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Part 4540
Revised January 2020

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

CONTROL SLIDES:

	Part 4540A	**Part 4540B**
	10 Slide/Set	98 Slide/Set

PRODUCT SPECIFICATIONS:
Tissue: Positive staining liver.
Fixation: Formalin 10%, Phosphate Buffered (Part 1090).
Section/Glass: Paraffin sections cut at 4 microns on Superfrost™ Plus slides.
Quality Control Stain: PAS quality control stained slide(s) included.
Reactivity: Guaranteed product specific reactivity for one year from date of receipt. Revalidate after one year to verify continued reactivity.
Storage: 15-30°C in a light deprived and humidity controlled environment.
Intended Use: To verify histological techniques and reagent reactivity.

Before using unstained control slides, review the enclosed stained slide(s) to ensure that this tissue source is acceptable for testing needs.

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

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.

PRESTAINING PREPARATION:

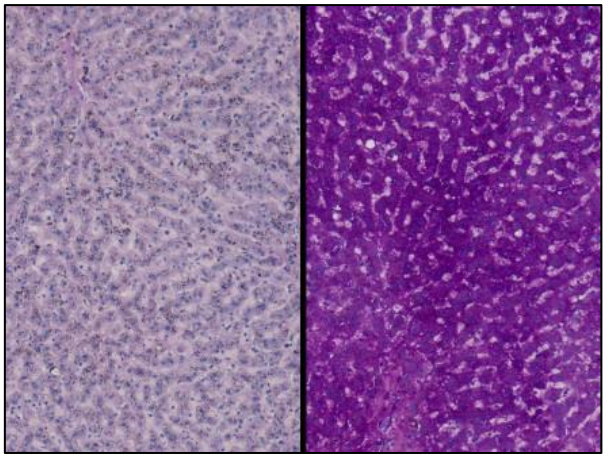
1. Additional control and patient slides required for digestion steps.
2. Heat dry sections in oven according to your laboratory protocol.

NEWCOMER SUPPLY VALIDATION PROCEDURE:

3. 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).*
4. Digestion Step: Proceed to Step #7 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 #4b and #4c to 37°C.*
   e. *Place slides labeled **"with"** in preheated Amylase Digestion Solution and slides labeled **"without"** in preheated Phosphate Buffer, pH 6.0.*
   f. *Incubate both for 60 minutes at 37°C.*
   g. *Proceed to Step #6.*
5. **Microwave Modification:** *See Procedure Note #3 (page 2).*
   a. *Follow Steps #4a through #4c.*
   b. *Place slides labeled **"with"** in a plastic Coplin jar containing the Amylase Digestion Solution. Place slides labeled **"without"** in a plastic Coplin jar containing Phosphate Buffer, pH 6.0 and microwave both for 1 minute at 37°C.*
6. Wash all slides in running tap water for 5 minutes; rinse in distilled water. Combine slides for remaining steps.
7. Place in Solution A: Periodic Acid 0.5%, Aqueous for 10 minutes.
8. Wash in three changes of tap water; rinse in distilled water.
9. Place in Solution B: Schiff Reagent, McManus for 20 minutes.
10. Wash in lukewarm tap water for 5-10 minutes.
11. Stain with Solution C: Hematoxylin Stain, Harris, 1 to 5 minutes, depending on preference of nuclear stain intensity.
12. Wash in tap water for 2-3 minutes.
13. Differentiate in Solution D: Acid Alcohol 1%; 1-2 quick dips.
14. Wash in tap water for 1 minute.
15. Blue in Solution E: Lithium Carbonate, Saturated Aqueous; 3-4 dips.
16. Wash in several changes of tap water; rinse in distilled water.
17. 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.

RESULTS:

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



With and without glycogen digestion

PROCEDURE NOTES:

1. Drain slides after each step to prevent solution carry over.
2. Do not allow sections to dry out at any point during procedure.
3. The suggested microwave procedure has been tested at Newcomer Supply. This procedure is a guideline and techniques should be developed for use in your laboratory.
4. Sigma α-Amylase from Porcine Pancreas (A3176) is the α-Amylase used in the digestion steps.
5. 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.