骨组织番红固绿染色实验报告

一、实验器材及试剂

1、 实验器材

名称	厂家	型号
脱水机	武汉俊杰电子有限公司	JJ-12J
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
番红固绿(骨组织)染液套装	Wanwu	G1053
分化液	Wanwu	G1040
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I 20 min -二甲苯 II 20min -无水乙醇I 5min - 无水乙醇II 5min - 75%酒精 5 min，自来水洗。

2、固绿染色：切片入骨组织固绿染色液 1 - 5min，水洗去多余染液，至软骨呈无色，1%盐酸酒精浸泡 10s，自来水稍洗。

3、番红染色：切片入骨组织番红染色液 1 - 5s，四缸无水乙醇快速脱水分别 5s、2s、10s、第四缸镜检。

4、切片放入二甲苯透明 5min，中性树胶封片。

5、显微镜镜检，图像采集分析。

三、染色判读：

软骨呈红色或橙红色，成骨呈绿色。有些结缔组织也呈红色。

四、注意事项：

1、染色过程中，注意番红不能染过，易与绿色杂合呈紫蓝色。

2、若片子效果不佳，可用盐酸酒精分化水洗至颜色褪掉后重染。

Safranin fast green staining for bone experimental report

1. Experimental equipment and reagents

1.1 Experimental equipment

Name	Manufactor	Model
Dehydrator	Wuhan Junjie Electronics Co., Ltd	JJ-12J
Embedding machine	Wuhan Junjie Electronics Co., Ltd	JB-P5
Pathological section machine	Shanghai Leica Instrument Co., Ltd	RM2016
Frozen platform	Wuhan Junjie Electronics Co., Ltd	JB-L5
KD-P Water Bath	Kedee	KD-P
Oven	Tianjin Lai Bo Rui Instrument Equipment Co., Ltd	GFL-230
Slides	Wanwu	
Orthostatic microscope	NIKON, JAPAN	NIKON ECLIPSE E100
Image system	NIKON, JAPAN	NIKON DS-U3

1.2 Main experimental reagents

Reagent name	Manufactor	Article number
Absolute ethanol	Sinopharm Group Chemical Reagent Co. LTD	100092683
Xylene	Sinopharm Group Chemical Reagent Co. LTD	10023418
Saffron Fast Green dye solution (Bone Tissue)	Wanwu kit	G1053
Differentiation solution	Wanwu	G1040
Neutral resin	Sinopharm Group Chemical Reagent Co. LTD	10004160

2. Experimental steps

2.1 Paraffin slides dewaxed as follow: Two changes of pure xylene for 20 min. Two changes of pure ethanol for 5min. 75% ethanol for 5min. Keep slides in tap water.

2.2 Fast green staining: The slides stained in fast green dye solution for 1-5 minutes, washed away the excess dye solution until the cartilage was colorless, and soaked in 1% hydrochloric acid and alcohol for 10s. And then washing them with tap water.

2.3 Saffron staining: The slides were stained in saffron dye solution for 1-5s, and then put into four cylinders of absolute ethanol, where three for rapid dehydration for 5s, 2s, 10s respectively, and kept in the fourth cylinder.

2.4 The slides were immersed in xylene to transparent for 5min, sealing with neutral resin.

2.5 Observed under microscope, and took images for analysis.

3. The results were as follows

Cartilage was red or orange-red, and bone formation was green. And some connective tissues were red.

4. Precautions

4.1 During the dyeing process, the saffron staining cannot be overtime, because it is easy to hybridize with fast green to become purple-blue.

4.2 If the result of the staining is not well, it can be washed with hydrochloric acid solution and alcohol until the color fades, and then re-stained.