Jones Staining Kit

REF 860-019

05279348001

IVD Σ 40

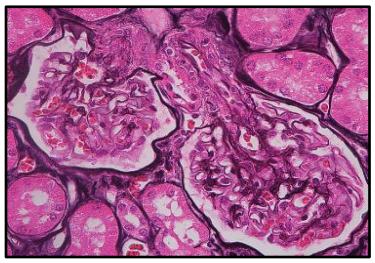

Figure 1. Jones Staining Kit staining kidney tissue.

INTENDED USE

The Jones Staining Kit is intended for laboratory use as a qualitative histologic stain to demonstrate capillary basement membranes by light microscopy in sections of formalin-fixed, paraffin-embedded (FFPE) tissue stained on the BenchMark Special Stains instrument.

This product should be interpreted by a qualified pathologist in conjunction with histological examination, relevant clinical information, and proper controls.

This product is intended for in vitro diagnostic (IVD) use.

SUMMARY AND EXPLANATION

Jones Staining Kit is a modification of Jones Methenamine Silver procedure.[1]

The glomerulus has a glomerular capillary basement membrane between it and the urinary space which is then encapsulated by the Bowman's capsule.[2,3] Glomerular disease occurs when there is a disruption to the glomerular filtration barrier that may allow red blood cells or plasma proteins to pass through the barrier.[4]

The Jones Staining Kit can highlight extracellular matrix proteins, adhesions to the Bowman's capsule, and breaks within the capillary basement membrane. This stain provides contrast and resolution of these delicate structures.[5,6,7]

The Jones Staining Kit demonstrates capillary basement membranes to aid the pathologist in the diagnosis of glomerular disease in kidney tissue.

PRINCIPLE OF THE PROCEDURE

The staining reaction is based on aldehyde reduction of silver ions to metallic silver under alkaline conditions. Jones Periodic Acid is used to oxidize carbohydrates to aldehyde groups. The combined Jones Silver A and Jones Silver B solutions form a methenamine-silver complex that is easily reduced to metallic silver by the aldehyde groups. To enhance staining contrast, toning of tissue specimen is completed using gold chloride within the Toner reagent. Fixer, with thiosulfate, stops the reaction and removes any unreduced silver from the section.[5,8] Nuclei are stained with Jones Hematoxylin and Jones Eosin is applied to provide a contrasting background.

This kit is optimized for use on BenchMark Special Stains instruments. The reagents are applied to tissue on microscope slides and mixed over the entire specimen.

MATERIAL PROVIDED

The reagent vials are supplied in barcode labeled carriers to insert into the reagent tray of the instrument. Each kit contains sufficient reagent for 40 tests:

One 19 mL vial of Jones Periodic Acid contains approximately 1% periodic acid.

One 19 mL vial of Jones Silver A contains approximately 1% silver nitrate.

One 15 mL vial of Jones Silver B contains approximately 2% sodium borate and approximately 14% methenamine.

One 15 mL vial of Toner contains approximately 1% gold chloride.

One 15 mL vial of Fixer contains approximately 2% sodium thiosulfate.

One 19 mL vial of Jones Hematoxylin contains modified Mayer's Hematoxylin (contains sodium iodate and ethylene glycol).

One 15 mL vial of Jones Eosin contains approximately 1% Eosin Y in an alcohol solution.

Seven vial inserts with sipping straws.

Reconstitution, Mixing, Dilution, Titration

No reconstitution, mixing, dilution, or titration of kit reagents is required. Further dilution of any of the reagents may result in unsatisfactory staining.

The reagents in this kit have been optimally diluted for use on BenchMark Special Stains instruments.

MATERIALS REQUIRED BUT NOT PROVIDED

Not all products listed in the method sheet may be available in all geographies. Consult your local support representative.

The following reagents and materials may be required for staining but are not provided:

1. Recommended control tissue
2. Microscope slides, positively charged
3. BenchMark Special Stains instrument
4. BenchMark Special Stains Deparaffinization Solution (10X) (Cat. No. 860-036 / 06523102001)
5. BenchMark Special Stains Liquid Coverslip (Cat. No. 860-034 / 06523072001)
6. BenchMark Special Stains Wash II (Cat. No. 860-041 / 08309817001)
7. General purpose laboratory equipment

STORAGE AND STABILITY

The Jones Staining Kit should be stored at 2-8°C. Refrigerated kit components should be brought to room temperature prior to use.

When properly stored, unopened and opened reagents are stable to the date indicated on the label. Do not use reagent beyond the expiration date indicated on the kit.

There are no obvious signs to indicate instability of these reagents; therefore, controls should be run simultaneously with unknown specimens. Contact your local support representative if positive control material shows a decrease in staining as it could indicate reagent instability.

SPECIMEN PREPARATION

Routinely processed, FFPE tissues are required for use with this assay and BenchMark Special Stains. The recommended tissue fixative is 10% neutral buffered formalin.[9]

Perform specimen collection and storage according to Histotechnology; A Self Instructional Text.[9] Cut sections to the appropriate thickness, approximately 2-4 µm, and place the sections on positively charged glass slides.

1. Dry the slides.[9]
2. Print appropriate barcode label(s).
3. Apply barcode labels to the frosted end of the slides prior to loading the slides onto the instrument (see the instrument User Guide for correct application of labels).

Refer to the Instructions for Use section for the recommended protocol for the BenchMark Special Stains instrument.

WARNINGS AND PRECAUTIONS

1. For in vitro diagnostic (IVD) use.
2. For professional use only.
3. **CAUTION**: In the United States, Federal law restricts this device to sale by or on the order of a physician. (Rx Only)
4. Do not use beyond the specified number of tests.
5. Positively charged slides may be susceptible to environmental stresses resulting in inappropriate staining. Ask your Roche representative for more information on how to use these types of slides.
6. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions. In the event of exposure, the health directives of the responsible authorities should be followed.[10,11]
7. Avoid contact of reagents with eyes and mucous membranes. If reagents come in contact with sensitive areas, wash with copious amounts of water.
8. Avoid microbial contamination of reagents as it may cause incorrect results.
9. For further information on the use of this device, refer to the BenchMark Special Stains instrument User Guide, and instructions for use of all necessary components located at navifyportal.roche.com.
10. Consult local and/or state authorities with regard to recommended method of disposal.
11. Product safety labeling primarily follows EU GHS guidance. Safety data sheet available for professional user on request.
12. To report suspected serious incidents related to this device, contact the local Roche representative and the competent authority of the Member State or Country in which the user is established.

This product contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

Table 1. Hazard information.

Hazard	Code	Statement
Danger 	H225	Highly flammable liquid and vapour.
	H314	Causes severe skin burns and eye damage.
	H317	May cause an allergic skin reaction.
	H360FD	May damage fertility. May damage the unborn child.
	H371	May cause damage to organs.
	H373	May cause damage to organs through prolonged or repeated exposure.
	H410	Very toxic to aquatic life with long lasting effects.
	P201	Obtain special instructions before use.
	P210	Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
	P260	Do not breathe mist or vapours.
	P273	Avoid release to the environment.
	P280	Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection.
	P303 + P361 + P353	IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water.
	P304 + P340 + P310	IF INHALED: Remove person to fresh air and keep comfortable for breathing. Immediately call a POISON CENTER/ doctor.
	P305 + P351 + P338 + P310	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/ doctor.
	P308 + P311	IF exposed or concerned: Call a POISON CENTER/ doctor.
	P370 + P378	In case of fire: Use dry sand, dry chemical or alcohol-resistant foam to extinguish.
	P391	Collect spillage.

EUH208: This product contains Gold chloride, hydrochloride, trihydrate, Sodium iodate. May produce an allergic reaction.

INSTRUCTIONS FOR USE

Prepare Reagent Vial

Before first use, a vial insert and sipping straw must be placed in the reagent vial.

Remove the shipping cap from the vial and place the insert and straw into the vial. The insert and sipping straw should be left in the vial, once the vial has been opened.

Staining Procedure

1. Load reagents and slides onto the instrument.
2. Place the soft cap into the slot on the reagent holder when the reagent is in use.
3. Perform the staining run according to the recommended protocol in Table 2, and the instructions in the User Guide.
4. When the run is complete, remove the slides from the instrument.
5. Use the soft cap to cover the reagent vial when reagent is not in use.
6. After use, store the reagents according to the recommended storage conditions.

Recommended Protocol

The parameters for the automated procedures can be displayed, printed and edited according to the procedure in the instrument User Guide.

The following procedures allow flexibility to accommodate user preference. This product has been optimized for use with the BenchMark Special Stains instruments but the user must validate results obtained with this product.

Table 2. Recommended staining protocol for Jones Staining Kit on a BenchMark Special Stains instrument.

Staining Procedure	S Jones
Protocol Step	**Method**
Deparaffinization	Select to automate paraffin removal.
Baking (optional)	The default is not selected. 75°C for 4 minutes is recommended.
Optimize Stain Intensity (Jones Silver B)	The default is 60°C for 12 minutes * Select to enable adjustment of staining intensity.** Select temperature from 55 - 60°C: 55°C, lighter staining intensity 60°C, darker staining intensity Select an incubation time from 8-20 minutes: 8 minutes, lighter stain 20 minutes, darker stain
Eosin or Light Green Counterstain	Select Eosin Counterstain to run the default protocol.
Optimize Hematoxylin Intensity (Jones Hematoxylin)	The default is 8 minutes. Select to optimize hematoxylin:** 8 minutes, lighter nuclear staining 16 minutes, darker nuclear staining
Optimize Eosin (Jones Eosin)	The default is 8 minutes. Select to optimize eosin:** 4 minutes, lighter counterstain 12 minutes, darker counterstain

**** If high background/mirroring is seen with the default protocol (60°C 12minutes):***

- Lower Jones Silver B step incubation temperature from 60°C to 55°C based on the allowed temperature dial-ability range (55°C to 60°C).
- Use Silver B incubation time (8 minutes, 12 minutes, and 16 minutes) to adjust the silver intensity as needed (shorter time stains lighter, and longer time stains darker).

***** To adjust staining preferences, increment the stain temperature and incubation time one parameter at a time.***

Recommended Post-Instrument Processing

1. Rinse slides in two changes of 95% ethanol to remove the leftover solution, followed by three changes of 100% ethanol.
2. Clear slides in three changes of Xylene.
3. Coverslip with permanent mounting media.

Compatible with the VENTANA HE 600 system coverslipping protocol. For further instructions, refer to the VENTANA HE 600 system User Guide.

QUALITY CONTROL PROCEDURE

An example of a positive control material would be FFPE human tissue that contains basement membrane.[10] Control tissue should be fresh autopsy, biopsy, or surgical

specimen prepared or fixed as soon as possible in a manner identical to test sections. Such tissues should monitor all steps of the analysis, from tissue preparation through staining.

Use of a tissue section fixed or processed differently from the test specimen provides control for all reagents and method steps except fixation and tissue processing. The cellular components of other tissue elements may serve as the negative control.

Optimal laboratory practice is to include a positive control section on the same slide as the test tissue. This helps identify any failures applying reagents to the slide. Control tissue may contain both positive and negative staining elements and serve as both the positive and negative control.

The control tissue must be tested with each run.

Known positive tissue controls should only be utilized for monitoring the correct performance of processed tissues and test reagents, not as an aid in formulating a specific diagnosis of patient samples.

If the positive tissue components fail to demonstrate positive staining, results with the test specimens should be considered invalid. If the negative components demonstrate positive staining, results with patient specimens should also be considered invalid.

Unexplained discrepancies in control results should be referred to the local support representative immediately. If quality control results do not meet specifications, patient results are invalid. The cause must be identified and corrected, and the patient samples repeated.

STAINING INTERPRETATION / EXPECTED RESULTS

Jones Staining Kit is tested to demonstrate basement membranes.

- Basement Membranes: Black
- Nuclei: Pink to Purple
- Background: Pink

SPECIFIC LIMITATIONS

Brown discoloration of glass slides has been observed with Jones Staining Kit. At expected levels, discoloration should not interfere with staining interpretation.

If high background/mirroring is seen with the default protocol (60°C 12 minutes):

- Lower Jones Silver B step incubation temperature from 60°C to 55°C based on the allowed temperature dial-ability range (55°C to 60°C).
- Use Silver B incubation time (8 minutes, 12 minutes, and 16 minutes) to adjust the silver intensity as needed (shorter time stains lighter, and longer time stains darker).

Only positively charged microscope slides have been used and validated for this assay.

PERFORMANCE CHARACTERISTICS

ANALYTICAL PERFORMANCE

Staining tests for sensitivity, specificity, and precision were conducted and the results are listed below.

Sensitivity and Specificity

Analytical sensitivity and specificity was evaluated in normal and diseased tissues cut at approximately 3 µm. All evaluated tissue cases (66/66) passed for acceptable staining as shown in Table 3.

Table 3. Sensitivity/Specificity of Jones Staining Kit was determined by testing the following FFPE normal and diseased tissues.

Tissue	# passed / # tested
Kidney (Normal)	41 / 41
Focal segmental glomerulosclerosis (Kidney)	4 / 4
Glomerular disease (Kidney)	4 / 4
Membranous glomerulonephritis (Kidney)	4 / 4
Membranoproliferative glomerulonephritis (Kidney)	3 / 3
Lupus nephropathy (Kidney)	7 / 7
Diabetic glomerulosclerosis (Kidney)	3 / 3

Precision

Precision of Jones Staining Kit was determined across multiple runs, days, instruments, and reagent lots using multiple cut slides from 6 normal kidney cases. Test cases were cut at approximately 2 µm. All acceptance criteria were fully met. Precision slides studies were performed according to Table 4

Table 4. Precision slide studies for Jones Staining Kit.

Parameters Tested	# of conditions	# passed / # tested
Run to Run	3 runs, same day	54 / 54
Day to Day	5 days	90 / 90
Instrument to Instrument	3 instruments	54 / 54
Intra Run	same day, same instrument	54 / 54
Lot to lot	3 lots	54 / 54

The results demonstrated no significant difference in staining intensity among the slides.

TROUBLESHOOTING

1. Section thickness may affect quality and intensity of staining. If staining is inappropriate, contact your local support representative for assistance.
2. Necrotic or autolyzed tissue may exhibit nonspecific staining.
3. If the positive control is negative, tissue may have been improperly collected, fixed, or deparaffinized. Follow the proper procedure for collection, storage, and fixation.
4. If the positive control is negative, check that the slide has the proper barcode label. If the slide is labeled properly, check the other positive controls from the same run to determine if the controls were properly stained.
5. If excessive background staining occurs: incomplete paraffin removal could cause staining artifacts or no staining. If all paraffin is not removed from the slide, repeat the staining run using the extended deparaffinization option, if available.
6. If tissue sections wash off the slide, confirm the slides are positively charged.
7. Extended stay of the slides on-board the instrument after run completion may affect quality and intensity of the staining. If the staining is inappropriate, remove slides promptly at the end of the run and proceed to post-instrument processing.
8. For corrective action, refer to the Instructions for Use section, the instrument User Guide or contact your local support representative.

REFERENCES

1. Koski JP. Silver methenamine-borate (SMB): Cost reduction with technical improvement in silver nitrate-gold chloride impregnations. J Histotechnol. 1981;3:115.
2. Kitching AR, Hutton HL. The Players: Cells Involved in Glomerular Disease. Clin J Am Soc Nephrol. 2016;11(9):1664-1674.
3. Mac-Moune Lai F, Szeto CC, Choi PC, et al. Isolate Diffuse Thickening of Glomerular Capillary Basement Membrane: A Renal Lesion in Prediabetes? Mod Pathol. 2004;17(12):1506-1512.
4. Hebert LA, Parikh S, Prosek J, et al. Differential Diagnosis of Glomerular Disease: A Systematic and Inclusive Approach. Am J Nephrol. 2013;38(3):253-266.
5. Bancroft JD, Layton C. Connective and Other Mesenchymal Tissues with Their Stains. In: Bancroft's Theory and Practice of Histological Techniques.2019:153-175.
6. Cathro HP, Shen SS, Truong LD. Diagnostic Histochemistry in Medical Diseases of the Kidney. Semin Diagn Pathol. 2018;35(6):360-369.
7. Herrera GA, Turbat-Herrera EA. Renal Diseases with Organized Deposits: An Algorithmic Approach to Classification and Clinicopathologic Diagnosis. Arch Pathol Lab Med. 2010;134(4):512-531.
8. Jones DB. Nephrotic Glomerulonephritis. Am J Pathol. 1957;33(2):313-329.
9. Carson F, Hladik C. Histotechnology: A Self Instructional Text, 3rd edition. Hong Kong: American Society for Clinical Pathology Press; 2009.
10. Occupational Safety and Health Standards: Occupational exposure to hazardous chemicals in laboratories. (29 CFR Part 1910.1450). Fed. Register.
11. Directive 2000/54/EC of the European Parliament and Council of 18 September 2000 on the protection of workers from risks related to exposure to biological agents at work.

NOTE: A point (period/stop) is always used in this document as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Symbols

Ventana uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see elabdoc.roche.com/Symbols for definition of symbols used):

GTIN	Global Trade Item Number
UDI	Unique Device Identification
	Indicates the entity importing the medical device into the European Union

REVISION HISTORY

Rev	Updates
G	Updates to Warnings and Precautions, Recommended Protocol, Specific Limitations, References, Symbols and Intellectual Property

INTELLECTUAL PROPERTY

CONTACT INFORMATION

Ventana Medical Systems, Inc.
1910 E. Innovation Park Drive
Tucson, Arizona 85755
USA
+1 520 887 2155
+1 800 227 2155 (USA)

www.roche.com

EC REP

Roche Diagnostics GmbH
Sandhofer Strasse 116
D-68305 Mannheim
Germany
+800 5505 6606

CE