

# Vonkossa Stain

# PRODUCT INFORMATION:

PERFORMANCE CHARACTERISTICS:

REFSSP023100 ml (Ready to use)SSP023250 ml (Ready to use)SSP023500 ml (Ready to use)

Staining Interpretation: Calcium Salts: Black Nuclei : Red Cytoplasm : Light Pink

# SUMMARY AND EXPLANATION

#### For Laboratory use only

Vonkossa Stain Kit is intended for use in the histological visualization of calcium deposits in paraffin sections. This stain is useful for the diagnosis of calcinosis. It can also be used to detect calcifications in tumors. Von Kossa stain can be used on all types of tissues that may have abnormal calcium deposits like the joints, blood vessels and kidneys which are most often affected.

# PRINCIPLE OF THE PROCEDURE

Von Kossa stain is widely used in histology to detect the presence of mineral deposits like abnormal calcium deposits in the body. The principle of this staining is based on the transformation of calcium salts into silver salts: Von Kossa's technique is clearly a two-step reaction: first, silver cations react with the components of calcium deposits, resulting in a transient yellow coloration. This coloration is transient and gradually changes through grey to black in the second step, where the organic material reduces the bound silver to black metallic silver with the aid of light. Counterstaining with nuclear fast red allows to stain the nuclei of the cells in red and their cytoplasm in pink.

			Pack Sizes		
Kit Contents	Product Code	Storage Conditions	100ml	250ml	500ml
5% Silver Nitrate (Reagent A)	IPS048	2-8°C	100ml	250ml	500ml
5% Sodium Thiosulphate (Reagent B)	IPS051	RT	100ml	250ml	500ml
0.5% Nuclear fast Red (Reagent C)	IPS085	RT	100ml	250ml	500ml

## STORAGE AND HANDLING

Storage Recommendations: Store at Recommended temperatures. When stored at the appropriate conditions, the reagents are stable until expiry. Do not use the reagents after expiration date provided on the vial.

To ensure proper regent delivery and stability, replace the dispenser cap after every use and immediately place the vials at room temperature away from sunlight in an upright position.

#### **SPECIMEN PREPARATION**

## **RECOMMENDED POSITIVE CONTROLS:**

1. Tissues with Calcium deposits

#### SAMPLE PREPARATION AND FIXATION:

 Formalin-fixed, Paraffin-embedded tissue sections of 3- 5 µm thickness on microscopic slides.

#### DS-SSP023-B

PRECAUTIONS

- 1. Normal precautions exercised in handling laboratory reagents should be followed.
- 2. This product should be used by qualified and trained professional users only
- 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 and gloves, while handling reagents
- 7. Avoid microbial contamination of reagents as it may lead to incorrect results

### MATERIALS REQUIRED, BUT NOT PROVIDED:

- Xylenes
- Graded alcohols (50%, 70%, 95%, absolute)
- DPX Mountant
- Microscopic slides (positively charged)
- Slide holder
- Cover slips
- Coplin jars
- Incandescent Light source

## STAINING PROCEDURE:

- 1. Deparaffinize the tissue sections and hydrate to distilled water.
- Incubate slide in Silver Nitrate Solution (5%), Reagent A, for 30 minutes or longer while exposing to either light at 100 watts or Sunlight. (For best results, keep light source nearer to the slide) (~ 60cm) during Silver Nitrate impregnation procedure.
- 3. Rinse in 3 changes of distilled water.
- 4. Incubate slide in Sodium Thiosulfate Solution (5%), Reagent B for 2 minutes.
- 5. Rinse for 2 minutes in running tap water followed by 2 changes of distilled water.
- Stain tissue section with 05% Nuclear Fast Red Solution, Reagent C for 5 minutes.
- Rinse for 2 minutes in running tap water followed by 2 changes of distilled water.
- 8. Dehydrate quickly in Graded Alcohols.
- 9. Clear in 3 changes of xylene and mount the slide with compatible medium.

#### QUALITY CONTROL

The recommended positive tissue control for Vonkossa stain is the tissue with calcium deposits

#### PERFORMANCE CHARACTERISTICS

The Vonkossa stain for **Calcium Salts** stains in **Black** color, the **Cell Nuclei** stains in **red** color and the **cytoplasm** stains in **light pink** color.



#### TROUBLESHOOTING

- 1. Follow the specific protocol recommendations according to data sheet provided
- 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
- Do not allow the section to dry out during the entire staining process
- 4. Excessive or incomplete counterstaining may compromise the interpretation of the results
- If unusual results occur, contact PathnSitu Technical Support at +91-40-2701 5544 or E-mail: <u>techsupport@pathnsitu.com</u>

# LIMITATIONS AND WARRANTY

- 1. Authorized and skilled/trained personnel only may use the product.
- The clinical interpretation of any test results should be evaluated within the context of the patient's medical history and other diagnostic test results.
- A qualified trained pathologist must perform the evaluation of the test results.
- 4. The product comes with no warranties beyond the provided description
- Use appropriate volume/concentration to cover entire tissue sections and optimum conditions to avoid false positive and negative results.
- 6. Use appropriate/recommended buffer/instruments/all consumables with appropriate incubation timings to obtain optimal results.
- Always recommend using known positive and negative controls to evaluate the test result.
- 8. Unexpected reactions may occur in untested tissues due to tissue component variability.
- False positive results can arise from no stringent washing practices and other contributing factors.
- In instances where localization differs from the specifications outlined in the datasheet, clinical coordination or prompt technical support is advised.
- 11. Maintain recommended storage conditions.
- 12. Refer entire data sheet to know any further limitations about the product.
- No warranties whether expressed or implied, extend beyond the description.
- 14. PathnSitu is not liable for property damage, personal injury, time or effort or economic loss caused by this product.

#### BIBLIOGRAPHY

- Von Kossa and his staining technique; Marlon R. Schneider1, Accepted: 3 November 2021 / Published online: 20 November 2021
- Identification and Characterization of Vascular Calcification– Associated Factor, a Novel Gene Upregulated During Vascular Calcification In Vitro and In Vivo; M. Yvonne Alexander, Fiona L. Wilkinson, John Paul Kirton, Claire Farrington Rock, Georgina D.M. Collett, Maria Jeziorska, J. Vincent Smyth, Anthony M. Heagerty, Ann E. Canfield
- HISTOCHEMICAL M ETHODS FOR CALCIUM, S.M McGEE Russell, Department of zoology, Birkbeck college, University of London, Male street W.C, London, June 6, 1957

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**EXPLANATION OF SYMBOLS**