



# Instructions For Use

## OAA-IFU

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## Orcein Solution

### Description and Principle

Orcein Solution may be used in histology procedures for the visualization of Hepatitis B surface Antigen (HBsAg), elastic fibers, and copper associated proteins. HBsAg appears as irregular shaped aggregates in the cytoplasmic region of the cells. This reagent may be used on formalin-fixed, paraffin-embedded or frozen sections.

### Expected Results

HBsAg:	Dark Brown/Purple
Elastic Fibers:	Dark Brown/Purple
Copper Assoc. Proteins:	Dark Purple
Background:	Light Reddish/Purple

### Kit Contents

#### Additional Kit Reagents Sold Separately

	Storage
1. Potassium Permanganate Sol. (5%)	18-25°C
2. Sulfuric Acid Solution (3%)	18-25°C
3. Oxalic Acid Solution (2%)	18-25°C
4. Orcein Solution	18-25°C
5. Differentiating Solution	18-25°C

### Suggested Controls (not provided)

Known hepatitis positive liver, Lung for elastic fiber, Any well fixed tissue cut 3-5 microns.

### Uses/Limitations

For In-Vitro Diagnostic use only.

Do not use if reagents become cloudy or precipitate

Do not use past expiration date.

Use caution when handling reagents.

Non-Sterile

Intended for FFPE sections cut at 5-10µm.

This procedure has not been optimized for frozen sections.

Frozen sections may require protocol modification.

### Storage

Store at room temperature (18-25°C).

### Safety and Precautions

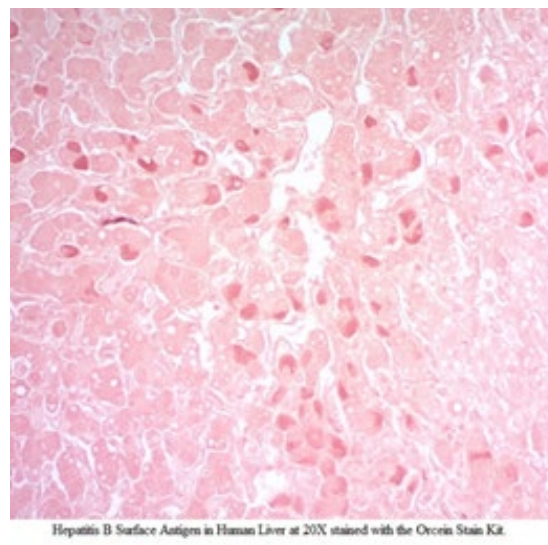
Please see current Safety Data Sheets (SDS) for this product and components GHS classification, pictograms, and full hazard/precautionary statements.

### Procedure

#### Prepare Oxidizing Immediately Prior to Beginning Procedure:

Combine:	50 ml	Distilled Water
	5 ml	Potassium Permanganate Solution (5%)
	3 ml	Sulfuric Acid Solution (3%)
		Mix thoroughly.

1. Deparaffinize sections if necessary and hydrate to distilled water.
2. Incubate slide in freshly prepared Oxidizing Solution for 10 minutes.
3. Rinse slide briefly in running tap water followed by 1 dip in distilled water.
4. Incubate slide in Oxalic Acid Solution (2%) for 10 minutes or until clear.  
Note: Section should be colorless following this step.



5. Rinse slide for 1 minute in running tap water followed by 2 dips in distilled water.

6. Incubate slide in coplin jar containing Orcein solution for 4-8 hours (2 hours is sufficient for elastin). Note: Ensure tissue is fully immersed in staining jar. Close lid to prevent evaporation.

7. Rinse slide in Alcohol, Reagent (70%).

8. Differentiate in Differentiating Solution for 10-60 seconds.

9. Dip slide in Alcohol, Reagent (70%) and check slide microscopically for proper differentiation.

**Note:** Repeat step 8 if necessary.

10. Dehydrate quickly in 3 changes of absolute alcohol.

11. Clear, and mount in synthetic resin.

**Note:** If darker staining is preferred:


1) Incubation time in Orcein solution may be increased.




and/or

2) Differentiation may be omitted by replacing steps 7-9 with a simple rinse with deionized water.

### References

1. Deodhar K.P., Tapp E., Scheuer P.J. Orcein staining of Hepatitis B Antigen in paraffin sections of Liver Biopsies. Journal of Clinical Pathology; vol. 28: pages 66-70, 1975.
2. Salaspuro, M., Sipponen, P. Demonstration of an intracellular copper-binding protein by Orcein staining in long-standing cholestatic liver diseases. Gut, 1976, volume 17: pages 787-790.

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