

KOVA® Stain

A Stain for Urinary Sediment



INTENDED USE

KOVA Stain is a modification of the Sternheimer-Malbin stain¹ suitable for use as an aid in the differentiation of cellular elements found by microscopic examination of urinary sediment. Relative number, cell type and casts assembled by qualitative or semiquantitative examination of urine sediment are adequate in most cases for diagnostic purposes.^{2,3}

DESCRIPTION

KOVA Stain contains crystal violet, safranin O and ammonium oxalate in a stabilized dilute ethanol solution. For in vitro diagnostic use.

STABILITY AND STORAGE

Store at room temperature (18 – 30°C). Do not refrigerate or freeze. Crystals or debris may be observed in the stain, this is not uncommon for Sternheimer-Malbin stain. These artifacts may be safely removed by filtration. Stable per the expiration date on the label.

STANDARDIZED URINALYSIS PROCEDURE:

SPECIMEN COLLECTION

- For the best chemical and microscopic results, analyze a clean, fresh, first morning urine specimen.
- Due to the increased concentration of urine constituents, the first morning specimen is most useful. Constituents such as casts may be better observed under a microscope in the concentrated, first morning specimen.
- A random specimen (collected from an ambulatory patient who has eaten two to three hours earlier) is more suitable for the detection of reducing sugars.
- Disposable plastic specimen cups or disposable plastic containers with lids are suitable for sample collection. The use of Kova Cups, Kova Tubes and Kova Caps is recommended.
- Following collection, process the urine specimen as soon as possible. Processing within four hours is imperative to avoid deterioration of the sediment or a change in the chemical and physical composition. If this is not possible, refrigerate the specimen between 2° and 8°C.⁴ Do not freeze.

PHYSICAL TESTS:

- Appearance: Record the color and turbidity.
- Specific Gravity: Measure and record the specific gravity by use of a temperature compensated refractometer, hydrometer or urinometer.
- Osmolality: Measure and record the osmolality by use of an osmometer.

CHEMICAL TESTS:

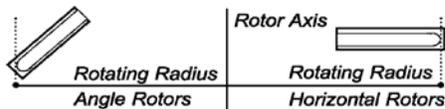
- Mix the urine well enough to resuspend any sediment that has settled to the bottom.
- Decant 12ml of the mixed urine into a graduated KOVA Tube.
- Using reagent test strips, perform chemical testing according to the manufacturer's instructions.
- Record the results.

CENTRIFUGATION AND MICROSCOPIC EXAMINATION

- Transfer a thoroughly mixed aliquot of KOVA-Trol or urine specimen to a KOVA Tube, filling it to the 12ml graduation.
- Centrifuge the KOVA Tubes (each containing 12ml of urine specimen or KOVA-Trol) at a relative centrifugal force (rcf) of 400 for five minutes; approximately 1500 revolutions per minute (rpm) with a 6-inch radius rotor. Formula used:

$$rcf = 28.38 (R) \left(\frac{N}{1000} \right)^2 \quad R = \text{radius of rotor in inches} \\ N = \text{revolutions per minute}$$

The rotating radius is the distance measured from the rotor axis to the tip of the liquid inside the tubes at the greatest horizontal distance from the rotor axis.



- Remove the KOVA Tubes from the centrifuge being careful not to disturb or dislodge the sediment.
- Insert a KOVA Petter into the KOVA Tube. Push the KOVA Petter to the bottom of the KOVA Tube until it seats firmly (at the 1ml graduation).
- Decant and discard 11ml from the KOVA Tube while the KOVA Petter is locked in position in the KOVA Tube. This will retain 1ml of urine sediment at the bottom of the KOVA Tube. With the use of the KOVA Decanting Rack, 10 tubes can be

decanted simultaneously. To release the KOVA Tubes, squeeze the top of the rack while pulling the tube straight up.

- Withdraw the KOVA Petter from the KOVA Tube.
- Add one drop of KOVA Stain¹ to the 1ml of urine sediment.
- Using the KOVA Petter, gently resuspend the sediment and stain until a homogeneous mixture is obtained.
- Withdraw a small sample of the urine sediment stain mixture by squeezing the bulb of the KOVA Petter.
- Transfer the sediment mixture to the KOVA Slide by placing one drop in the corner of the well. The chamber will fill by capillary action.
- Remove any excess specimen remaining on the open recessed area by touching the open edge with absorbent material.
- Place the KOVA Slide on a microscopic stage under the objective lens.
- Scan the slide chamber under low power magnification (10X eyepiece/10X objective) to enumerate casts. Enumerate all other formed elements under high power magnification (10X eyepiece/40X objective).⁵ Do not reuse KOVA products.

PRODUCT PERFORMANCE

Microscopic Characteristics of Sediment

Element	Staining Characteristic		Comment	
Red Blood Cells	Urine pH acid— slight purple neutral — pink (unstained) alkaline — dark purple			
	(Nuclei)	(Cytoplasm)		
White Blood Cells	Neutrophilic (dark staining)	red-purple	violet to purple granularity	larger than dark staining cells cytoplasmic granules with or without Brownian movement
	Glitter Cells	light blue to almost colorless	pale blue	
Epithelial Cells	Renal Tubular	dark purple	orange-purple	single rounded Nucleus differentiates from leukocytes
	Bladder Tubular	dark blue	pale blue	
	Squamous	purple	pink to violet	
Hyaline Casts	(Inclusions)	(Matrix)	very homogenous	
	----	pink to light purple		
Fine Granular Casts	purple granules	pink		
Course Granular Casts	deep purple granules	purple	Matrix is often not visible	
Fatty Casts	fat unstained	pink		
Red Cells Casts	lavender intact cells	pink		
Blood Casts (hemoglobin)	dark purple	---	hemoglobin from degenerated red blood cells	
Waxy Casts	---	light to dark purple	sharper outline than hyaline casts	
Bacteria	dead-dark purple	living and active — unstained to pink	motility of organisms is not impaired	
Mycelia Spores or Fungi	light purple			
Trichomonas Vaginalis	pale blue			
Background	pale pink			

MATERIALS PROVIDED

Product Number	Description	Packaging
87116	KOVA Stain	3 x 25ml

KOVA System products are available separately.

KOVA SYSTEM FOR QUALITY CONTROL - KOVA-Trol™, KOVA Liqua-Trol™

Human urine controls for physical, chemical and microscopic quality control.

Product Number	Description	Determinations Per Package
87329	KOVA-Trol I - High Abnormal without Abnormal Urobilinogen value assignment	4 x 15 mls
87325	KOVA-Trol I - High Abnormal Without Abnormal Urobilinogen Value Assignment	4 x 60 mls
87326	KOVA-Trol I - High Abnormal Without Abnormal Urobilinogen Value Assignment	10 x 60 mls
87332	KOVA-Trol I - High Abnormal With Urobilinogen Value Assignment	4 x 60 mls
87333	KOVA-Trol I - High Abnormal With Urobilinogen Value Assignment	10 x 60 mls
87334	KOVA-Trol I - High Abnormal With Urobilinogen Value Assignment	4 x 15 mls
87130	KOVA-Trol II - Low Abnormal	4 x 15 mls
87128	KOVA-Trol II - Low Abnormal	10 x 60 mls
87331	KOVA-Trol III - Normal with hCG	4 x 15 mls
87327	KOVA-Trol III - Normal with hCG	4 x 60 mls
87328	KOVA-Trol III - Normal with hCG	10 x 60 mls
87122	KOVA Liqua-Trol II - Normal with hCG with microscopics	2 x 120 mls
87123	KOVA Liqua-Trol II - Normal with hCG with microscopics	4 x 120 mls
87176	KOVA Liqua-Trol I - (Abnormal) with microscopics	2 x 120 mls
87177	KOVA Liqua-Trol I - (Abnormal) with microscopics	4 x 120 mls

KOVA SYSTEM AND SYSTEM COMPONENTS

Product Number	Description	Determinations Per Package
87153	KOVA System Super Pac 1000 w/Caps 100 KOVA Glasstic® Slide 10 (10 chambered), 1000 KOVA Petters, 1000 KOVA Super Tubes, 1000 KOVA Caps	1000
87154	KOVA System Super Pac 1000 100 KOVA Glasstic Slide 10 (10 chambered), 1000 KOVA Petters, 1000 KOVA Super Tubes,	1000
87155	KOVA System Pac II 400 100 KOVA Slide II (4 chambered), 400 KOVA Petters, 400 KOVA Super Tubes,	400
87156	KOVA System Value Pac 500 50 KOVA Glasstic Slide 10 (10 chambered), 500 KOVA Petters, 500 Economy Tubes	500
87141	KOVA KO-LEC-PAC® 500 KOVA Super Tubes, 500 KOVA Caps, 500 KOVA Cups, 500 Labels and 5 Transport Racks	500 (5 x 100)
87100	KOVA Slide II with Grid for quantitation; 100 x 4 well slides; each chamber contains 6.6 µl and has a 3mm x 3mm grid with each 1mm grid square = 0.15 µl	400

87118	KOVA Slide II (without grid) 100 x 4 well slides	400
87146	KOVA Glasstic® Slide 10 100 x 10 well slides in crystal clear Plexiglas*	1000
87157	KOVA Glasstic Slide 10 50 x 10 well slides in crystal clear Plexiglas*	500
87144	KOVA Glasstic Slide 10 with Grid 100 x 10 well slides in crystal clear Plexiglas* with quantitation grids; each chamber contains 6.6 µl and has a 3 mm x 3 mm grid with fine divisions of 0.33 mm x 0.33 mm. Test procedure includes method for quantitating cells per µl of patient samples.	1000
87137	KOVA Super Tube Graduated 12ml non-sterile disposable collection and centrifuge tubes made of high impact, unbreakable plastic to eliminate cracking or breaking during centrifugation.	500
87138	KOVA Economy Tube As above but in economical, styrene plastic.	500
87135	KOVA Petter Disposable plastic transfer pipette designed to retain 1.0 ml of urine after centrifugation. The unique lock tip provides a one-step contamination free decanting method.	500
87139	KOVA Cap Recommended to prevent spillage during transportation, as well as aerosol contamination during centrifugation.	500
87136	KOVA Decanting Rack Rack for decanting up to 10 specimens.	1 Rack

BIBLIOGRAPHY

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2. Henry, J.B. (Ed.): Todd-Stanford-Davidsohn: Clinical Diagnosis and Management by Laboratory Methods. 16th Edition, Vol. 1, W.B. Saunders Co., Philadelphia, 1979.
3. Welber, J.M. and Greene, J.A., Jr.: Examination of the Urine. New York, Meredith Publishing Co., 1966.
4. Haber, M.H.: A Primer of Microscope Urinalysis. ICL Scientific, 1978.
5. Siegle, M.D.: Urinoscopy - First the Microscope. Lab. Med. 12: 781-784, 1981.

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* Plexiglas is a trademark of Rohm & Haas.

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