



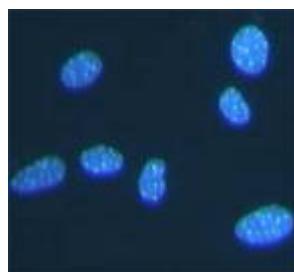
碧云天生物技术/Beyotime Biotechnology  
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## 细胞凋亡-Hoechst染色试剂盒

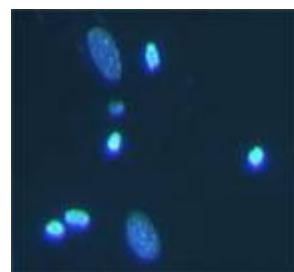
产品编号	产品名称	包装
C0003	细胞凋亡—Hoechst染色试剂盒	100次

### 产品简介:

➤ 碧云天生产的细胞凋亡—Hoechst染色试剂盒(Hoechst Staining Kit)为您提供了一种经典而又快速简便的细胞凋亡检测方法。细胞发生凋亡时，染色质会固缩。所以Hoechst 33258染色后，在荧光显微镜下观察，正常细胞的细胞核呈正常的蓝色，而凋亡细胞的细胞核会呈致密浓染，或呈碎块状致密浓染，颜色有些发白。典型的Hoechst 33258染色后的细胞图片参考下图。



正常细胞核



刺激后有致密浓染的凋亡细胞

- 只需25分钟即可完成细胞凋亡检测的整个过程。
- 本试剂盒提供了固定液，染色液，及抗荧光淬灭封片液。
- 本试剂盒对贴壁细胞，悬浮细胞和组织切片均适用。
- 足够检测100个样品。

### 包装清单:

产品编号	产品名称	包装
C0003-1	固定液	50ml
C0003-2	Hoechst 33258染色液	50ml
C0003-3	抗荧光淬灭封片液	5ml
—	说明书	1份

### 保存条件:

4°C保存，Hoechst 33258染色液需4°C避光保存，半年有效。

### 注意事项:

- 需可以观察蓝色荧光的荧光显微镜或激光共聚焦显微镜。
- 需PBS或0.9% NaCl溶液。
- 需自备盖玻片与载玻片。盖玻片与载玻片可以向碧云天订购。
- 荧光物质均易发生淬灭，染色后的样品宜避光保存。可以向碧云天选购各种型号的载玻片存储盒。
- 在使用抗淬灭封片液的情况下可以减缓淬灭，但仍宜尽量避光。
- 本产品仅限于专业人员的科学研究用，不得用于临床诊断或治疗，不得用于食品或药品，不得存放于普通住宅内。
- 为了您的安全和健康，请穿实验服并戴一次性手套操作。

### 使用说明:

#### 1. 贴壁细胞

- 取洁净盖玻片在70%乙醇中浸泡5分钟或更长时间，无菌超净台内吹干或用无菌的PBS或0.9%NaCl等溶液洗涤三遍，再用细胞培养液洗涤一遍。将盖玻片置于六孔板内，种入细胞培养过夜，使约为50%-80%满。
- 刺激细胞发生凋亡后，吸尽培养液，加入**0.5ml固定液，固定10分钟或更长时间(可4°C过夜)**。
- 去固定液，用**PBS或0.9% NaCl洗两遍，每次3分钟**，吸尽液体。洗涤时宜用摇床，或手动晃动。
- 加入**0.5ml Hoechst 33258染色液，染色5分钟**。也宜用摇床，或手动晃动数次。
- 去染色液，用**PBS或0.9% NaCl洗两遍，每次3分钟**，吸尽液体。洗涤时宜用摇床，或手动晃动。
- 滴一滴抗荧光淬灭封片液于载玻片上，盖上贴有细胞的盖玻片，让细胞接触封片液，尽量避免气泡。
- 荧光显微镜可检测到呈蓝色的细胞核。激发波长350nm左右，发射波长460nm左右，图谱请参考下图。

## 2. 悬浮细胞

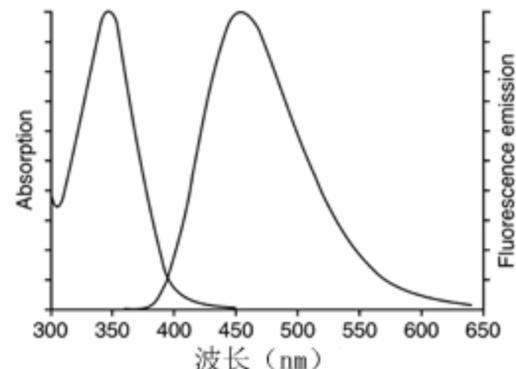
- a. 离心收集细胞样品于1.5ml离心管内，加入**0.5ml固定液**，缓缓悬起细胞，**固定10分钟**或更长时间(可4°C过夜)。
- b. 离心去固定液，用**PBS或0.9% NaCl洗两遍，每次3分钟**。洗涤期间手动晃动数次。
- c. 离心后吸去大部分液体保留约50μl液体，再缓缓悬起细胞，滴加至载玻片上，尽量使细胞分布均匀。
- d. 稍晾干，使细胞贴在载玻片上不易随液体流动。
- e. 均匀滴上**0.5ml Hoechst 33258染色液，染色5分钟**。用吸水纸从边缘吸去液体，微晾干。
- f. 去染色液，用**PBS或0.9% NaCl洗两遍，每次3分钟**，吸尽液体。洗涤时宜用摇床，或手动晃动。
- g. 滴一滴抗荧光淬灭封片液于载玻片上，盖上一洁净的盖玻片，尽量避免气泡。
- h. 荧光显微镜可检测到呈蓝色的细胞核。激发波长350nm左右，发射波长460nm左右，图谱请参考下图。

## 3. 组织切片

- a. 常规包埋切片后，根据切片的不同类型，处理至可以用于免疫组化染色。
- b. **PBS或0.9% NaCl洗两遍，每次3分钟**，吸尽液体。洗涤时宜用摇床，或手动晃动。可在六孔板中操作。
- c. 加入**0.5ml Hoechst 33258染色液，染色5分钟**。也宜用摇床，或手动晃动数次。
- d. 去染色液，用**PBS或0.9% NaCl洗两遍，每次3分钟**，吸尽液体。洗涤时宜用摇床，或手动晃动。
- e. 小心将切片置于载玻片上，滴一滴抗淬灭封片液，盖上一洁净的盖玻片，尽量避免气泡。
- f. 荧光显微镜可检测到呈蓝色的细胞核。激发波长350nm左右，发射波长460nm左右，图谱请参考右图。

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Hoechst 33258 的吸收光谱和发射光谱  
左侧峰为吸收光谱，右侧峰为发射光谱

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