# 骨组织 Goldner 三色法染色实验报告

## 一、实验器材及试剂

1、 实验器材

| 名称 | 厂家 | 型号 |
| --- | --- | --- |
| 脱水机 | DIAPATH | Donatello |
| 包埋机 | 武汉俊杰电子有限公司 | JB-P5 |
| 病理切片机 | 上海徕卡仪器有限公司 | RM2016 |
| 冻台 | 武汉俊杰电子有限公司 | JB-L5 |
| 组织摊片机 | 浙江省金华市科迪仪器设备有限公司 | KD-P |
| 烤箱 | 天津市莱玻瑞仪器设备有限公司 | GFL-230 |
| 载玻片 | Wanwu | |
| 正置光学显微镜 | 日本尼康 | NIKON ECLIPSE E100 |
| 成像系统 | 日本尼康 | NIKON DS-U3 |

2、 主要实验试剂

| 试剂名称 | 厂家 | 货号 |
| --- | --- | --- |
| 无水乙醇 | 国药集团化学试剂有限公司 | 100092683 |
| 二甲苯 | 国药集团化学试剂有限公司 | 10023418 |
| Goldner 染液套装 | Wanwu | G1064 |
| 冰醋酸 | 国药集团化学试剂有限公司 | 10000218 |
| 中性树胶 | 国药集团化学试剂有限公司 | 10004160 |

## 二、实验步骤

**1、石蜡切片脱蜡至水：**依次将切片放入二甲苯I20min-二甲苯II20min-无水乙醇I5min-无水乙醇II5min-75%酒精 5min，自来水洗。

**2、细胞核染色：**Goldner 染液 A 与 Goldner 染液 B 液等比混合，切片入混合液染色 20min，自来水洗，1%盐酸酒精溶液分化 2s，自来水冲洗，蒸馏水洗。

**3、 Goldner 染液 C：**切片入第一缸 Goldner 染液 C 浸染 5-10 min，0.2%冰醋酸快速漂洗每缸 3s。

网址：http://www.wanwusw.com
细胞网址：http://www.hfwanwu.com
地址：合肥市蜀山区长江西路248号11层
电话： 400-1016-218
邮箱：2028438226@qq.com

合肥万物 合肥万物 合肥万物

**4、Goldner染液D染色：**切片入Goldner染液D浸染3min，显微镜下控制，以胶原处Goldner染液C褪色为准。

**5、Goldner染液C染色：**切片第二缸Goldner染液C染3-5min，0.2%冰醋酸快速漂洗每缸3s。

**6、Goldner染液E染色：**切片入Goldner染液E染3-5min，0.2%的冰醋酸三缸分化每缸2s，无水乙醇三缸脱水各2s、3s、5s。

**7、透明封片：**切片放入第三缸无水乙醇5min，二甲苯透明5min，中性树胶封片。

**8、**显微镜镜检，图像采集分析。

**三、结果判读：**

细胞核蓝紫色，矿化骨绿色，非矿化骨是橙红色。

**四、注意事项：**

1、根据切片的数量对铁苏木素染液定期进行更换，也可以现配现用；

2、Goldner 染液 D 与 Goldner 染液 C 的染色程度要控制好，胶原部分不能是太红，会影响 Goldner 染液 D 的着色；

3、冰醋酸分化 Goldner 染液 E 的过程中，要分化适当，不能过深或者过浅。

网址：http://www.wanwusw.com
细胞网址：http://www.hfwanwu.com
地址：合肥市蜀山区长江西路248号11层
电话： 400-1016-218
邮箱：2028438226@qq.com

合肥万物 合肥万物 合肥万物

# Bone Tissue Goldner Trichrome Staining

# Experiment Report

## I. Experimental equipment and reagents

1. Experimental equipment

| Equipment name | Manufacturer | Model No. |
|---|---|---|
| Dehydrator | DIAPATH | Donatello |
| Embedding center | Wuhan Junjie Electronics Co., Ltd. | JB-P5 |
| Pathological microtome | Shanghai Leica Instrument Co., Ltd. | RM2016 |
| Cooling plate | Wuhan Junjie Electronics Co., Ltd. | JB-L5 |
| Tissue spreading water bath | Zhejiang Jinhua Kedi Instrumental Equipment Co., Ltd. | KD-P |
| Oven | Tianjin Leibo Terry Equipment Co., Ltd. | GFL-230 |
| Microscope slide | Wanwu | |
| Upright electron microscope | JAPAN NIKON | NIKON ECLIPSE E100 |
| Imaging system | JAPAN NIKON | NIKON DS-U3 |

2. Main experiment reagents

| Reagent name | Manufacturer | Item No. |
|---|---|---|
| Anhydrous ethanol | Sinopharm Chemical Reagent Co.,Ltd. | 100092683 |
| Xylene | Sinopharm Chemical Reagent Co.,Ltd. | 10023418 |
| Goldner staining solution suit | Wanwu | G1064 |
| Glacial acetic acid | Sinopharm Chemical Reagent Co.,Ltd. | 10000218 |
| Neutral balsam | Sinopharm Chemical Reagent Co.,Ltd. | 10004160 |

## II. Experimental procedure

**1. Deparaffinize the paraffin sections to water:** Place the sections in sequence in Xylene I for 20min - Xylene II for 20min - Anhydrous ethanol I for 5min - Anhydrous ethanol II for 5min - 75% Ethyl alcohol for 5min , wash with tap water.

**2. Nuclear staining:** Goldner staining solution A and Goldner staining solution B are mixed in equal proportions, put sections in the mixed solution and stain for 20 min, wash with tap water,

网址：http://www.wanwusw.com
细胞网址：http://www.hfwanwu.com
地址：合肥市蜀山区长江西路248号11层
电话： 400-1016-218
邮箱：2028438226@qq.com

合肥万物

then differentiate with 1% hydrochloric acid alcohol solution for 2s, resin with tap water, and wash with distilled water.

**3. Staining with Goldner staining solution C:** Immerse sections into first jar of Goldner staining solution C to stain for 5-10 minutes, then rinse quickly with 0.2% glacial acetic acid, for 3 seconds per tank.

**4. Staining with Goldner staining solution D:** Dip the sections into Goldner staining solution D to stain for 3 min, control under microscope, subject to the fading of Goldner staining solution C at the collagen area.

5. **Staining with Goldner staining solution C:** Dip the sections in second jar of Goldner staining solution C and stain for 3-5min, rinse quickly with 0.2% glacial acetic acid for 3s per jar.

6. **Staining with Goldner staining solution E:** Put the sections into Goldner staining solution E and stain for 3-5min, the differentiate with three jars of 0.2% glacial acetic acid, for 2s per jar, and dehydrate with three jars of anhydrous ethanol, for 2s, 3s and 5s respectively.

**7. Transparency and sealing:** Place the sections into the third jar of anhydrous ethanol for 5min, Xylene transparency for 5min, lastly, seal with neutral balsam.

8. Microscope inspection, image acquisition and analysis.

**III. Interpretation of results:**

The cell nucleus are blue-violet, the mineralized bone is green, and the non-mineralized bone is orange-red.

**IV. Precautions:**

1. According to the number of sections, replace the iron hematoxylin staining solution regularly and it can also be prepared when using.

2. The staining degree of Goldner staining solution D and Goldner staining solution C must be well controlled. The collagen part cannot be too red, it will affect the coloring of Goldner staining solution D.

3. During the differentiating of Goldner staining solution E by glacial acetic acid, the differentiation should be proper, and not be too deep or too shallow.