改良天狼星红染色试剂盒

货号： G1472
规格： 2×50mL/2×500mL
保存： 室温，避光保存，有效期为 1 年。

产品组成：

名称		2×50mL	2×500mL	保存
试剂(A):铁苏木素染色工作液	试剂(A1):铁苏木素储备液	25mL	250mL	室温，避光
	试剂(A2):铁苏木素稀释液	25mL	250mL	室温，避光
临用前按照A1:A2=1：1的比例混匀制备染色工作液，建议在4小时内使用。				
试剂(B):天狼星红染色液		50 mL	500mL	室温，避光

产品介绍：

胶原纤维(Collagen Fiber)是结缔组织中分布最广含量最多的一种纤维，广泛分布于各种脏器，其中I型胶原纤维主要是骨、皮肤、肌腱；II型胶原纤维主要是软骨胶原；III型胶原纤维主要在胚胎组织、成人血管、胃肠道；IV型胶原纤维主要在基膜中。天狼星红与其衬染液都是强酸性染料，易与胶原分子中的碱性基团结合，吸附牢固。偏振光镜检查，胶原纤维有正的单轴双折射光的属性，与天狼星红复合染色液结合后，可增强双折射，提高分辨率，从而区分两型胶原纤维。

改良天狼星红染色试剂盒主要由铁苏木素染色液和天狼星红染色液组成，不含苦味酸。主要用于各种组织病变时对胶原纤维异常变化的研究，在普通光学显微镜下心脏血管等组织的胶原纤维被染成红色，在偏振光镜下对各种纤维化病变的分型和分级研究有一定的帮助作用。采用免疫组化技术也可显示I、III型胶原纤维，但所用抗体昂贵，操作费时，而采用天狼星红染色试剂便宜，操作简单。

自备材料：

10%福尔马林固定液、普通光学显微镜或偏振光显微镜

操作步骤：*（仅供参考）*

1. 组织固定于 10%福尔马林固定液，常规脱水包埋。
2. 切片切 3-8μm，常规脱蜡至水。
3. 临用前配制铁苏木素染色液，滴加染色 5-10min，蒸馏水洗 10-20s 洗去多余染色液。
4. （可选）自来水浸洗 5-10min，蒸馏水清洗 3 次，每次 5-10s。
5. 天狼星红染色液滴染 15-30min，对于较易上色组织可把染色时间控制在 5-10min 内。
6. 流水稍微冲洗，去除切片表面染液。
7. 从 75%开始系列乙醇脱水，二甲苯透明，中性树胶封固。

染色结果：

1）光学显微镜观察结果	胶原纤维	红色
	细胞核	棕褐色到黑色
	肌纤维	黄色
2）偏振光镜观察结果	I 型胶原纤维	强橙黄色或亮红色
	III 型胶原纤维	绿色

注意事项：

1. 为使在偏振光镜下显示清晰，本法的切片厚度以 6～7μm 为宜。
2. 不进行铁苏木素染色细胞核可能着浅红色，属于正常现象，用铁苏木素染色后则不会出现这种状况。
3. 本法染色稳定，不易褪色。
4. 为了您的安全和健康，请穿实验服并戴一次性手套操作。

Modified Sirius Red Stain Kit（No Picric Acid）

Cat: G1472
Size: 2×50mL/2×500mL
Storage: RT, avoid light, valid for 1 year.

Kit Components

Reagent		2×50mL	2×500mL	Storage
Reagent(A):Iron Hemat--oxylin Working solution	A1:Iron Hematoxylin Stock Solution	25mL	250mL	RT, avoid light
	A2:Iron Hematoxylin Diluent Solution	25mL	250mL	RT, avoid light
Before use, mix A1 with A2 in equal to form Iron Hematoxylin Working Solution，it is recommended to use within 4 h.				
Reagent(B):Sirius Red Staining Solution		50 mL	500 mL	RT, avoid light

Introduction

Collagen fiber is the most widely distributed and abundant fiber in connective tissue. It is widely distributed in various organs. Type I collagen fibers are mainly bone, skin and tendon; Type II collagen fibers are mainly cartilage collagen; Type III collagen fibers are mainly found in embryonic tissues, adult blood vessels and gastrointestinal tract; Type IV collagen fibers are mainly in the basement membrane. Sirius red and its lining dye solution are strong acid dyes, which are easy to combine with the basic groups in collagen molecules and have strong adsorption. Polarized light microscopy shows that the collagen fibers have the property of positive uniaxial birefringence. When combining with Sirius red compound staining solution, it could enhance the birefringence and improve the resolution, so as to distinguish the two types of collagen fibers.

Modified Sirius Red Stain Kit（No Picric Acid） is mainly composed of Iron Hematoxylin Stain and Sirius Red Stain, without picric acid. It is mainly used in the study of collagen fiber abnormality or fiber proliferation in various tissue diseases. Under the ordinary optical microscope, the collagen fibers of heart and blood vessels and other tissues are dyed red, which is helpful for the classification and grading of various fibrotic diseases under the polarized light microscope. Immunohistochemical technique can also display type I and III collagen fibers, but the antibodies used are expensive and time-consuming, while Sirius red staining reagent is cheap and easy to operate.

Self Provided Materials

10% formalin fixative solution, ordinary optical microscope or polarized light microscope

Protocol(*for reference only*)

1. Fix the tissue in 10% formalin fixative solution and embed by routine dehydration.
2. Cut the slices into 3-8 μm. Conventional dewaxing to water.
3. Prepare Iron Hematoxylin Staining Solution before use, stain the section with it for 5-10min, and wash it with distilled water for 10-20s to remove the excess staining solution.
4. (Optional) Immerse in tap water for 5-10min, and wash with distilled water for 3 times, 5-10s each time.
5. Sirius Red Staining Solution shall be used for dripping dyeing for 15-30min. For tissues that are easy to be stained, the dyeing time can be controlled within 5-10min.
6. Rinse slightly with running water to remove the staining liquid on the surface of the slice.
7. Dehydrate by series of ethanol and transparent by xylene, finally seal with neutral gum.

Result

1）Optical microscope observations	Collagen fiber	Red
	Nucleus	Brown to Black
	Muscle fiber	Yellow
2）Polarized light microscope observations	Type I collagen fiber	Strong Orange Yellow or Light Red
	Type III collagen fiber	Green

Note

1. In order to display clearly under the polarized light microscope, the slice thickness should be 6-7μm.
2. The nucleus without iron hematoxylin staining may be light red, which is a normal phenomenon, but it will not occur after iron hematoxylin staining.
3. This method is stable and not easy to fade.
4. For your safety and health, please wear experimental clothes and disposable gloves.