

Instructions For Use GRS-IFU

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Reticulum Stain Kit

(Modified Gomori's)

Description and Principle

The Reticulum Stain Kit (Modified Gomori's) is intended for use in histological demonstration of reticular fibers. The main function of reticular fibers is to provide support. They are normally found throughout the body, particularly in liver, lymph node, spleen and kidney. Ammoniacal silver stains are a common method for demonstrating reticular fibers

Hexose sugars of reticular fibers are oxidized to aldehydes by a potassium permanganate solution. Ferric ammonium sulfate binds to reticulum and is replaced by silver from an ammoniacal silver solution. Impregnated silver is reduced to a visible metallic form with formalin and toned with gold chloride.

Expected Results

Reticulum: Black Nuclei: Red

| Kit Contents | Storage |
|--|----------------|
| Potassium Permanganate Solution | 18-25° C |
| 2. Sulfuric Acid Solution (1N) | 18-25° C |
| 3. Potassium Metabisulfite Solution (3%) | 18-25° C |
| 4. Ferric Ammonium Sulfate Solution | 18-25° C |
| 5. Silver Nitrate Solution (10%) | 2-8° C |
| 6. Potassium Hydroxide Solution (10%) | 18-25° C |
| 7. Formalin Solution (20%) | 18-25° C |
| 8. Gold Chloride Solution (0.2%) | 2-8° C |
| 9. Sodium Thiosulfate Solution (5%) | 18-25° C |
| 10. Nuclear Fast Red Solution | 18-25° C |

Suggested Controls (not provided)

Liver, Kidney, Lymph Node, Spleen.

Uses/Limitations

For In-Vitro Diagnostic use only. Do not use past expiration date. Use caution when handling reagents. Non-Sterile

Intended for FFPE sections cut at 5-10 µm.

This procedure has not been optimized for frozen sections.

Frozen sections may require protocol modification.

Storage

Mixed storage conditions. Store according to individual label instructions.

Safety and Precautions

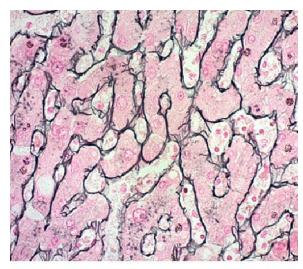
Please see current Safety Data Sheets (SDS) for this product and components GHS classification, pictograms, and full hazard/precautionary statements.

Required but not Included

Concentrated Ammonium Hydroxide Solution (25-30%)

Important Notes

- 1. All glassware used in this procedure should be chemically cleaned and rinsed thoroughly in distilled water.
- 2. Do $\underline{\text{not}}$ use metal forceps to remove slides from reagents. Use plastic forceps only.
- 3. Equilibrate all reagents to room temperature prior to use.



Reticular fibers Human Liver stained using Reticulum Stain Kit (Modified Gomori's) viewed at 40x

Preparation of Reagent Prior to Beginning:

- 1. Prepare working Acidified Potassium Permanganate Solution by mixing 2.5ml of Sulfuric Acid Solution (SAQ015) with 47.5ml of Potassium Permanganate Solution (PPD250). Mixed solution is stable for 2 days.
- 2. Prepare working Ammoniacal Silver Solution using chemically cleaned glassware in a chemical fume hood as follows:

Mix 2.5ml of Potassium Hydroxide Solution (PHC015) with 10ml of Silver Nitrate (10%) Solution (SNX065). Add concentrated ammonium hydroxide (25-30%); drop by drop, while swirling the flask continuously, until precipitate just dissolves. A few potassium hydroxide crystals will remain. Carefully add Silver Nitrate Solution (10%), drop by drop, until one drop causes the solution to become cloudy. Measure the resulting volume, dilute with an equal volume of distilled water. Filter into chemically cleaned stain jar.

Note: Use extreme care in preparation and use of Ammoniacal Silver Solution. Store Ammoniacal Silver Solution in a refrigerator to avoid the formation of explosive compounds. If Ammoniacal Silver Solution is exposed to sunlight, it will explode. Dispose of waste observing all local, state and federal laws.

Procedure:

- 1. Deparaffinize sections if necessary and hydrate to distilled water.
- 2. Place slide in working Acidified Potassium Permanganate Solution for 1 minute.
- 3. Rinse in 3 changes of distilled water.
- 4. Differentiate in Potassium Metabisulfite Solution for 1 minute.
- 5. Rinse in running tap water for 3 minutes.
- Rinse in distilled water.
- 7. Apply Ferric Ammonium Sulfate Solution for 30 seconds.

- 8. Immediately rinse slides in running tap water for 2 minutes.
- 9. Rinse in 2 quick changes of distilled water.
- 10. Apply working Ammoniacal Silver Solution for 1 minute.
- 11. Rinse quickly in 3 changes of distilled water.
- 12. Place slide in 20% formalin for 3 minutes.
- 13. Rinse in running tap water for 3 minutes.
- 14. Rinse in 2 changes of distilled water.
- 15. Apply Gold Chloride Solution for 2-5 minutes.
- 16. Rinse in 2 changes of distilled water.
- 17. Apply Sodium Thiosulfate Solution for 1-2 minutes.
- 18. Rinse in tap water for 2 minutes.
- 19. Counterstain using Nuclear Fast Red Solution for 5 minutes.
- 20. Rinse in tap water.
- 21. Rinse in distilled water.
- 22. Dehydrate through graded alcohols.
- 23. Clear, and mount in synthetic resin.

References

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