SUBJECT: GARVEY STAIN FOR SENILE PLAQUE AND NEUROFIBRILLARY TANGLES

I. PRINCIPLE (Purpose):
This method is based on the enhancement of some silver techniques by ammonia. It consists of sensitization in Ammoniacal Silver Nitrate, followed by reduction in a physical developer consisting of Silver Nitrate, Hydroquinone and Gum Mastic.

II. ROLE:
A. Research Analyst

V. SPECIMENS (Samples):
A. 10% Neutral Buffered Formalin fixed tissue, cut paraffin sections at 8-10 microns
B. Brain- quality control

VI. MATERIALS, REAGENTS, EQUIPMENT and SAFETY PRECAUTIONS:
List all materials etc used, with order number and expiration date where applicable as well as any specific safety requirements.

A. Equipment
1. Coplin Jars
2. Forceps
3. Stir plate
4. Magnetic stir bar
B. Reagents

1. ALCOHOLIC AMMONIA SOLUTION
Storage: Make fresh before use.
95% Ethanol……………………………………45.0 ml
Ammonium Hydroxide, concentrate………………5.0 ml

2. 1% SILVER NITRATE SOLUTION
Storage: Make fresh before use.
Silver Nitrate……………………………………0.25 gm
Distilled Water…………………………………25.0 ml

3. AMMONIACAL SILVER NITRATE SOLUTION
Storage: Make fresh before use.
1% Silver Nitrate Solution…………………………20.0 ml
add Ammonium Hydroxide Concentrate drop by drop, stirring after each drop, until the precipitate is dissolved. This usually takes 5 to 10 drops. Then add
1% Silver Nitrate Solution…………………….0.75 ml
add distilled water to this solution to make 40 ml.

4. 2.5 % GUM MASTIC SOLUTION
Storage: In refrigerator, in glass bottle.
Gum Mastic……………………………………….2.5 gm
Absolute Alcohol………………………………..100.0 ml

5. 1.6 % HYDROQUINONE SOLUTION
Storage: Make fresh before use.
Hydroquinone……………………………………0.4 gm
Distilled Water………………………………….25.0 ml

6. REDUCING SOLUTION
Storage: Make fresh before use.
Hydroquinone solution……………………….25.0 ml
Gum Mastic solution…………………………15.0 ml
1% Silver Nitrate solution…………………..1.0 ml, add just before use.

7. 1% GOLD CHLORIDE SOLUTION
Storage: In refrigerator, in glass bottle.
Gold Chloride……………………………………1.0 gm
Distilled Water………………………………..100.0 ml

8. 5% SODIUM THIOSULFATE SOLUTION
Storage: At room temperature, in glass bottle.
Sodium Thiosulfate……………………………5.0 gm
Distilled Water………………………………..100.0 ml

VII. QUALITY CONTROL
A. Use brain as positive control.
   1. Senile plaque, neurofibrillary tangles………………brown-black
   2. Background………………………………………colorless
B. Quality control check according to BCP H.15 Quality Control of Histologic Stains
X. PROCEDURE

A. Deparaffinize and hydrate to distilled water.
B. Pretreat sections in Alcoholic Ammonia for 10 minutes.
C. Rinse in distilled water.
D. Sensitize slides in Ammoniacal Silver Nitrate for 15 seconds. In a 600 watt microwave oven and allow to stand for 1 minute. Return slides to the microwave for 10 seconds and allow standing for another minute.
E. Rinse in two changes of distilled water.
F. Dehydrate to 100% Ethanol.
G. Treat with Gum Mastic for 5 minutes.
H. Quickly remove slides from Gum Mastic and rinse in distilled water.
I. Place in Reducing Solution. Reduce in microwave oven for 10 seconds.
J. Remove slides from the microwave and check the slides under the microscope.
K. If the nerve is black, place the slides in 100% Ethanol. If not, return slides to the Reducing Solution and check every 30 seconds until nerve is black.
L. Rinse in distilled water.
M. Tone slides in Gold Chloride until sections turn brown to gray.
N. Place in Sodium Thiosulfate for 1 minute.
O. Dehydrate in two changes each of 95% Ethanol and 100% Ethanol. Clear in two changes of Xylene.
P. Mount in synthetic resin.

XIII. REFERENCES:

XV. HISTORY BLOCK

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XVI. APPROVAL BLOCK:

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Date this version is removed from the manual:

Date the procedure is retired: