S0330 Schiff's Reagent GCC Diagnostics

USE

Schiffs Reagent is generally used to demonstrate PAS [Periodic acid-Schiff] reaction on leukocytes and muccopolysaccharides. Lymphocyte staining with PAS reaction is useful in reaching therapeutic decisions in cases of Lymphocytic leukaemia.
PAS is widely used to demonstrate muccopolysaccharides in tissue sections.
With Diastase digestion the PAS reaction finds use as an aid in the diagnosis of glycogen storage disease.
Fungal organisms can also be demonstrated in sections using Schiffs reagent.
A standard tecnique is given below as example.

PRINCIPLE OF TEST [PAS]

Treatment with Periodic acid oxidises glycols to aldehydes. Reaction with Schiffs reagent stains the glycol containing cellular components. Reaction can be carried out on blood, bone marrow smears, sections or tissue touch preps.
When used on blood/bone marrow smears, this reaction may be useful in recognising some cases of Erythroleukaemia and acute lymphoblastic leukaemia.
Diastase hydrolyses starch, glycogen and degradation products originating in these polysaccharides present in tissue. These digestion by-products are rinsed away prior to staining.

ADDITIONAL REAGENTS REQUIRED

1 - Periodic acid solution [Corrosive]
2 - Haematoxylin counterstain Gill No. 3
3 - Formaldehyde 37-38%
4 – Ethanol 95% (IMS)
5 – Diastase (a-amylase)

Reagents provided are for 'In-Vitro Diagnostic use only'. Use this product in a well ventilated place or in a fume hood. Wear eye & skin protection when in use. Standard precautions in the handling of laboratory reagents should be followed.

STORAGE AND STABILITY

Store Schiffs reagent in packaging provided at cool room temperature <12 C in the dark.
Note expiry date on pack.
Reagent are suitable for use only if: Schiffs reagent is water white. If product has deteriorated before expiry date please contact us for replacement.

SPECIMEN COLLECTION AND STORAGE

All blood derivatives should be considered potentially infectious. Take appropriate precautions.
Use freshly prepared whole, EDTA or heparinized blood or bone marrow smears. Fix as soon as possible.

For polysaccharides, fix tissue in 10% NBF, Zenkers or Bouin's fixative. Some carbohydrates are water soluble, prolonged fixation in aqueous fixatives may lead to their loss.

For Glycogen, use alcoholic formalin or Carnoys fluid as fixative.
Picric acid containing fixatives will increase time required for diastase digestion.

REAGENT PREPARATION AND SET-UP

1 - Fixative, add 5ml of 37% Formaldehyde to 45ml 95% Ethanol (IMS).

2 - Beaker or Coplin jar of Periodic acid, return to bottle after use. Use 4-6- times and discard.

3 – Coplin jar of Schiffs reagent, return to bottle after use. Use 2-3 times and discard.

4 - Beaker haematoxylin Gill No 3, return to bottle after use. Use until reagent no longer performs acceptably.

5 – Beaker (200ml) de-ionised water (2-3 changes required).

TECHNIQUE [Blood, bone marrow, tissue touch preps]

1 - Fix air dried smears for 1 minute at room temperature in Fixative solution.
2 - Rinse slides for 1 minute in slowly running tap water.
3 - Transfer to Periodic acid reagent for 5 minutes at room temperature.
4 - Rinse in 2-3 changes of de-ionised water.
5 - Transfer to Colpin jar (Schiffs reagent), keep lid on . Stain for 15 minutes at room temperature.
6 - Rinse in running tap water for 3-5 minutes.
7 - Counterstain in Haematoxylin Gill No 3 for 1-2 minutes.
8 - Rinse slides in running tap water for a **few seconds only**.
9 - Air dry and examine or mount in DPX or similar and examine under oil immersion.

TECHNIQUE [Tissue Sections]

1 - Deparaffinize and bring sections to water.
2 - Transfer to periodic acid reagent for 5 minutes at room temperature
3 - Rinse in several changes of de-ioninsed water.
4 - Transfer to Coplin jar (Schiffs reagent), keep lid on. Stain for 15 minutes at room temperature.
5 – Rinse in running tap water for 3-5 minutes.
6 - Counterstain in Haematoxylin Gill No 3 for 1-2 minutes.
7 - Rinse in running tap water.
8 - Dehydrate, clear, mount in DPX or similar and examine.

DIASTASE DIGESTION [microwave method]

1 – Use a normal undigested control slide with test slide. Label one for digestion and one for PAS only.
2 - Deparaffinize and bring to water.

3 - To a Plastic Coplin jar add 0.2g a-Amylase and 40ml de-ionised water. Dissolve.
4 – Immerse slide in Coplin jar and microwave at 600 watts for 25 seconds.
5 – Rinse digested slide in running tap water for 5 minutes.
6 – Process both digested and undigested slides for PAS reaction as above under Tissue Section method.

QUALITY CONTROL

Smears prepared from healthy individuals may be included for control purposes.
Tissue section known to be PAS positive and/or containing glycogen should be included each time a section is stained.

EXPECTED OBSERVATIONS.

Normal – Early myeloid precursors do not stain. Diffuse and granular staining increases with maturation. Erythrocyte precursors do not stain. Megakaryocytes and platelets stain strongly. Monocytes stain faintly and may show fine granules.

Erythroleukaemia – Early erythroid precursors may show intense cytoplasmic granular staining. Diffuse staining may be present in more mature nucleated cells.

Acute Lymphoblastic Leukaemia – PAS staining is variable. In many cases, some precursor cells show Coarse granules of block-like positive reaction.

Acute Granulocytic Leukaemia – Myeloblasts are usually negative, although faint reaction product may occasionally be observed.

Tissue Sections – PAS positive material in tissue stain pink and nuclei blue. Diastase digested slides will have no obvious PAS staining of glycogen when compared to the undigested glycogen positive control slide.

SAFETY DATA SHEET SECTION

[PAS reaction reagents]

Use these reagents in a ventilated area / hood and wear eye and skin protection when using. Do not consume these reagents.

Periodic acid , contains < 1% w/v periodic acid (aq). Irritant and may cause burns to eyes and sensitive skin. If in contact with eyes treat with eye-bath for 5 minutes.
If in contact wash with soap and water.
Schiffs Reagent – Harmful and corrosive. Contains < 2% HCl. Harmful by ingestion. Harmful in contact with eyes, may cause irritation / burns. Harmful by inhalation (vapour of SO2 Sulphur dioxide).If inhaled and you feel unwell move to a clean air area and rest. If in contact with eyes treat eye-bath for 5 minutes. If in contact with skin wash immediately with plenty of soap and water.. Seek medical attention if soreness persists. Contains HCl and SO2.
Haematoxylin Gill No 3 – Harmful by ingestion. Contains haematoxylin dye and aluminium salts.

R: 20, 21, 22, 36, 38 S: 20/21, 24/25, 36/37

For more complete information on Health & Safety, Storage, Fire-fighting, Transport , Spillage etc please read the MSDS for this product.

ACCIDENTAL SPILLAGE & WASTE DISPOSAL

In the volumes supplied in the kit and when in use, this product is unlikely to present a serious spillage risk. However, the following information is provided to deal with any spillage or disposal problem that may arise.

Periodic acid reagent – mop up with cloth and wash are down with water or a solution of sodium carbonate in water to neutralise the acidic effect of the acid. Dispose of spillage or waste reagent down the public sewer diluting greatly with water.

Schiffs Reagent – same treatment as Periodic acid.

Haematoxylin Gill No 3. – Same treatment as Periodic acid.

UNSATISFACTORY PERFORMANCE

As part of our duty to monitor product performance and our policy of continual improvement. Please report to us any unsatisfactory performance you may experience with this product. If any reagent degrades before expiry date of shelf life we will replace that reagent free of charge.

GCC Diagnostics guarantees that the highest quality reagents are supplied with this product and that the product conforms to the information contained in this leaflet.
The user should however, determine the suitability of this product for their particular use.
If you wish to report any findings to us or if you require help or further information on the use of this product please contact us.

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