

## 硫堇染色液(0.4%)

货号: G1900

规格: 50mL

保存: 室温, 避光保存, 有效期 6 个月。

### 产品介绍:

染色体是细胞内具有遗传性质的遗传物质深度压缩形成的聚合体, 易被碱性染料染成深色, 所以叫染色体(染色质), 染色体和染色质是同一物质在细胞分裂间期和分裂期的不同形态表现而已。染色体出现于分裂期, 染色质出现于间期, 呈丝状, 其本质都是脱氧核糖核酸(DNA)和蛋白质的组合(即核蛋白组成的), 不均匀地分布于细胞核中, 是遗传信息(基因)的主要载体, 但不是唯一载体(如细胞质内的线粒体)。

许多染料对 DNA 都有不同程度的亲和性, 故可以作为染色体的染色。在染色体的常规染色中, 一般用姬姆萨、地衣红、福尔根、石炭酸复红等可获得较好的染色效果。硫堇染色液(0.4%)可用于口腔黏膜、尿液、羊水、绒毛细胞以及人工培养细胞等样本的染色体染色, 尤其适用于较难上色的性染色质的染色。硫堇染色亦可用于肥大细胞的染色, 异染性物质呈紫红色, 其他呈蓝色。

### 自备材料:

PBS 或生理盐水、蒸馏水、载玻片、盖玻片、AF 固定液、1mol/L HCl、显微镜

### 操作步骤: (仅供参考)

#### (一)样本处理

1. 口腔黏膜细胞:
  - 1) 用 PBS 或生理盐水漱口数次, 尽量去除口腔内细菌和其他杂物。
  - 2) 操作人员一手拉住患者的下唇, 一手用压舌板或牙签钝头端刮取两侧颊部或下唇内侧的粘液, 丢弃第一次刮取的细胞。
  - 3) 同一部位连续刮取数次, 将刮取物涮入装有 5mL 生理盐水的离心管中。
  - 4) 1500g 离心 10-15min, 弃上清液, 留取细胞团。
  - 5) 加入新鲜固定液(甲醇:冰乙酸=3:1)10mL, 轻轻混匀制成悬液, 室温放置 30min。
  - 6) 取一滴悬液至预冷的干净载玻片上, 晾干。
2. 尿液中脱落细胞:
  - 1) 取患者干净的中段尿液, 混匀, 吸取 10mL 至离心管中。
  - 2) 1500-2000g 离心 10-15min, 弃上清液, 留取细胞团。
  - 3) 加入新鲜固定液(甲醇:冰乙酸=3:1)10mL, 轻轻混匀制成悬液, 室温放置 30min。
  - 4) 1500g 离心 10-15min, 弃上清液, 留取细胞团。
  - 5) 根据细胞的多少, 加入数滴新配制的固定液, 充分混匀制成悬液。
  - 6) 取一滴悬液至预冷的干净载玻片上, 晾干。
3. 羊水细胞:
  - 1) 按妇科常规经腹壁穿刺妊娠 16 周左右孕妇的羊水 10mL 至离心管中, 抽取羊水时先抽取 2-3mL 丢弃, 以免母体细胞的污染。
  - 2) 1500g 离心 10min, 弃上清液, 留取细胞团。
  - 3) 加入新鲜固定液(甲醇:冰乙酸=3:1)10mL, 轻轻混匀制成悬液, 室温放置 30min。
  - 4) 1500g 离心 15min, 弃上清液, 留取细胞团。
  - 5) 根据细胞的多少, 加入数滴新配制的固定液, 充分混匀制成悬液。
  - 6) 取一滴悬液至预冷的干净载玻片上, 晾干。

#### (二)染色质染色

1. 玻片标本置于 1mol/L HCl 中, 37°C 孵育 20min。
2. 蒸馏水充分冲洗, 自然干燥。
3. 将玻片样本浸入硫堇染色液(0.4%), 染色 15-20min。
4. 蒸馏水冲洗, 自然干燥。





5. 在低倍镜下查找均匀分散的细胞群，转油镜认真观察。

### 染色结果：

异染性物质呈紫红色，其他呈蓝色。

### 注意事项：

1. 在为了获得细胞沉淀的离心的过程中，对于特殊细胞，如果细胞沉淀不充分，可以适当提高离心力或延长离心时间。
2. 为了您的安全和健康，请穿实验服并戴一次性手套操作。



## Thionin Blue Stain Solution, 0.4%

**Cat:** G1900

**Size:** 50mL

**Storage:** RT, avoid light, valid for 6 months.

### Introduction

Chromosome is a kind of polymer formed by deep compression of genetic material with genetic properties in cells, which is easy to be dyed dark by basic dyes, so it is called chromosome (chromatin). Chromosome and chromatin are different morphological manifestations of the same substance in cell division and interphase. Chromosomes appear in the division stage, chromatin appears in the interphase, and it is filiform. The essence of both are the combination of DNA and protein (composed of nucleoprotein). It is unevenly distributed in the nucleus, and it is the main carrier of genetic information (genes), but not the only carrier (such as mitochondria in the cytoplasm).

Many dyes have different degrees of affinity to DNA, so they can be used as chromosome staining. In the routine chromosome staining, Giemsa, Orcein, Feulgen and Carboic Fuchsin are generally used to obtain better staining effect. Thionin Blue Stain Solution, 0.4% can be used for chromosome staining of oral mucosa, urine, amniotic fluid, villus cells and cultured cells, especially for sex chromatin which is difficult to be stained. Thionine staining can also be used for mast cell staining. The heterochromatin is purplish red, others are blue.

### Self Provided Materials

PBS or Physiological saline or Distilled water, Slide, Fixative (methanol: acetic acid = 3:1), 1M HCl, Microscope

### Protocol(for reference only)

#### Sample treatment

##### *For Oral Mucosa Cells*

1. Wash the mouth several times with PBS or physiological saline, and try to remove bacteria and other impurities in the mouth.
2. The operator holds the patient's lower lip with one hand, scrapes the mucus on both cheeks or the inner side of the lower lip with the tongue depressor or the blunt end of toothpick with the other hand, and discards the cells scraped for the first time.
3. Scrape the same part several times continuously, and rinse the scraped object into the centrifuge tube containing 5mL physiological saline.
4. Centrifuge with 1500g for 10-15mins, discard supernatant and retain cell mass.
5. Add 10mL fresh fixative (methanol: acetic acid = 3:1), mix gently to make suspension, and place it at room temperature for 30mins.
6. Take a drop of suspension onto the precooled clean slide and dry it.

##### *For Exfoliated Cells in urine*

1. Take the clean middle urine of the patient, mix well, and drop 10mL into the centrifuge tube.
2. Centrifuge with 1500-2000g for 10-15mins, discard supernatant and retain cell mass.
3. Add 10mL fresh fixative (methanol: acetic acid = 3:1), mix gently to make suspension, and place it at room temperature for 30mins.
4. Centrifuge with 1500g for 10-15mins discard supernatant and retain cell mass.
5. According to the number of cells, add a few drops of newly prepared fixative, and mix them well to make suspension.
6. Take a drop of suspension onto the precooled clean slide and dry it.

##### *For Amniotic Fluid Cells*

1. According to the gynecological routine, take 10 mL amniotic fluid of pregnant women about 16 weeks of gestation through abdominal wall puncture to the centrifuge tube and discard 2-3 mL amniotic fluid first taken to avoid the pollution of mother cells.
2. Centrifuge with 1500g for 10mins, discard supernatant and retain cell mass.
3. Add 10mL fresh fixative (methanol: acetic acid = 3:1), mix gently to make suspension, and place it at room temperature for 30mins.
4. Centrifuge with 1500g for 15mins, discard supernatant and retain cell mass.





5. According to the number of cells, add a few drops of newly prepared fixative, and mix them well to make suspension.
6. Take a drop of suspension onto the precooled clean slide and dry it.

#### **Chromatin staining**

1. Place the slide samples in 1M HCl and incubate at 37°C for 20mins.
2. Rinse with distilled water and dry naturally.
3. Place the slide sample in Thionin Blue Stain Solution, 0.4% and stain for 15-20mins.
4. Rinse with distilled water and dry naturally.
5. Look for the evenly distributed cell group under low power microscope, and observe carefully with oil immersion lens.

#### **Result**

Heterochromatic substances are purplish red, others are blue.

#### **Note**

1. In the process to obtain the centrifugation of cell precipitation, for special cells, if the cell precipitation is not enough, can appropriately increase the centrifugal force or extend the centrifugation time.
2. For your safety and health, please wear experimental clothes and disposable gloves.

