alien red blood cells ≥72%	7	148	2	157
32% ≤ anomalous erythrocyte <72%	2	1	6	9
add up the total	32	156	12	200

△ Note: See 1.7 for determination methodology.

3. Discussion

In this study, 200 fresh morning urine specimens of outpatients and inpatients were collected from the Affiliated Hospital of Hebei University, and the erythrocytes and their morphology in urine were analyzed. In the dry chemistry test, the determination index of urinary occult blood is hemoglobin in urine, and the main principle is that peroxidase is similar to the structure of ferrohemoglobin in blood, and when catalyzed, it will generate new ecological oxygen and produce discoloration. Hemoglobin in this test is more sensitive, but myoglobin, strong oxidants, bacteriuria, etc. can cause its false positives, women's urine test on vacation is also usually positive; when the concentration of VC is too high, it will also interfere with the results to produce false-negative results, so the specificity is low . Urinary sediment analysis is more efficient by applying the principles of flow cytometry and electrical impedance analysis, but crystals and yeast, which are similar in morphology and size to erythrocytes, can be mistaken for erythrocytes, and its specificity (93.8%) is higher than the dry chemistry specificity (86%); the sensitivity (95.2%) is lower than the dry chemistry sensitivity (97.1%). Some studies have proved that the combination of urine sediment and dry chemistry can effectively improve the accuracy of routine urine examination[6-8] , which is better than the two methods alone. Manual phase contrast microscopy is the gold standard for the detection of erythrocytes and their morphology, but the results will be affected by personal operation, etc. At the same time, the use of manual microscopy will greatly increase the workload of the staff, and it is not suitable for the detection of large quantities of specimens.

In this study, it can be found through the results of the urine sediment test analyzer and manual phase contrast microscopy detection of red blood cell morphology, when the abnormal morphology of red blood cells in the specimen is greater than 72%, the highest compliance rate of the instrument and manual phase contrast microscopy, 94.8%; when the abnormal morphology of red blood cells in the specimen is less than 32%, the compliance rate of the instrument and manual phase contrast microscopy is 71.9%; when the abnormal morphology of red blood cells in the specimen is in the range of 32%-72%, i.e., when the number of normal morphology erythrocytes and abnormal morphology erythrocytes in the specimen were equal, the compliance rate was the lowest, which was only 50%, and at this time, the single data could no longer provide accurate test results. The

results of this study showed that the compliance rate of the urine sediment test analyzer in the urine specimen erythrocyte morphology detection was 88.5%, and it was more accurate when the number of abnormal morphology erythrocytes accounted for a larger proportion.

Based on the above results, we analyzed that: when the abnormal morphology red blood cells in the urine were predominant and all were spiny, due to the uniform size of their morphology and the generation of similar electrical impedance, they might be determined by the instrument to be 1.0 (homogeneous, anomalous erythrocytes ＜32%); when the normal red blood cells in the urine were predominant, but due to the change of osmolality or the morphology change generated by the long immersion time, they might be determined by the instrument to be 2.0 (non-homogeneous, anomalous morphology ≥72%) or 3.0 (mixed, 32% ≤ abnormal morphology <72%); when the number of normal morphology erythrocytes and abnormal morphology erythrocytes in the urine is comparable, the cellular morphology is more complex, and the accuracy of instrumental determination is reduced, manual phase contrast microscopy detection is required.

To sum up, in the urine routine testing of this common clinical project, the manual phase contrast microscopy as the gold standard, urine dry chemical analyzer and urine sediment detection analyzer can improve the sensitivity and specificity of urine erythrocyte detection, compared with the traditional manual microscopy, its speed greatly improved, has a very good future prospects for development. However, in the detection of urine erythrocyte morphology, in order to avoid the influence of the instrument itself and other interfering factors, to improve the accuracy, manual phase contrast microscopy is essential, urine dry chemistry test analyzer, urine sediment test analyzer and manual phase contrast microscopy of the three joint, in order to provide more accurate results for the clinic.

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