V02



普鲁士蓝染色试剂盒(伊红法)

货号: G1424

规格: 2×50mL/2×100mL

保存:室温,避光保存,有效期1年。

产品组成:

٠.		A. A				
		名称	2×50mL	2×100mL	保存	
	试剂(A): Perls	A1: Perls 染色液 A	25mL	50mL	室温,避光	
	染色工作液	A2: Perls 染色液 B	25mL	50mL	室温	
	临用前,取 A1、A2 等量混合,即为 Perls stain,不宜提前配制。					
	试剂(B): 伊红染色	50mL	100mL	室温,避光		

产品介绍:

普鲁士蓝反应(Prussian Blue Reaction)是一种对含有颗粒铁的细胞或组织中的非血红素铁(含铁血黄素、铁转运蛋白、铁沉积等)进行染色的方法,在缺血性贫血、溶血性贫血、血色素沉着症和血色素代谢异常等疾病验证方面起重要作用。该染色方法可以区分含铁血黄素和其他色素,但是受限于染色原理,只适用三价铁离子含量较多的组织和细胞染色,联合其他增强试剂可用于铁死亡的检测。

普鲁士蓝反应最早被 Perls 提出,因此最常被称为 Perl 铁染色,但是自 1867 年提出以来有众多化学家和病理学家对该反应进行优化,其中优化比较完善使用也比较多的一种称为 Lillie 法,能够对二价铁和三价铁进行分别标记。本试剂盒即为即用型的 Lillie 法三价铁染色试剂盒,采用优质原料配制,试剂稳定,染色特异性良好。

自备材料:

固定液(见注意事项 2)、系列乙醇、蒸馏水

操作步骤: (仅供参考)

- 1、组织推荐固定于中性福尔马林固定液(10%)中,常规脱水包埋。(见注意事项2)
- 2、石蜡切片推荐厚度 5μm, 常规脱蜡至水。冰冻切片推荐厚度 10μm, 常规复温复水。
- 3、切片滴加 Perls 染色工作液染色 15~30min,蒸馏水冲洗 2~5min。(见注意事项 4)
- 4、滴加伊红染色液覆盖切片染色 15~30s, 蒸馏水快洗 1~5s。
- 5、75%/85%/95%乙醇快速脱水,每级2-3s,无水乙醇脱水两次,每次30s,二甲苯透明,中性树胶封片。

染色结果:

含铁血黄素或铁蛋白中的三价铁	蓝色
组织背景	红色

对照(可选):

取相同对照切片脱蜡至水,1%盐酸水溶液室温处理20min后正常染色。结果为染色阴性。

取少量铁盐配成不同浓度的水溶液试样(常用 0.1%氯化铁或硫酸铁水溶液),滴入一滴染色工作液应观察到明显蓝色沉淀出现,结果为试剂阳性。

取少量亚铁盐配成不同浓度的水溶液试样(常用 0.1%硫酸亚铁水溶液),滴入一滴染色工作液应观察到 无明显变化或有微量变色,结果为试剂阴性。

注意事项:

- 1、石蜡切片脱蜡应充分,系列乙醇应及时换新。
- 2、该染色法组织取材应避免使用酸性固定液或螯合剂处理导致铁离子丢失,推荐使用 G2161-中性福尔马林固定液(10%)或 P1110-4%组织细胞固定液进行组织固定。
- 3、整个操作过程中须避免铁离子污染,清洗用水以蒸馏水为宜,因自来水常内含铁质。
- 4、 该染色对铁含量有要求,染色前建议进行使用脾脏石蜡切片或铁盐进行对照染色确定试剂有效性。
- 5、为了您的安全和健康,请穿实验服并戴一次性手套操作。

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Prussian Blue Iron Stain Kit (With Eosin)

Cat: G1424

Size: 2×50 mL/ 2×100 mL

Storage: RT, avoid light, valid for 1 year.

Kit Components

Reagent		2×50mL	2×100mL	Storage	
Reagent(A):Perls Stain	A1: Perls Stain A	25mL	50mL	RT, avoid light	
Working Solution	A2: Perls Stain B	25mL	50mL	RT	
Before use, mix equal parts of A1 and A2 to form Perls Stain. It is not suitable to prepare in advan					
Reagent(B): Eosin Soluti	50mL	100mL	RT, avoid light		

Introduction

The Prussian Blue Reaction is a method of staining non heme iron (containing heme, iron transporters, iron deposition, etc.) in cells or tissues containing granular iron. It plays an important role in the validation of diseases such as ischemic anemia, hemolytic anemia, hemochromatosis, and abnormal hemoglobin metabolism. This staining method can effectively distinguish between hemosiderin and other pigments, but due to the principle of staining, it is only suitable for staining tissues and cells with a high content of trivalent iron ions. In combination with other enhancing reagents, it can also be used for the detection of iron death models.

The Prussian blue reaction was first proposed by Perls and is therefore most commonly referred to as the Perl iron staining method. However, since its proposal in 1867, numerous chemists and pathologists have optimized this reaction, among which the Lillie method, which is more widely used and well optimized, can label divalent and trivalent iron separately. This kit is a ready to use Lillie method ferric iron staining kit, prepared with high-quality raw materials, with stable reagents and good staining specificity.

Self Provided Materials

Fixative(See Note 2), Series of ethanol, Distilled water

Protocol(*for reference only*)

- 1. The organization recommends fixation in neutral formalin fixative (10%), conventional dehydration and embedding. (See Note 2)
- 2. The recommended thickness for paraffin sections is 5μm. Conventional dewaxing to water. The recommended thickness for frozen sections is 10μm. Regular rewarming and rehydration.
- 3. Stain with Perls Stain Solution for 15-30min and rinsed with distilled water for 2-5min.(See Note 4)
- 4. Re-dye with Eosin Solution for 15-30s. Quickly rinse with distilled water for 1-5s.
- 5. Dehydrate by 75%/85%/95% ethanol for each 2-3s, then dehydrate by absolute ethanol twice, each for 30s. Transparent by xylene, then seal with resinene.

Result

Hemosiderin or Ferric Iron	Blue	
Background	Red	

Negative Control(Optional)

Take the same control section and dewax it to water, then treat it with a 1% hydrochloric acid solution at room temperature for 20min before staining normally. The result is negative for staining.

Take some ferrous salt and prepare an aqueous solution samples (commonly 0.1% ferrous sulfate aqueous solution), and drop a drop of staining solution, the result is negative for kit.

Take some iron salt and prepare an aqueous solution samples (commonly 0.1% ferric chloride or ferric sulfate aqueous solution). Drop a drop of the staining solution, the result is a positive for kit.

Note

- 1. Section dewaxing should be as clean as possible. Series of ethanol should be replaced frequently.
- 2. This staining method for tissue sampling should avoid using acidic fixative or chelating agent treatment that may cause iron ion loss. It is recommended to use G2161-neutral formalin fixative (10%) or P1110-4% tissue cell fixative for tissue fixation.
- 3. During the whole operation process, the container should be clean and avoid the use of metal iron products. When washing sections and containers, distilled water is suitable, because ordinary water contains iron.
- 4. This staining requires iron content, and it is recommended to use spleen paraffin sections or iron salts for comparative staining before staining to determine the effectiveness of the reagent.

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