

Masson Fontana Stain Kit

PRODUCT INFORMATION:

REF

SSP026 25 Reactions

SSP026 50 Reactions

PERFORMANCE CHARACTERISTICS:

Staining Interpretation:

Melanin : Black

Nuclei : Pink

SUMMARY AND EXPLANATION

For laboratory use only

This reagent is intended for use in qualitatively identifying melanin pigment by light microscopy in formalin-fixed, paraffin-embedded (FFPE) tissue sections using histochemical staining methodology. The interpretation of any positive or negative staining must be complemented by the evaluation of proper known controls (positive and negative). It must be made within the context of the patient's clinical history and other diagnostic tests. A qualified and trained pathologist/personnel must evaluate the test.

PRINCIPLE OF THE PROCEDURE

The Masson Fontana stain is based on the ability of melanin pigment to reduce silver nitrate (AgNO_3) to metallic silver (Ag) without the need for an external reducer, resulting in a black precipitate visible under a light microscope. The presence of melanin in the tissue is thus demonstrated by black staining. Gold chloride is used as a toning agent to enhance contrast and improve the clarity of melanin visualization. To prevent non-specific background staining from unreduced silver, sodium thiosulfate is applied to remove any excess silver salts. Finally, safranin is used as a counterstain to highlight other tissue components for better morphological assessment.

REAGENTS PROVIDED

Kit Contents	Product Code	Storage conditions	Pack Sizes	
			25 tests	50 tests
Silver Nitrate Solution - B (Reagent A)	IPS061	2-8°C	50ml	100ml
Gold Chloride Solution (Reagent B)	IPS050	2-8°C	25ml	50ml
Sodium Thiosulphate Solution - A (Reagent C)	IPS051	RT	25ml	50ml
Safranin Solution (Reagent D)	IPS097	RT	25ml	50ml

STORAGE AND HANDLING

Storage Recommendations: Store at recommended storage conditions. When stored at the appropriate conditions, the reagents are stable until the expiry date.

Do not use the reagents after the expiration date provided on the vial.

To ensure proper reagent performance and stability, replace the dispenser cap after every use. Immediately place the vials in the recommended storage conditions, keeping them away from sunlight and heat in an upright position.

During transport, short-term exposure to 2-8°C does not affect product performance.

Laboratory Use Only

SPECIMEN PREPARATION

Recommended positive controls: Skin, Malignant melanoma

Sample preparation and fixation: Formalin-fixed, Paraffin-embedded tissue sections of 4-5 µm thickness

PRECAUTIONS

1. This product should be used by qualified and trained professional users only.
2. The product contains hazardous reagents, must use gloves while handling.
3. It can cause serious eye and skin irritation. Refer to Material Safety Datasheet for any updated risk, hazard or safety information.
4. Dispose of waste observing all local, state, provincial or national regulations.
5. Do not use reagents after expiration date.
6. Use protective clothing or laboratory aprons, while handling reagents.
7. Avoid contamination of reagents as it may lead to incorrect results.

MATERIALS REQUIRED, BUT NOT PROVIDED

- Positive control and Negative control slides
- Ammonium Hydroxide, concentrated
- Xylenes
- Alcohol (50%, 70%, 95%, Absolute)
- Mounting Medium
- Microscopic slides (Positively charged)
- Slide holder
- Cover slips
- Coplin jars
- Distilled water

PREPARATION OF WORKING SOLUTION

Ammoniacal Silver Nitrate working solution:

1. Take the required volume of Silver Nitrate Solution - B (Reagent A) in a clean conical flask.
2. While continuously shaking or swirling the flask, add concentrated ammonium hydroxide (**not provided**) drop by drop until the precipitate formed is completely dissolved. (**Do not add excess ammonium hydroxide**)
3. Dilute the resulting solution to the given volume using distilled or deionized water to make it to a final volume.

Note: The Ammoniacal silver nitrate working solution cannot be reused.

Reference volume for Ammoniacal Silver Nitrate working solution preparation:

- Silver Nitrate Solution - B (Reagent A): 2ml
- Distilled Water: 48ml

STAINING PROCEDURE

1. Deparaffinize in three changes of xylene and hydrate to distilled water via decreasing concentrations of alcohols (100%, 70%, and 50%) for 3 minutes each.
2. Preheat the Ammoniacal silver nitrate working solution (Refer to working solution preparation above) for 15 min at 60 °C.
3. Place the slides in preheated Ammoniacal silver nitrate solution for 45 min in water bath at 60° C
4. Rinse the slides in distilled water.
5. Incubate the sections with Gold Chloride Solution (Reagent B) for 5 minutes.
6. Rinse slides thoroughly in distilled water.
7. Incubate slides with Sodium Thiosulphate Solution - A (Reagent C) for 5 minutes.
8. Rinse the slides in distilled water for 2 minutes.
9. Counterstain with Safranin Solution (Reagent D) for 5-10 Seconds.
10. Rinse slides in distilled water for 30 seconds.
11. Dehydrate quickly through 95% alcohol and 2 changes of 100% alcohol.
12. Clear in 3 changes of xylene for 2 minutes each.
13. Cover slip with compatible mounting medium.

QUALITY CONTROL

The recommended positive tissue control for Masson Fontana is the skin, Malignant/pigmented melanoma.

PERFORMANCE CHARACTERISTICS

Masson Fontana stain kit stains **Melanin** in **black** colour, and **other tissues** **Elements** will be stained **pink** in colour.

TROUBLESHOOTING

1. Follow the specific protocol recommendations according to the data sheet provided.
2. Tissue staining is dependent on the handling and processing of the tissue prior to staining. Improper fixation, tissue processing, freezing, thawing, washing, drying, heating, sectioning or contamination with other tissues or fluids may produce artifacts, reagent trapping or inaccurate results.
3. Do not allow the section to dry out during the entire staining process.
4. Gently mix all the reagents prior to use.
5. Excessive or incomplete counterstaining may compromise the interpretation of the results.
6. If unusual results occur, contact PathnSitu Technical Support at +91-40-2701 5544 or E-mail: techsupport@pathnsitu.com

LIMITATIONS AND WARRANTY

1. This product is intended for use only by authorised, trained, and qualified personnel.
2. A qualified and trained pathologist/personnel must interpret the results of the test.
3. Interpretation of test results must be made in conjunction with relevant background information and additional laboratory findings.
4. Always use the recommended volume and concentration of reagents to ensure complete coverage of the tissue section and to minimise the risk of false-positive or false-negative results.
5. Use appropriate buffers, instruments, consumables, and incubation conditions as recommended to achieve optimal staining performance.
6. It is strongly recommended to include known positive and negative controls when performing the test to ensure the validity of results.
7. The product has been validated on formalin-fixed, paraffin-embedded (FFPE) tissues. The end user must establish performance on other tissue types.
8. Unexpected results may occur in untested tissues due to inherent variability in tissue components.
9. False-positive reactions may occur due to insufficient washing, inappropriate protocol conditions, or other contributing factors.
10. In instances where the staining pattern or localisation differs from the specifications outlined in this datasheet, please get in touch with technical support for guidance.
11. Maintain the product under the recommended storage conditions to preserve reagent stability and performance.
12. Do not use reagents that appear cloudy, discoloured, or show signs of contamination. Discard any components showing signs of deterioration.
13. Silver Nitrate is light sensitive. Avoid exposing silver nitrate to bright light, including direct sunlight, as it can cause the chemical to break down.
14. This product is intended for single-use application only. Once applied to a tissue section, reagents should not be recovered or reused, as this may compromise test integrity and specificity.
15. PathnSitu makes no warranties beyond those expressly stated in the product description.
16. PathnSitu shall not be liable for property damage, personal injury, time or effort, or economic loss arising from the use of this product.
17. Please refer to the complete datasheet for all instructions, precautions, and additional product limitations.
18. For detailed information and specifications on individual components, please refer to Product Material Safety Data Sheet (MSDS)

BIBLIOGRAPHY

1. A Novel Histochemical Method for a Simultaneous Staining of Melanin and Collagen Fibers Víctor S. Carriel, Jose Aneiros-Fernandez, Salvador Arias-Santiago, Ingrid J. Garzón, Miguel Alaminos, and Antonio Campos
2. Relationship between skin response to ultraviolet exposure and skin color type S. Del Bino, J. Sok, E. Bessac and F. Bernerd*
3. Melanoma of the Urinary Bladder: A Review of the Literature Anthony Kodzo-Grey Venyo

EXPLANATION OF SYMBOLS

LOT

Lot number / Batch number



Storage limitation



Date of manufacture



Manufacturer address



Expiry

RT

Room Temperature

REF

Catalogue number