

Principle

Intended Use

For the determination of leukocyte esterase in urine.
AUTION 9EB Strips are manufactured for use only in the AUTION MAX AX-4280 Urine Analyzer.

Clinical Significance

Leukocyturia is an important sign of inflammation of the urinary tract, including kidneys, bladder, urethra and prostate. The source of inflammation can be bacterial, chlamydia, monilial, viral, mycoplasmal, trauma, and foreign bodies.

Methodology

AUTION Strips 9EB are multi-parameter test strips with leukocytes intended for use with the AUTION MAX AX-4280 Urine Analyzer to measure certain constituents in urine. The AUTION 9EB consist of 9 pads impregnated with chemicals specific for the determination of a particular constituent affixed to a plastic strip.

One of the pads permits the determination of leukocytes. An additional correction pad is included, to compensate for the natural color of urine and its effect on the color reaction of the leukocytes pad.

The AUTION MAX AX-4280 Urine Analyzer utilizes dual wavelength reflectance spectroscopy, in combination with AUTION Strips reagent chemistry, to provide semi-quantitative results of leukocytes. At the defined wavelength for leukocytes, the AUTION MAX AX-4280 Urine Analyzer analyzes the color and the intensity of reflected light from the leukocytes pad to calculate clinically meaningful leukocytes results.

Chemical Reaction Scheme

Measurement of leukocyte esterase activity



TAI = 3-(N-Toluenesulfonyl-L-alanyloxy)indole
MMB = 2-Methoxy-4-(N-morpholino)benzenediazonium

Specimen Collection and Preparation

Acceptable Sample Containers

Sterile collection bottles
BD yellow top urinalysis tubes
BD tiger top urinalysis tubes with preservative.

Gray Top culture tubes are not acceptable.

Sample Collection

A clean freshly voided midstream specimen should be collected in a clean container for routine analysis, and a sterile container for UACII requests. Infant bag collections are acceptable for children ≤ 2 years of age. Other acceptable specimens include catheterized specimens, suprapubic and ostomy collections, as well as kidney or bladder collections from the operating room.

BD tiger top urinalysis preservative tubes must be filled to a level between the marked minimum and maximum lines on the tubes (7-9 mL). Under-filled or over-filled tubes are unacceptable.

For best results, BD yellow top urinalysis tubes without preservative require eight (8) mL for UA or UACII. Urine specimens with a volume < 3 mL will be diluted for microscopic analysis, if possible. Urine specimens with a volume < 1 mL may not have enough volume for microscopic analysis.

Specimens exhibiting gross hematuria cannot be tested on the AX-4280. Gross hematuria may cause incorrect results in subsequent samples.

If analysis cannot be performed within one hour after collection, immediately refrigerate (2° and 8° C) the specimen. Bring the specimen to room temperature prior to analysis. Do not centrifuge the specimen prior to analysis.

The specimen volume placed on the iQ System must be at least 3 mL. If testing on the AX-4280 module only, the minimum volume is 2 mls. If testing on the iQSeries module only, the minimum volume is 2 mL.

Sample Stability and Handling

Urine collected without preservative at room temperature must be delivered to the lab within 1 hour of collection.

Urine collected without preservative and immediately placed on ice must be delivered within 4 hours of collection.

Urine collected in BD urinalysis preservative tubes will be accepted up to 48 hours after collection.

All specimens should be handled using the principles of Universal Precautions, and must be capped tightly.

Specimens that leak are unacceptable for analysis.

Reagents

Contents: 100 AUTION strips 9EB per container
Reorder # 800-3510

Reactive Ingredients

for leukocytes pad on AUTION test strip	Concentration
3-(N-Toluenesulfonyl-L-alanyloxy)indole (TAI)	0.49 mg
2-Methoxy-4-(N-morpholino)benzenediazonium	(MMB) 0.17 mg

Storage and Handling

Store AUTION strips between 1° and 30°C. DO NOT FREEZE. PROTECT AGAINST HEAT, LIGHT AND MOISTURE (Ambient Humidity). Each vial contains a desiccant to prevent exposure to moisture in the air (humidity). Immediately re-cap vials after removal of desired number of strips.

Reagent Stability

AUTION Strips are stable for two years after the date of manufacture, when stored in their original, unopened container, and maintained at 1° and 30°C.

Once AUTION test strips are placed in the hopper, they are only stable for 72 hours. Allow the hopper to empty at least once every 24 hours.

Acceptable Reagent Performance

Any discoloration of the pad(s) may indicate deterioration. If discoloration is observed or if control/patient results are questionable or in conflict with expected results, check the following:

Leukocytes-Urine, on AUTION 9EB multi-parameter test strip
ARKRAY USA, Inc., manufacturer

Technical Procedure 3306

1. Confirm the AUTION strips are still within the expiration date indicated on the vial.
2. Controls are within the expected range.
3. Results are equivalent when fresh product is tested

If a problem still exists, contact IRIS Customer Service Support at (800) 776-4747

Precautions and Warnings

For In Vitro Diagnostic Use: Utilize standard precautions required for the handling of all laboratory reagents.

Warnings: Toxic. AUTION strips contain one or more of the following chemicals: Diazonium salt and phenol

Gloves: Avoid contact with skin and mucous membranes. Wearing of gloves, when handling blood and body fluids, is included in the Center for Disease Control's recommended universal precautions.

Equipment

This test on the AUTION 9EB strips is used with the AUTION MAX AX-4280 Urine Analyzer. The analyzer is manufactured and supplied by ARKRAY FACTORY, Inc. in Japan and distributed by Iris Diagnostics, A Division of IRIS International, Inc., Chatsworth, California.

For technical assistance, contact IRIS Customer Service Support at (800) 776-4747.

Calibration

Weekly calibration verification is performed using one white and one gray check strip on the AUTION MAX AX-4280 Urine Analyzer. Refer to the Iris Maintenance Procedure, AX-4280 module weekly schedule and check off sheet.

Quality Control

At least two levels of control material should be analyzed each shift. Parallel testing between the old shipment or lot number and the new shipment or lot number will be done to assure acceptable strip performance.

The following controls should be prepared and used in accordance with the package inserts. Allow controls to come to room temperature and mix well for several minutes before testing. Quality Control results should be evaluated and handled with respect to the Clinical Chemistry Quality Control Procedure #3000.T. Strip lot changes are documented on the IRIS reagent log sheet.

Quality Control Material

Control	Storage
MAS Liquid UA Abnormal Control 1	+2°C to +8°C*
MAS Liquid UA Normal Control 3	+2°C to +8°C*

*Urine controls are received and stored at 2°C to 8°C. Bottles of controls in use are stored at +2°C to +8°C and are good for 30 days

Testing Procedure

Follow the correct testing set-up, testing and control procedures, as outlined in the AUTION MAX AX-4280 Urine Analyzer [Operating Manual](#).



Standard Reporting Format

Leukocyte esterase results, using AUTION Strips, in combination with the AUTION MAX AX-4280 Urine Analyzer, are reported in clinically useful and common units of measure. Results can be printed directly from the urine analyzer and/or transferred to the LIS.

1. Results are reported semi-quantitatively as Negative, Trace, Small, Moderate and Large.
2. Reference interval: Negative.
3. Normal urine specimens should not produce a positive reaction. Small amounts of leukocyte esterase, causing a positive reaction should be repeated, using a fresh urine specimen, from the same patient. Positive results require further testing for pyuria.

Procedural Notes

Limitations

The leukocytes test pad reacts with esterase derived from granulocytic white blood cells (WBC) in the urine specimen.

Interferences

1. Urine with glucose more than 500 mg/dL, protein more than 300 mg/dL, low pH, and elevated specific gravity can cause false negative results.
2. Urine with formaldehyde can cause false positive results.

Carryover

Studies were performed to assess the amount of analyte carried over by the AUTION MAX AX-4280 Urine Analyzer from one specimen reaction into subsequent specimen reactions. The studies consisted of the measurement of four identical high control samples immediately followed by four identical low control samples. Results of the testing demonstrated zero carryover for bilirubin as measured by the AUTION MAX AX-4280.

Gross hematuria may cause incorrect results in subsequent samples. Do not test specimens exhibiting gross hematuria. If carryover is suspected, sample a few tubes of saline and rerun samples that followed a sample that may have caused carryover.

Performance Characteristics

The performance characteristics of the leukocytes pad test on the AUTION Strips have been determined in clinical and analytical studies. In clinical studies, the sensitivity of the leukocytes pad test depends on several inherent factors, including pH, Specific Gravity and interfering factors (see [Interferences](#)). Generally, the leukocytes reagent pad test on the AUTION strips have been developed to be specific for leukocytes being tested.

Sensitivity and Range

Sensitivity and linearity studies were performed using a series of contrived control materials which covered a wide range of known analyte concentrations. The results of the studies using the AUTION MAX AX-4280 Urine Analyzer demonstrated analytical sensitivity and linearity over the measurement range as shown for leukocytes.

1. Analytical Sensitivity: Approximately 25 Leukocytes/uL
2. Analytical Measurement Range: 25 – 500 Leukocytes/uL

Semiquantitative result	Trace	Small	Moderate	Large	
Value (Leu/μL)	<25	25	75	250	500

Method Comparison

The performance of the AUTION MAX AX-4280 Urine Analyzer was evaluated in comparison with a commercially available automated urinalysis system. Urine samples were obtained from a hospital laboratory and included both normal and abnormal levels of urine leukocytes. Some of the native urine samples were spiked to elevated levels in order to achieve the desired range of abnormal values. Both urinalysis systems provide semi-quantitative results.

Therefore, for purposes of data analysis, for leukocytes, results for the individual urine samples were referred to the cut-off value for each system to discriminate between negative (normal) and positive (abnormal) findings. Overall agreement, sensitivity (positive agreement), and specificity (negative agreement) between the AUTION MAX AX- 4280 Urine Analyzer and the comparative system are shown in the table below.

Analyte	No. of Samples	Overall Agreement (%)	Sensitivity (%)	Specificity (%)
Leukocytes	180	98.9	100.0	98.3

Validation Studies performed at UCDHS

See attached validation method comparison sheet.

Precision

Reproducibility of the AUTION MAX AX-4280 Urine Analyzer was evaluated by performing replicate measurements of a control material within a single run. Measurement results were reported by the instrument in the reflectance values format instead of semi-quantitative values in order to allow for a continuous measurement output.

Within Run Precision Data

Analyte	Number of Replicates	Reflectance Value		CV (%)
		Mean	S.D.	
Leukocytes	21	43.02	1.22	2.85

Precision Studies performed at UCDHS

See attached AUTION MAX AX-4280 Precision Reflectance Report sheet.

Additional Information

For more detailed information on the AUTION Strips 9EB multi-parameter test strips with leukocytes and the AUTION MAX AX-4280 Urine Analyzer, refer to the [AUTION Strips 9EB for Urine Chemistry](#) package insert and the AUTION MAX AX-4280 Urine Analyzer [Operating Manual](#).

References

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8. Schersten, B. and Fritz, H. Subnormal Levels of Glucose in Urine. *JAMA* 1967. 201; 129-132.
9. *AUTION Strips 9EB For Urine Chemistry*, package insert, issued: Dec. 2001, Rev.: Dec. 2011; supplied by ARKRAY, Inc., Kyoto, Japan.

