



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	Servicebio	
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	Servicebio	G1064
冰醋酸	国药集团化学试剂有限公司	10000218
中性树脂	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120min-二甲苯120min-无水乙醇15min-无水乙醇15min-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。



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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。

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。



Bone Tissue Goldner Trichrome Staining

Experiment Report

1. Experimental Equipment and Reagents

1.1 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	Servicebio	
Upright electron microscope	JAPAN NIKON	NIKON ECLIPSE E100
Imaging system	JAPAN NIKON	NIKON DS-U3

1.2 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	Servicebio	G1064
Glacial acetic acid	Sinopharm Chemical Reagent Co.,Ltd.	10000218
Neutral balsam	Sinopharm Chemical Reagent Co.,Ltd.	10004160

2. Experimental Procedure

2.1 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.



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

2.3 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.

2.4 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.

2.5 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.

2.6 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.

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

2.8 Microscope inspection, image acquisition and analysis.

3. Interpretation of Results:

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

4. Precautions:

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

4.2 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.

4.3 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.



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