

## 碱性固绿染色液

货号: G2551

规格: 2×50mL

保存: 室温, 避光保存, 有效期 1 年。

### 产品组成:

名称	2×50mL	保存
试剂(A): 酸性分化液	50mL	室温, 避光
试剂(B): 碱性固绿染色液	B1: 碱性固绿 A	25mL
	B2: 碱性固绿 B	25mL
临用前, 取 B1、B2 等量混合, 即为碱性固绿染色液, 即配即用。		

### 产品介绍:

不同的氨基酸带有不同化学性质的侧链基团, 有的带有碱性侧链, 有的带有酸性侧链, 由此组成的蛋白质具有不同数目的碱性基团和酸性基团, 这些基团会使蛋白质在不同的 pH 溶液中带有不同的净电荷, 整个蛋白质分子带正电荷多, 即为碱性蛋白(等电点偏向酸性); 整个蛋白质带负电荷多, 即为酸性蛋白(等电点偏向酸性)。

碱性固绿染色液是利用碱性蛋白与带有负电荷的碱性化染料固绿结合进行染色。细胞内含量最丰富的碱性蛋白是组蛋白, 主要存在于细胞核内。因此染色后细胞核大部分被染成绿色。该染色液仅用于科研领域, 不宜用于临床诊断或其他用途。

### 自备材料:

玻片、70%乙醇、水浴锅、显微镜

### 操作步骤: (仅供参考)

#### (一) 对于组织染色

1. 组织固定于 10%福尔马林中, 常规脱水包埋。切片厚 4 $\mu$ m, 常规脱蜡至水。
2. 切片浸入酸性分化液中, 60 $^{\circ}$ C 水浴 30min, 蒸馏水洗两次, 每次 1min。
3. 滤纸吸去残留水分。
4. 切片浸入碱性固绿染色液染色 5~15min, 蒸馏水洗 30s。
5. 室温晾干, 中性树胶封固。

#### (二) 对于贴壁细胞或爬片染色

1. 使用中性福尔马林固定液或组织细胞固定液室温固定 10-20min。
2. 按照培养基添加量的一半加入酸性分化液, 烘箱 60 $^{\circ}$ C 孵育 30min, 蒸馏水洗两次, 每次 1min。
3. 吸去多余水分。
4. 滴加同分化液等量的碱性固绿染色液染色 5~15min, 蒸馏水洗 30s。
5. 带水观察。

#### (三) 对于血涂片或悬浮细胞染色

1. 取新鲜血液 1 滴滴于载玻片一端, 推片, 室温晾干。
2. 涂片浸入 70%乙醇中固定 5min, 室温晾干。
3. 涂片浸入酸性分化液中, 60 $^{\circ}$ C 水浴 30min。
4. 流水充分水洗, 滤纸吸去残留水分。
5. 涂片浸入碱性固绿染色液染色 15~30min。
6. 流水冲洗, 室温晾干。
7. 直接镜检或滴加 1 滴中性树胶, 加盖盖玻片进行封片观察。

### 染色结果:

细胞核大部分区域	绿色
细胞质、核仁	不着色

**注意事项：**

1. 血液涂片或骨髓涂片应厚薄均匀，以免影响染色效果。
2. 血细胞涂片染色要求新鲜全血或 EDTA 抗凝血。
3. 酸性分化液孵育后，冲洗应彻底，否则会干扰固绿的染色。
4. 染色过深可用甲醇或酒精适当脱色，最好不复染。
5. pH 值对染色有一定影响，载玻片应清洁、无酸碱污染，以免影响染色效果。
6. 为了您的安全和健康，请穿实验服并戴一次性手套操作。

## Alkaline Fast Green Stain Solution

**Cat:** G2551

**Size:** 2×50mL

**Storage:** RT, avoid light, valid for 1 year.

### Kit Components

Reagent		2×50mL	Storage
Reagent(A): Acid Differentiation Solution		50mL	RT, avoid light
Reagent(B): Alkaline Fast Green Solution	B1: Alkaline Fast Green Solution A	25mL	RT, avoid light
	B2: Alkaline Fast Green Solution B	25mL	RT, avoid light
Before use, mix B1 with B2 in equal amount to form Alkaline Fast Green Solution. It is ready to use.			

### Introduction

Different amino acids have side chain groups with different chemical properties. Some have basic side chains, some have acid side chains, and the resulting proteins have different numbers of basic and acid groups. These groups will make proteins with different net charges in different pH solutions, and the whole protein molecule has more positive charges, that is, basic protein (isoelectric point tends to be alkaline); The whole protein has more negative charge, that is acid protein (isoelectric point tends to be acid).

The principle of Alkaline Fast Green Stain Solution is that basic protein combines with basic dye containing negative charge to show color. The most abundant basic protein in the cell is histone, which mainly exists in the nucleus. So after dyeing, most of the nucleus are dyed green. The staining solution is only used in scientific research field, and is not suitable for clinical diagnosis or other purposes.

### Self Provided Materials

Slide, 70% ethanol, Water bath, Microscope.

### Protocol (for reference only)

#### For Tissue Staining

1. Fix the tissue in 10% formalin and routinely dehydrate and embed. Slice into 4μm. Conventional dewaxing to water.
2. Immerse the section in Acid Differentiation Solution, soak in 60°C water for 30 min, and wash twice with distilled water for 1 min each time.
3. Filter paper absorbs residual water.
4. Dip the section into Alkaline Fast Green Solution for 5-15 min and wash with distilled water for 30 s.
5. Dry at room temperature and seal with neutral gum.

#### For Adherent Cells or Climbing Section Staining

1. The cells were fixed in neutral formalin or tissue cell solution at room temperature for 10-20 min.
2. Add Acid Differentiation Solution according to half of the added amount of medium, incubate in warm at 60 °C for 30min, and wash twice with distilled water for 1min each time. 3. Absorb excess water.
3. Add the same amount of Alkaline Fast Green Solution to dye for 5-15 min, and wash with distilled water for 30 seconds.
4. Observe with water.

#### For Blood Smear or Suspension Cell Staining

1. Take 1 drop of fresh blood to one end of the slide, push the slide and dry at room temperature.
2. Immerse the smear in 70% ethanol for 5min and dry it at room temperature.
3. Immerse the smear in Acid Differentiation Solution and incubate in 60 °C water bath for 30min.
4. Rinse with running water completely and absorb residual water by filter paper .
5. Immerse the smear in Alkaline Fast Green Solution for 15-30 min.
6. Rinse with running water and dry at room temperature.
7. View under the microscope directly or after sealing with resinene.

### Result

Cytoplasm and Nucleolus	Green
Most areas of nucleus	Colorless

## Note

1. The blood smear or bone marrow smear should be uniform in thickness to avoid affecting the staining effect.
2. Blood cell smear staining requires fresh whole blood or EDTA anticoagulant.
3. After incubation with Acid Differentiation Solution, wash thoroughly, otherwise it will interfere with the staining of fast green.
4. If the dye color is too deep, it can be decolorized properly with methanol or alcohol, and it is better not to be re-dyed.
5. The pH value has certain influence on the dyeing. The slide should be clean and free of acid and alkali pollution to avoid affecting the dyeing effect.
6. For your safety and health, please wear experimental clothes and disposable gloves.