

Instructions For Use  
**KAA-1-IFU**

Rev. Date: Jan. 10, 2006

Revision: 1

Page 1 of 4

P.O. Box 3286 - Logan, Utah 84323, U.S.A. - Tel. (800) 729-8350 - Fax (435) 755-0015 - [www.scytek.com](http://www.scytek.com)

## GMS Stain Kit

(Modified Gomori Methenamine-Silver Nitrate Stain for Fungus and  
Pneumocystis carinii)

### Description:

The Modified Gomori Methenamine-Silver Nitrate Stain (GMS Stain Kit) is intended for use in the histologic visualization of fungi, basement membrane and some opportunistic organisms such as *Pneumocystis carinii*. *Pneumocystis carinii* is an opportunistic pathogen that causes severe pulmonary disease in humans, dogs, rats, mice and other vertebrate species with acquired, induced, or inherited immune deficiency syndromes. In addition, this procedure will demonstrate *Actinomyces* and related species, *Nocardia* asteroid, and certain encapsulated bacteria.

Fungi: Black  
P. Carinii: Black  
Mucin: Gray  
Mycelia (inner): Rose  
Hyphae (inner): Rose  
Background: Light Green

### Uses/Limitations:

For In-Vitro Diagnostic use only.  
Histological applications.  
Air-dried smears.  
Do not use past expiration date.  
Use caution when handling these reagents.

### Availability/Contents:

<u>Item #</u>	<u>Kit Contents</u>	<u>Storage Conditions</u>
SNS-125	Silver Nitrate (125 ml)	2-8° Centigrade
MET-125	Methenamine (125 ml)	2-8° Centigrade
GCB-100	Gold Chloride (100 ml)	2-8° Centigrade
BOR-15	Borax Solution (15 ml)	Room Temperature
SBC-100	Sodium Bisulfite (100 ml)	Room Temperature
CHR-100	Chromic Acid (100 ml)	Room Temperature
STG-100	Sodium Thiosulfate (100 ml)	Room Temperature
FGN-100	Fast Green Solution (100 ml)	Room Temperature

Instructions For Use  
KAA-1-IFU

Rev. Date: Jan. 10, 2006

Revision: 1

Page 2 of 4

P.O. Box 3286 - Logan, Utah 84323, U.S.A. - Tel. (800) 729-8350 - Fax (435) 755-0015 - [www.scytek.com](http://www.scytek.com)

**Precautions:** Chromic Acid is a strong oxidizer. Contact with other material may cause fire. Contact with skin and eyes can cause burns. Harmful if swallowed. Harmful to respiratory and gastrointestinal system. Can cause cancer and birth defects.

Borax solution is irritating to skin, eyes, and respiratory system. Target organs are central nervous system and kidneys. Harmful if swallowed. Possible risk to unborn child.

Handle all components with care, wearing gloves and eye protection.

ScyTek Laboratories may not be held liable for injury due to mishandling.

Follow all Federal, State, and local regulations regarding disposal.

Use in chemical fume hood whenever possible.

**Important Notes:**

1. All glassware used in this procedure should be chemically cleaned and rinsed thoroughly in distilled water.
2. Failure to adequately remove the alcohol used in deparaffination will result in reduction of the chromic acid solution. Reduction of the chromic acid solution will result in a change in color from orange to brown. Discard the reagent if color change is noted.
3. Do not use metal forceps to remove slides from reagents. Use plastic forceps only.
4. Prewarm all reagents to room temperature prior to use.

**Procedure (Standard):**

1. Deparaffinize sections if necessary and hydrate to distilled water.
2. Incubate slide in Chromic Acid solution for 10 minutes.
3. Rinse in tap water followed by 2 changes of distilled water.
4. Incubate slide in Sodium Bisulfite solution for 1 minute (to remove any residual chromic acid).
5. Rinse in tap water followed by 2 changes of distilled water.
6. Combine the following for a working GMS solution:

25 ml Silver Nitrate  
25 ml Methenamine  
2 ml Borax Solution

Note: Mixed solution may not be stored for reuse later.

## Instructions For Use KAA-1-IFU

Rev. Date: Jan. 10, 2006

Revision: 1

Page 3 of 4

P.O. Box 3286 - Logan, Utah 84323, U.S.A. - Tel. (800) 729-8350 - Fax (435) 755-0015 - [www.scytek.com](http://www.scytek.com)

7. Place working GMS solution in 60° centigrade water bath and allow temperature to equilibrate.
8. Incubate slide in working GMS solution for 10-15 minutes. Using plastic forceps, dip slide in distilled water and check under a microscope for evaluation of silver impregnation. Fungi should be dark brown. If color is not sufficient, return the slide to working GMS solution for 2-3 minutes and check again.
9. Rinse in 4 changes of distilled water.
10. Incubate slide in Gold Chloride solution for 15-30 seconds.
11. Rinse in 4 changes of distilled water.
12. Incubate slide in Sodium Thiosulfate for 2 minutes.
13. Rinse in tap water followed by 2 changes of distilled water.
14. Incubate slide in Fast Green Solution for 2 minutes.
15. Rinse in distilled water.
16. Dehydrate through graded alcohols.
17. Clear, and mount in synthetic resin.

### Procedure (Microwave):

*Note: These instructions were developed using a standard 500 watt microwave oven. Heating times should be modified as needed depending on the microwave oven used.*

1. Deparaffinize sections if necessary and hydrate to distilled water.
2. Place slide in plastic coplin jar filled with Chromic Acid solution. Cap jar loosely!
3. Place jar in microwave oven and heat on high power for 10 seconds. Allow slide to remain in warm solution for 3 minutes.
4. Rinse in tap water followed by 2 changes of distilled water.
5. Incubate slide in Sodium Bisulfite solution for 1 minute (to remove any residual chromic acid).
6. Rinse in tap water followed by 2 changes of distilled water.

## Instructions For Use KAA-1-IFU

Rev. Date: Jan. 10, 2006

Revision: 1

Page 4 of 4

P.O. Box 3286 - Logan, Utah 84323, U.S.A. - Tel. (800) 729-8350 - Fax (435) 755-0015 - [www.scytek.com](http://www.scytek.com)

7. Combine the following for a working GMS solution:
- |                      |
|----------------------|
| 25 ml Silver Nitrate |
| 25 ml Methenamine    |
| 2 ml Borax Solution  |

Note: Mixed solution may not be stored for reuse later.

8. Place working GMS solution (loosely capped) in microwave oven for 40 seconds. Remove and pour several times between coplin jar and a clear graduated cylinder to mix thoroughly (use protective glove to avoid burning hand). Mixed solution remains in coplin jar.
9. Incubate slide in working GMS solution (heated) for 2-6 minutes until the tissue is medium brown in color. Using plastic forceps, dip slide in distilled water and check under a microscope for evaluation of silver impregnation. Fungi should be dark brown. If color is not sufficient, return the slide to working GMS solution for 1-2 minutes and check again. Reheat solution if needed.
10. Rinse in 4 changes of distilled water.
11. Incubate slide in Gold Chloride solution for 15-30 seconds.
12. Rinse in 4 changes of distilled water.
13. Incubate slide in Sodium Thiosulfate for 2 minutes.
14. Rinse in tap water followed by 2 changes of distilled water.
15. Incubate slide in Fast-Light Green for 2 minutes.
16. Rinse in distilled water.
17. Dehydrate through graded alcohols.
18. Clear, and mount in synthetic resin.

### References:

1. McManus, J.F.A. and Mowry, R. 1955. Staining Methods and Histologic and Histochemical. Grocott, pp 194-197.
2. Koski, J.P. 1981. Silver methenamine-borate (SMB); Cost reduction with technical improvement in silver nitrate-gold chloride impregnation's. Journal of Histotechnology 4:115.
3. Procop, G.W. et al. 2004. Detection of *Pneumocystis jiroveci* in Respiratory Specimens by Four Staining Methods. Journal of Clinical Microbiology. July 2004, Vol. 42, No. 7, pp 3333-3335.
4. Raab, S.S. et al. 1994. Utility of Gomori methenamine silver stains in bronchoalveolar lavage specimens. Modern Pathology, June 1994, Vol. 7, No. 5, pp 599-604.
5. Sale, G.E. 1978. Rapid Methenamine Silver Stain. Arch Path Lab Med, 1978, 102, pp 351-352.
6. Sheehan, D.C., Hrapchak, B.B. 1980. Theory and Practice of Histotechnology, 2<sup>nd</sup> edition, CV Mosby Company, St. Louis, MO.