

masson 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	上海徠卡仪器有限公司	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
masson 染液套装	Wanwu	G1006
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、切片浸入 Masson A 液中浸泡过夜，自来水洗。
- 3、切片入 Masson B 液及 Masson C 液等比混合的染液，浸染 1min，自来水洗，1%盐酸酒精分化，自来水洗。
- 4、切片入 Masson D 液浸染 6 min，自来水漂洗。
- 5、Masson E 液浸染 1 min。
- 6、不水洗，稍沥干直接入 Masson F 液染 2-30s。
- 7、切片用 1%冰醋酸漂洗分化，两缸无水乙醇脱水。
- 8、透明封片：切片放入第三缸无水乙醇 5min，二甲苯 5min 透明，中性树胶封片。
- 9、显微镜镜检，图像采集分析。

三、结果判读：

胶原纤维呈蓝色；肌纤维、纤维素和红细胞呈红色。

四、注意事项：

- 1、Masson A 液染完后水洗要稍快，切片洗干净即可。
- 2、根据切片的数量对混合的染液进行更换，也可以现配现用；
- 3、Masson D 液的染色程度要控制好，胶原部分不能是红色或者太红，会影响后续 Masson F 液的着色；
- 4、冰醋酸分化过程中，若是分化过度，胶原蓝色太浅，若分化不足，易与红色叠加呈紫蓝色。

Masson 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
Masson dye solution set	Wanwu	G1006
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 Masson A overnight, rinse with tap water.
- 2.3 Masson B, Masson C were prepared into Masson solution according to the ratio of 1:1. Then stain with Masson solution for 1 min, rinse with tap water. And differentiation it with 1% hydrochloric acid alcohol, rinse with tap water.
- 2.4 Soak the slices in Masson D for 6 min, rinse with tap water; Masson E for 1 min; Masson F for 2-30s.
- 2.5 Rinse the slices with 1% glacial acetic acid and then dehydration with two cup of anhydrous ethanol.
- 2.6 100% ethanol for 5 min; Xylene for 5 min; Finally seal with neutral gum.
- 2.7 Observe with microscope inspection, image acquisition and analysis.

3 Results

Color	Result
Blue	Collagen fibers
Red	Muscle fibers; Cellulose; Erythrocyte

4 Precautions

- 4.1 After treat the slices with Masson A , raine with the water should be quickly. It is sufficient to just wash the slices clean.
- 4.2 Replace the mixed dye solution according to the number of slices. And it can also be formulated when you need.
- 4.3 The staining degree of Masson D solution should be well controlled. The collagen part cannot be red, which will affect the subsequent coloration of Masson F solution;
- 4.4 During the differentiation of glacial acetic acid, if it is over-differentiated, the collagen blue will be too light, if it is insufficiently differentiated, it will be easily superimposed with red to become purple-blue .