

## **G-486, LUNA'S METHOD FOR ERYTHROCYTES AND EOSINOPHIL GRANULES**

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

**Sections:** Paraffin @ 6 microns

### **Procedure:**

1. Deparaffinize and hydrate to distilled water.
2. Stain for 5 minutes in Working Hematoxylin-Biebrich Scarlet Solution.

\*To prepare Working Hematoxylin-Biebrich Scarlet Solution:

First prepare Hematoxylin Working Solution by mixing:

1. **Weigert's Hematoxylin A** (G-486-1A).....22.5 ml
2. **Weigert's Hematoxylin B** (G-486-1B)... ..22.5 ml

Mix solutions well- then add...

3. **Biebrich Scarlet, 1% Aqueous** (G-486-2)...5 ml

Mix well.

3. Differentiate in **Acid Alcohol, 1%** (G-486-3) until the desired nuclear detail is achieved (usually 8 dips). Remove Acid Alcohol by rinsing in water.
4. Dip in **Lithium Carbonate, 0.5%** (G-486-4) until section turns blue and erythrocytes are bright red (usually 5 dips). Wash in running water for 2 minutes.
5. Dehydrate in 95% alcohol, absolute alcohol, and clear in xylene, two changes each.
6. Mount with **Permount** (M-18).

### **Stain Results:**

Eosinophil granules, erythrocytes and Charcott Leyden crystals	Red
Background	Blue

### **References:**

Luna, L.G., (ed.), Manual of Histologic Staining Methods of the AFIP, 3<sup>rd</sup> edition,  
McGraw-Hill, NY, p1111, c 1968.