



# Instructions For Use

## GRT-IFU

205 South 600 West Logan, Utah 84323, U.S.A. – Tel. (800) 729-8350 – Tel. (435) 755-9848 – Fax (435) 755-0015 – www.scytek.com Rev. 5, 12/5/2022

## Reticulum Stain Kit

### Description and Principle

The Reticulum Stain Kit 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 potassium permanganate. Ferric ammonium sulfate binds to reticulum and is replaced by silver from an ammoniacal silver solution. Impregnated silver is then reduced to a visible metallic form with formalin and toned with gold chloride.

### Expected Results

Reticulum: Black  
Background: Green

### Kit Contents

	<b>Storage</b>
1. Potassium Permanganate Solution (1%)	18-25° C
2. Potassium Metabisulfite Solution (3%)	18-25° C
3. Ferric Ammonium Sulfate Solution (3%)	18-25° C
4. Formalin Solution (20%)	18-25° C
5. Gold Chloride Solution (0.1%)	2-8° C
6. Sodium Thiosulfate Solution (5%)	18-25° C
7. Light Green Solution	18-25° C
8. Sodium Hydroxide Solution (3%)	18-25° C
9. Silver Nitrate Solution (10%)	2-8° 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 5-10µm.

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:

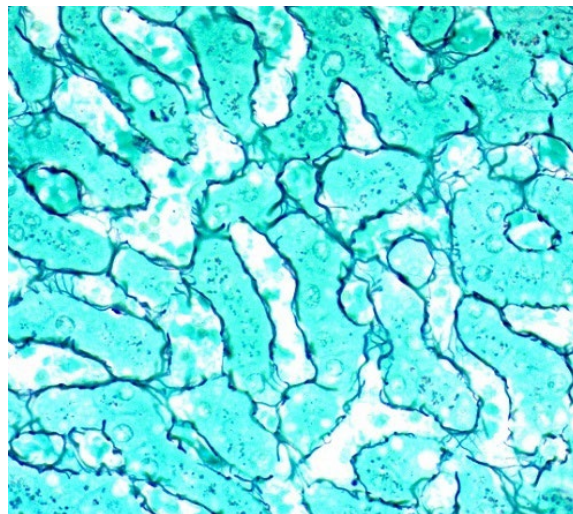
Concentrated Ammonium hydroxide Solution (25-30%)

### Important Notes

1. All glassware used in this procedure should be chemically cleaned and rinsed thoroughly in distilled water.

2. Do 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 in Human Liver stained with Reticulum Stain Kit.  
Viewed at 400X magnification.

### Preparation of Reagent Prior to Beginning:

**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.

1. Prepare working Ammoniacal Silver Solution using chemically cleaned glassware in a chemical fume hood as follows:

Add one (10ml) vial of Silver Nitrate Solution to 17ml of Distilled Water and mix completely. Add 1ml of Concentrated Ammonium hydroxide (25-30%) (Not Included in Kit) while continuously mixing. The mixture will initially turn brown and then become clear. Add 10ml of Sodium Hydroxide Solution (3%) and mix completely. If the solution does not remain colorless, add Concentrated Ammonium hydroxide (25-30%) drop by drop until no precipitate remains. Add Distilled Water to a total volume of 60ml and mix completely. Solution is now ready for use.

### Procedure:

1. Deparaffinize sections if necessary and hydrate to distilled water.
2. Place slide in Potassium Permanganate Solution (1%) for 5-10 minutes.
3. Rinse in 3 changes of distilled water.
4. Differentiate in Potassium Metabisulfite Solution (3%) until section is transparent.
5. Rinse in 3 changes of distilled water.
6. Apply Ferric Ammonium Sulfate Solution (3%) for 10 minutes.
7. Rinse in 2 quick changes of distilled water.
8. Apply working Ammoniacal Silver Solution for 2-3 minutes.

9. Rinse in 3 changes of distilled water.
10. Place slide in Formalin Solution (20%) for 1 minute.
11. Rinse in 3 changes of distilled water.
12. Apply Gold Chloride Solution (0.1%) for 3-5 minutes.
13. Rinse in 2 changes of distilled water.
14. Apply Sodium Thiosulfate Solution (5%) for 1-2 minutes to remove unreduced silver.
15. Rinse in tap water for 2 minutes.
16. Counterstain using Light Green Solution for 2-5 minutes.
17. Rinse quickly in Absolute Alcohol.
18. Dehydrate through 3 changes of Absolute Alcohol.
19. Clear, and mount in synthetic resin.

### **References**

1. Gomori, G., A Modification of the Silver Impregnation Method of Staining Reticular Fibers. American Journal of Clinical Pathology, Volume 21, Pages 897-899, 1951.



SeyTek Laboratories, Inc.  
205 South 600 West  
Logan, UT 84321  
435-755-9848  
U.S.A.



Emergo Europe  
Prinsessegracht 20  
2514 AP The Hague, The Netherlands