



FLAGELLA STAIN

INTENDED USE

Remel Flagella Stain is recommended for use in qualitative procedures to stain bacterial flagella and identify their location on the bacterial cell.

SUMMARY AND EXPLANATION

Flagella are structures embedded in the bacterial cell envelope that are responsible for motility of the microorganism.¹ Flagellar length, shape, curvature, number, and arrangement on the cell are phenotypic characteristics used for classification of bacteria especially non-fermentative aerobic, gram-negative bacilli and anaerobes.² The Flagella Stain is a modification of the Ryu stain which is a simple, rapid method for demonstrating bacterial flagella.³

PRINCIPLE

Bacterial flagella are too thin to be seen with an optical microscope. Flagella Stain forms a precipitate at the surface of the flagella which increases their apparent width and makes them visible with the optical microscope.

REAGENTS (CLASSICAL FORMULA)*

Tannic acid (CAS 1401-55-4)	20.0 g
Aluminum Potassium Sulfate (CAS 7784-24-9)	13.2 g
Crystal Violet (CAS 548-62-9)	1.2 g
5% Carbolic Acid (CAS 108-95-2)	100.0 ml
Ethyl Alcohol 95% (CAS 64-17-5)	10.0 ml
Demineralized Water (CAS 7732-18-5)	100.0 ml

*Adjusted as required to meet performance standards.

PRECAUTIONS

CAUTION! May cause irritation to skin, eyes, and respiratory tract. Avoid breathing vapor and eye/skin contact. **WARNING!** Flammable. Keep away from heat, sparks, and flame.

This product is for *In Vitro* diagnostic use and should be used by properly trained individuals. Precautions should be taken against the dangers of microbiological hazards by properly sterilizing specimens, containers, and media after use. Directions should be read and followed carefully. Refer to Material Safety Data Sheet for detailed information on reagent chemicals.

STORAGE

This product is ready for use and no further preparation is necessary. Store product in its original container at room temperature until used.

PRODUCT DETERIORATION

This product should not be used if (1) the color has changed, (2) the expiration date has passed, or (3) there are other signs of deterioration.

SPECIMEN COLLECTION, STORAGE, AND TRANSPORT

Specimens should be collected and handled following recommended guidelines.⁴

MATERIALS REQUIRED BUT NOT SUPPLIED

(1) Loop sterilization device, (2) Inoculating loop, swab, collection containers, (3) Incubators, alternative environmental systems, (4) Supplemental media, (5) Quality control organisms, (6) Microscope, glass slides, (7) Demineralized water.

PROCEDURE

The test isolate should be a young culture (24-48 hours) growing on a nonselective medium without dextrose. The age of the culture will depend on the organism being tested.

1. Place a drop of water directly on an isolated colony of the test isolate and a second drop of water on a clean microscope slide.
2. Gently touch the drop of water on the colony with an inoculating loop, holding the loop in place for a few seconds to allow the organism to swim into the loop.

3. Gently touch the loop to the drop of water on the slide, taking care not to mix or otherwise cause the flagella to break off.
4. Cover the organism in suspension with a cover slip.
5. Place a drop of Flagella Stain at one end of the cover slip and allow the stain to flow under the cover slip.
6. Allow slide to sit at room temperature for 10-15 minutes.
7. Examine slides under the microscope at 100X.

INTERPRETATION

Positive test - Dark staining red to blue-black flagella
Negative test - No flagella seen

QUALITY CONTROL

All lot numbers of Flagella Stain have been tested using the following quality control organisms and found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

CONTROL

Pseudomonas aeruginosa
ATCC® 27853
Proteus mirabilis
ATCC® 12453

RESULTS

Polar flagellation
Peritrichous flagellation

LIMITATIONS

1. Because a low pH can damage flagella, the test isolate should be grown on a medium that does not contain dextrose (e.g., sheep blood agar).³
2. Standard microscope slides, precleaned by the manufacturer, are acceptable for use.³
3. The best flagellation occurs in the logarithmic and early stationary phases of growth, usually within 24-48 hours of growth.³




BIBLIOGRAPHY

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2. Summanen, P., E.J. Baron, D.M. Citron, C. Strong, H.M. Wexler, and S.M. Finegold. 1993. Wadsworth Anaerobic Bacteriology Manual. 5th ed. Star Publishing Co., Belmont, CA.
3. Kodaka, H., A.Y. Armfield, G.L. Lombard, and V.R. Dowell, Jr. 1982. J. Clin. Microbiol. 16:948-952.
4. Versalovic, J., K.C. Carroll, G. Funke, J.H. Jorgensen, M.L. Landry, and D.W. Warnock. 2011. Manual of Clinical Microbiology. 10th ed. ASM Press, Washington, D.C.

PACKAGING

REF R40041, 50 ml/BtlEach

Symbol Legend

REF	Catalog Number
IVD	In Vitro Diagnostic Medical Device
LAB	For Laboratory Use
	Consult Instructions for Use (IFU)
	Temperature Limitation (Storage Temp.)
LOT	Batch Code (Lot Number)
	Use By (Expiration Date)

CAS (Chemical Abstracts Service Registry No.)

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