

维多利亚蓝染色实验报告

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
载玻片及盖玻片	江苏世泰实验器材有限公司	10127105P-G
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
维多利亚蓝染液套装	武汉谷歌生物科技有限公司	G1055
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、组织酸化：组画笔圈住组织，滴加维多利亚蓝酸化液（维多利亚蓝 A 与维多利亚蓝 B 1:1 混匀，现配现用）于组织上氧化 5min，切片入两缸水浸洗共 10s，稍甩干切片多余水分。

3、组织漂白：维多利亚蓝 C 于组织上漂白 2min，切片入 3 缸水浸洗，各 5s，稍甩干切片多余水分。

4、组织着色：70%的乙醇浸洗，切片入维多利亚蓝染液 D 浸染 24h(加盖)，70%的乙醇浸洗 2 次，每次约 10s，至玻片表面无染液附着为止，流水稍洗。

5、组织分化：镜检弹力纤维着色程度，着色深，切片入 75%乙醇分化 5s，自来水洗终止分化，镜检，反复分化水洗和镜检，至弹力纤维呈蓝色，背景淡蓝色或近乎无色；

6、复染：切片入维多利亚蓝染液 E 染色 1-5min，流水冲洗。

7、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。

8、显微镜镜检，图像采集分析。

三、结果判读：

弹力纤维呈蓝色，细胞核呈红色。

Victoria blue staining experimental report

1. Lab equipment and reagents

A. Lab equipment

Items	Manufacturer	Model
Dehydrator	DIAPATH	Donatello
embedding machine	Wuhan Junjie Electronics Co., Ltd.	JB-P5
Pathology microtome	Shanghai Leica Instruments Co., Ltd.	RM2016
Frozen platform	Wuhan Junjie Electronics Co., Ltd.	JB-L5
Water Bath-Slide Drier	Zhejiang Jinhua Kedi Instrumental Equipment CO.,LTD	KD-P
Laboratory oven	Tianjin Labotery Instrument Equipment Co., Ltd.	GFL-230
Microscope slide &cover glass	Citotest Labware Manufacturing Co.,Ltd	10127105P-G
Upright optical microscope	Nikon Japan	Nikon Eclipse E100
Imaging system	Nikon Japan	NIKON DS-U3

B. Chemical Reagents

Items	Manufacturer	Model
Absolute alcohol	Sinopharm Chemical Reagent Co., Ltd.	100092683
Xylene	Sinopharm Chemical Reagent Co., Ltd.	10023418
Victoria blue staining kit	Wanwu	G1055
Neutral balsam	Sinopharm Chemical Reagent Co., Ltd.	10004160

2. Experimental steps

(1) Paraffin section deparaffinization and rehydration: put the slides into xylene I 20minutes-xylene II 20 minutes-absolute ethanol I 5 min-absolute ethanol II 5 min-75% alcohol for 5 min, tap water washing.

(2) Tissue acidification: draw a circle to enclose the tissue, add Victoria blue acidification solution (Victoria Blue A and Victoria Blue B 1:1 and mix, ready for usage), oxidize the tissue for 5

minutes, dip the slides in two cylinders of water for 10s, and dry the slides to remove excess water.

(3) Tissue bleaching: The tissues are bleached with Victoria Blue C for 2 minutes, put the slides soak in 3 cylinders of water for 5 s, and dry the slides to remove excess water.

(4) Tissue coloring: dip into 70% ethanol, put the slides into Victoria blue staining D for 24h (covered), dip into 70% ethanol washing for 2 times, 10s each time, until there is no staining solution attached to the slide surface, running water washing.

(5) Tissue differentiation: checking the degree of staining of elastic fibers under a microscope, if deep coloring, put slides into 75% ethanol for 5s, tap water washing to terminate differentiation microscope examination, repeatedly differentiation, washing and microscope examination, until elastic fiber is blue, light blue background or nearly colorless;

(6) Counterstaining: staining with Victoria Blue Staining Solution E for 1-5 minutes and running water washing.

(7) Dehydration and sealing: put the slide into absolute ethanol I 5min-absolute ethanol II 5min-absolute ethanol III 5min-xylene I 5min-xylene II 5min transparent, neutral balsam sealing.

(8) Microscope examination, image collection and analysis.

3. Results

The elastic fiber is blue, nucleus is red.