

## **K-681, KLUVER-BARRERA METHOD FOR MYELIN AND NERVE CELLS**

**Fixation:** Formalin, 10% Buffered Neutral (F-113)

**Sections:** Paraffin @ 15 to 20 microns

**Staining:**

1. Deparaffinize and hydrate to 95% alcohol.
2. Leave in **Luxol Fast Blue Solution** (K-681-1) in 56°C oven overnight.
3. Rinse off excess stain with 95% alcohol.
4. Rinse in distilled water.
5. Differentiate the slides singly in **Lithium Carbonate Solution** (K-681-4) for 30 seconds.
6. Continue differentiation in 70% alcohol until the gray matter is clear and white matter sharply defined.
7. Check microscopically. Repeat the differentiation if necessary starting at step 5.
8. When differentiation is complete, place in distilled water.
9. When all slides have been collected in distilled water, add fresh distilled water.
10. Counterstain in (**Cresyl Violet Acetate**), Working solution for 6 minutes.  
To prepare working solution add 15 drops of **Acetic Acid, 10%** (K-681-3) to 100 ml of **Cresyl Violet Acetate, 0.1%** (K-681-2) just before use. Filter.
11. Rinse in 2 changes of 95% alcohol.
12. Continue the dehydration through two changes of absolute ethyl alcohol and xylene, two changes each, for 2 minutes.
13. Mount with resinous medium.

**Results:**

Myelin, including phospholipids	Blue to Green cells
Cells and cell products	Pink to violet

- NOTE: The Luxol Fast Blue procedure works well when combined with other procedures as the Bodian Method.

**References:**

Kluver,H., and Barrera, E. : A method for the combined staining of cells and fibers in the Nervous system. *J. Nueropath.Exp. Neurol.* 12:400-403, 1953.

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