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Part 4540
Revised December 2015

# Periodic Acid Schiff (PAS) Glycogen Control Slides – Technical Memo

**CONTROL SLIDES:** **Part 4540A** 10 Slide/Set **Part 4540B** 98 Slide/Set

**Periodic Acid Schiff (PAS) Glycogen Control Slides contain a section of positive staining liver.**

**PRODUCT DESCRIPTION:**
The enclosed positive control slides are intended to be used to verify histological techniques and reagent reactivity. These slides are to be used for the qualitative purpose of determining positive or negative results, and are not intended to be used for any quantitative purpose. The first serial section within the control box is stained and provided for your reference. ***Before using the unstained slides, review the enclosed stained slide with your pathologist to ensure that this tissue source is acceptable. Newcomer Supply will not accept a return with missing slides in the series. Newcomer Supply guarantees reactivity of these control slides for one year from the date of receipt. Revalidate after one year to verify continued reactivity. Store at 15-30°C in a light deprived and humidity controlled environment.***

These positive control slides were produced from human surgical or autopsy tissues under carefully controlled conditions. Reasonable measures are used to deliver quality control slides that are as consistent as possible. However, characteristics of quality control slides may be dissimilar due to variations in the reagents, stains, techniques, laboratory conditions, and tissue sources used. Newcomer Supply Laboratory uses a manual method of performing quality control procedures, specifically avoiding automation, in order to provide reactive control slides for even less aggressive methods of staining that our customers may be using.

**CONTROL SLIDE VALIDATION:**

| **With Periodic Acid Schiff (PAS) Stain Kit:** | **Part 9162A/B** | **Individual Stain Solution** |
|---|---|---|
| Solution A: Periodic Acid 0.5%, Aqueous | 250/500 ml | Part 13308 |
| Solution B: Schiff Reagent, McManus | 250/500 ml | Part 1371 |
| Solution C: Hematoxylin Stain, Harris | 250/500 ml | Part 12013 |
| Solution D: Acid Alcohol 1% | 250/500 ml | Part 10011 |
| Solution E: Lithium Carbonate, Saturated Aqueous | 250/500 ml | Part 12215 |
| α-Amylase (for glycogen digestion) | | |
| Phosphate Buffer, pH 6.0 (for glycogen digestion) | | Part 13312 |

***For storage requirements and expiration date refer to individual product labels.***

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**APPLICATION:**

Newcomer Supply Periodic Acid Schiff (PAS) Glycogen Control Slides are for the positive histochemical staining of glycogen in tissue sections, and can also be utilized as control slides for glycogen digestion steps and further identification of mucosubstances.

**METHOD:**

**Fixation:** Formalin 10%, Phosphate Buffered (Part 1090)
**Technique:** Paraffin sections cut at 5 microns on Superfrost® Plus
- Additional control and patient slides needed for digestion steps.

**Solutions:** All solutions manufactured by Newcomer Supply, Inc.

**NEWCOMER SUPPLY VALIDATION PROCEDURE:**

1. Deparaffinize sections thoroughly in three changes of xylene, 3 minutes each. Hydrate through two changes each of 100% and 95% ethyl alcohols, 10 dips each. Wash well with distilled water.
   a. *See Procedure Notes #1 and #2 (page 2).*
2. Digestion Step: Proceed to Step #5 if not running Digestion.
   a. *Two control slides and two patient slides are needed. Label one control slide and one patient slide **"with"**; label the other control slide and patient slide **"without".***
   b. *Prepare Amylase Digestion Solution and mix well.*
      α-Amylase *0.05 gm*
      *Phosphate Buffer, pH 6.0 (13312) 50 ml*
   c. *Prepare separate Coplin jar of Phosphate Buffer, pH 6.0.*
   d. *Preheat both solutions from Steps #2b and #2c to 37°C.*
   e. *Place slides labeled **"with"** in preheated Amylase Digestion Solution and slides labeled **"without"** in preheated Phosphate Buffer, pH 6.0. Incubate both for 60 minutes at 37°C.*
   f. *Proceed to Step #4.*
3. **Microwave Modification:** *See Procedure Note #3 (page 2).*
   a. *Follow Steps #2a through #2c.*
   b. *Place slides labeled **"with"** in a plastic Coplin jar containing the Amylase Digestion Solution and slides labeled **"without"** in a plastic Coplin jar containing Phosphate Buffer, pH 6.0 and microwave both for 1 minute at 37°C.*
4. Wash all slides in running tap water for 5 minutes; rinse in distilled water. Combine slides for remaining steps.
5. Place in Solution A: Periodic Acid 0.5%, Aqueous for 10 minutes.
6. Wash in three changes of tap water; rinse in distilled water.
7. Place in Solution B: Schiff Reagent, McManus for 20 minutes.
8. Wash in lukewarm tap water for 5-10 minutes.
9. Stain with Solution C: Hematoxylin Stain, Harris, 1 to 5 minutes, depending on preference of nuclear stain intensity.
10. Wash in tap water for 2-3 minutes.
11. Differentiate in Solution D: Acid Alcohol 1%; 1-2 quick dips.
12. Wash in tap water for 1 minute.
13. Blue sections in Solution E: Lithium Carbonate, Saturated Aqueous; 3-4 dips.
14. Wash in several changes of tap water; rinse in distilled water.
15. Dehydrate in two changes each of 95% and 100% ethyl alcohol. Clear in three changes of xylene, 10 dips each; coverslip with compatible mounting medium.

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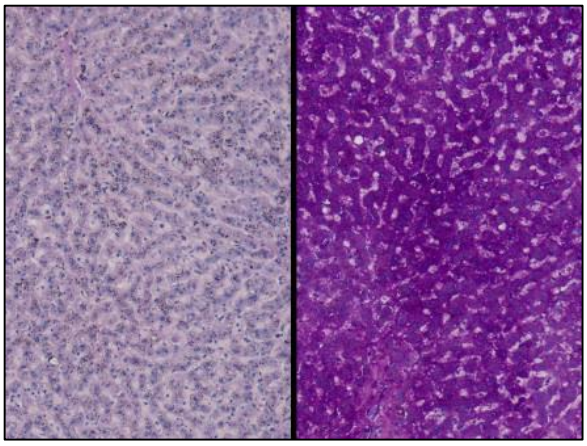
**RESULTS:**

| | |
|---|---|
| Glycogen digestion | Absence of magenta |
| Glycogen | Magenta |
| Acid & neutral epithelial mucin | Magenta |
| Nuclei | Blue |



*With and without glycogen digestion*

**PROCEDURE NOTES:**

1. Drain staining rack/slides after each step to prevent solution carry over.
2. Do not allow sections to dry out at any point during staining procedure.
3. The suggested microwave procedure has been tested at Newcomer Supply using an "EB Sciences", 850 watt microwave oven with temperature probe and agitation tubes. This procedure is reproducible in our laboratory. It is nonetheless a guideline and techniques should be developed for your laboratory which meet the requirements of your situation. Microwave devices should be placed in a fume hood or vented into a fume hood, according to manufacturer's instructions, to prevent exposure to chemical vapors.
4. Sigma α-Amylase from Porcine Pancreas (A3176) is the α-Amylase used in the digestion steps.
5. Newcomer Supply Schiff Reagent, McManus is stored at room temperature. There is no benefit to store this product at 4°C.
6. If using a xylene substitute, closely follow the manufacturer's recommendations for deparaffinization and clearing steps.

**REFERENCES:**

1. Bancroft, John D., and Marilyn Gamble. *Theory and Practice of Histological Techniques*. 6th ed. Oxford: Churchill Livingstone Elsevier, 2008. 168-171, 180.
2. Carson, Freida L., and Christa Hladik. *Histotechnology: A Self-Instructional Text*. 3rd ed. Chicago, Ill.: American Society of Clinical Pathologists, 2009.137-141.
3. Sheehan, Dezna C., and Barbara B. Hrapchak. *Theory and Practice of Histotechnology*. 2nd ed. St. Louis: Mosby, 1980. 164-168, 245.
4. Modifications developed by Newcomer Supply Laboratory.