刚果红染色实验报告

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	G6004
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
刚果红染液套装	Wanwu	G1056
分化液	Wanwu	G1005-3
返蓝液	Wanwu	G1005-4
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、刚果红染色：切片入刚果红 A 液浸染一夜，自来水洗 2min。

3、**背景分化：**刚果红 B 液分化 1s，至阳性斑块明显，背景基本无色，自来水洗。

4、**染核：**切片入刚果红C液染1min，自来水洗，分化液分化，自来水洗，返蓝液返蓝，流水冲洗。

5、脱水封片：切片依次放入无水乙醇I 5min -无水乙醇II 5min-无水乙醇III5min -二甲苯I5min -二甲苯II5min透明，中性树胶封片。

6、显微镜镜检，图像采集分析。

三、结果判读：

白光下淀粉样蛋白呈红色，细胞核呈浅蓝色

四、注意事项：

1、区分甲状腺胶质、弹力纤维和胶原纤维，可能都会染成红色；

2、分化时要掌握恰当，分化不足，胶原纤维也呈红色，分化过度，淀粉样变蛋白也可脱色。

3、若白光下未见阳性或者阳性很弱，则可在荧光下观察红光，

Congo Red staining report

1 Apparatus and reagents

1.1 Major apparatus

Name	Producer	Model
Dehydrator	DIAPATH	Donatello
Embedding machine	Wuhan Junjie Electronics Co., Ltd	JB-P5
Pathology slicer	Leica	RM2016
Frozen platform	Wuhan Junjie Electronics Co., Ltd	JB-L5
Organizer	KEDEE	KD-P
oven	Labotery	GFL-230
Glass slide	Wanwu	G6004
Upright optical microscope	Nikon	NIKON ECLIPSE E100
Imaging system	Nikon	NIKON DS-U3

1.2 Major reagents

Name	Producer	Code
Ethanol	SCRC	100092683
Xylene	SCRC	10023418
Congo Red dye solution set	Wanwu	G1003
Differentiation solution	Wanwu	G1005-3
Scott Tap Bluing	Wanwu	G1005-4
Neutral gum	SCRC	10004160

2 Procedure

2.1 Dewaxing as followed:

Xylene I for 20 min;

Xylene II for 20 min;

100% ethanol I for 5 min;

100% ethanol II for 5 min;

75% ethanol for 5 min;

Rinsing with tap water ;

2.2 Soak the slices in Congo Red A overnight, rinse with tap water for 2 min.

2.3 Treat the section with Congo Red B for 1s until the positive plaques were obvious, and the background was basically colorless, then rinse with tap water.

2.4 Stain sections with Congo Red C for 1 min, rinse with tap water.Then treat it with Differentiation solution and Scott Tap Bluing, each step required washing with water.

2.5 Dehydrate as followed:

100% ethanol I for 5 min;
100% ethanol II for 5 min;
100% ethanol III for 5 min;
Xylene I for 5 min;
Xylene II for 5 min;
Finally seal with neutral gum.

2.6 Observe with microscope inspection, image acquisition and analysis.

3 Results

Color	Result
Blue	Nucleus
Red (under white light)	Amyloid

4 Precautions

4.1 Please pay attention to distinguishing thyroid glial, elastic fiber and collagen fiber, which may be dyed red together;

4.2 It is necessary to master proper differentiation, insufficient differentiation, collagen fibers will also become red, over-differentiated, amyloid protein will also be decolorized.

4.3 If there is no positive or positive weakly under the white light, you can observe the red light under fluorescence.