

## **A-131 PINKERTON'S METHOD FOR RICKETTSIA**

**FIXATIVE:** Zenker's modified" Mercury free" (F-222), Regaud's (SO-424), Muller's (F-190) or Buffered Neutral Formalin, 10% (F-113)

**SECTIONS:** Cut Paraffin sections at 6 microns.

**STAINING:**

1. Deparaffinize and hydrate to distilled water.
2. Stain overnight in Methylene Blue, 1% Aq. (A-131-1). Rinse in 95% alcohol for 5 seconds to prevent loss of blue color. Rinse quickly in distilled water for 2-3 seconds.
3. Stain for 30 minutes in Basic Fuchsin, 0.25% (A-131 -2).
4. Decolorize rapidly in Citric Acid, 0.5% (A-131-3) for 1-2 seconds, never more than 3 seconds.
5. Differentiate further in absolute alcohol until nuclei stand out blue and rickettsia clumps red.
6. Clear with 2 changes of Xylene.
7. Mount with Permount (M-18).

**STAINING RESULTS:**

RICKETTSIA	BRIGHT RED
NUCLEI	BLUE

**REFERENCES:**

Simmons, J.S. and Gentzkow, C.J., Laboratory Methods of the United States Army, 5th ed., Lee & Febiger, Philadelphia, p.572.,1944.

Luna, L.G., (ed.) Manual of Histologic Staining Methods of the AFIP, 3rd edition, McGraw-Hill, N.Y., p. 237., c.1968.