V02

布氏杆菌染色试剂盒

货号：G3190
规格：2×50mL
保存：室温，避光保存，有效期 6 个月。

产品组成：

名称	2×50mL	保存
试剂(A)：沙黄染色液	50mL	室温，避光
试剂(B)：复染液	50mL	室温，避光

产品介绍：

布氏杆菌(Brucella)是一种革兰氏阴性的不运动细菌，牛、羊、猪等动物最易感染。布氏杆菌染色又称科兹洛夫斯基染色戒柯兹洛夫斯基染色。布氏杆菌染色液采用沙黄为染色剂，临床标本直接涂片，背景干净，胞核胞质对比强烈，胞内吞噬体清晰易辨认，细菌染色特征典型。所有操作应置于二级生物安全柜中进行，一旦发现绿色细菌中有红色点状细菌，应高度怀疑为布氏杆菌，及时通知 CDC，防止疫情扩散。

自备材料：

接种环或挑取细菌的其他工具、酒精灯 、载玻片、光学显微镜

操作步骤：(*仅供参考*)

1、 涂片：取待检细菌，于载玻片中央涂成薄层，尽量薄。如果需做对照，可以将大肠杆菌与待检细菌混合涂片。
2、 干燥：涂片后在室温下自然干燥，也可在酒精灯上略加温，使之迅速干燥。
3、 固定：手持载玻片一端，标本面朝上，在酒精灯的火焰外侧快速来回移动3~5次，每次1s，温度不宜过高，防止菌体蛋白变性，放置待凉后染色。
4、 染色：滴加沙黄染色液染色，在酒精灯的火焰上微微加热至出现蒸汽为止，通常需要30~60s。清水洗去染色液。
5、 复染：滴加复染液染色2~3min。水洗，吸干。
6、 镜检。

染色结果：

布氏杆菌	红色
大肠杆菌或其他菌	绿色

注意事项：

1、 涂片之前应事先在背面做好圆圈标记，以便判断后续试验的位置。
2、 取细菌时，应注意自我防护，拔或塞试管塞时，应将试管口通过火焰略加烧灼，最后将接种环在火焰上烧灼灭菌。
3、 加热固定涂片时，应注意玻片勿太靠近火焰，一般要求玻片温度不超过60℃，以玻片背面触及手背皮肤不觉过烫为宜。
4、 待检细菌培养时间也会影响染色，阳性菌培养时间过长或已死亡或细菌溶解，都常呈阴性反应。
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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Brucella Stain Kit(Kirschner Method)

Cat: G3190
Size: 2×50mL
Storage:RT, avoid light, valid for 6 months.

Kit Components

Reagent	2×50mL	Storage
Reagent (A): Safranine Staining Solution	50mL	RT, avoid light
Reagent (B): Redyeing Solution	50mL	RT, avoid light

Introduction

Brucella is a kind of gram-negative non motile bacteria, which is most susceptible to infection in cattle, sheep, pigs and other animals. Brucella staining is also called kozlovsky staining. The Brucella staining solution takes safranine as the staining agent. For clinical specimen, directly smear. The background is clean, the contrast between nucleus and cytoplasm is strong, the phagocyte in the cell is clear and easy to identify, and the characteristics of bacterial staining are typical. All operations shall be taken in clean bench. Once find red punctate bacteria in the green bacteria, it shall be highly suspected to be Brucella, and timely notify CDC to prevent the spread of the epidemic.

Self Provided Materials

Inoculating ring or other tools for picking up bacteria, Alcohol lamp, Slide, Optical microscope

Protocol(*for reference only*)

1. Smear: take the bacteria to be tested, and smear a layer in the center of the slide as thin as possible. If you need to make a comparison, you can make a mixture smear of E. coli and bacteria to be tested.
2. Dry: after smearing, dry naturally at room temperature or heat slightly on the alcohol lamp to make it dry quickly.
3. Fixation: hold one end of the slide with the specimen face up, and move it back and forth 3 to 5 times quickly outside the flame of the alcohol lamp for 1 s each time. The temperature should not be too high to prevent the denaturation of the bacterial protein. Dye it after cooling.
4. Dyeing: drop Safranine Staining Solution on the smear and dye, heat slightly on the flame of alcohol lamp until steam appears, usually for 30-60s. Remove the staining solution with clear water.
5. Drop Redyeing Solution and dye for 2-3min.Wash with water and absorb the exceed water.
6. View under the optical microscope.

Result

Brucella	Red
E. coli or other bacteria	Green

Note

1. Before smearing, make circle mark on the back in advance to judge the position of subsequent test.
2. When taking bacteria, pay attention to self-protection. When pulling or plugging the tube plug, slightly burn the tube mouth through the flame, and finally burn the inoculation ring on the flame for sterilization.
3. When heating to fix smear, note that the slide is not too close to the flame. Generally, the temperature of the slide is not more than 60 ℃.
4. The culture time of the bacteria to be tested will also affect the staining. If the culture time of the positive bacteria is too long or the bacteria are dead or dissolved, it will often show negative reaction.
5. For your safety and health, please wear experimental clothes and disposable gloves.