Beijing Solarbio Science & Technology Co., Ltd
Tel: 400-968-6088
Fax: 010-56371281
Http://www.solarbio.com

福尔马林-EDTA 脱钙液

货号： G2520
规格： 500mL
保存： 室温保存，有效期 1 年。

产品介绍：

在组织切片过程中，一些组织内含有骨质或钙化灶时，含钙的组织不宜直接用石蜡包埋切片。这是因为钙和石蜡之间的密度不同，较难切出完整的切片。对含钙组织最好固定之后，再进行脱钙或二者同时进行。然后进行下游操作如脱水、透明、浸蜡、包埋、切片。用于脱钙的试剂很多，脱钙剂包括有机酸、无机酸、乙二胺四乙酸(EDTA)以及电解法脱钙。EDTA 是一种相对较好的螯合脱钙剂，对组织结构影响最小，可以较好的保存组织的某些酶类，经 EDTA 脱钙后的组织可以进行免疫组化和原位杂交染色。但是该法脱钙速度太慢，一般脱需要数周至数月。

福尔马林-EDTA 脱钙液主要由福尔马林、EDTA 等组成，pH 值约为 7.2～7.4，相对于常规 EDTA 脱钙液，该试剂脱钙后对组织结构损害小，但脱钙速度更慢。其优点是：①经 EDTA 脱钙的组织染色结果好；②对组织的结构损害比常规 EDTA 脱钙液更小。其缺点是：①脱钙速度很慢，不适合常规标本脱钙使用；②脱钙后组织会稍微变硬；③不宜用化学方法确定脱钙终点。

操作步骤：（*仅供参考*）

1. 骨组织脱钙时，取材不易过厚，一般大约 5mm。
2. 组织固定后，用 PBS 清洗 3 次，每次 20min。
3. 组织用蒸馏水清洗 3 次，每次 20min。
4. 组织转移至 20~30 倍体积的 EDTA 脱钙液中，脱钙 10~30 天或更长时间。如果想加快脱钙速度，可以置于 37℃进行脱钙。如果必要，更换新的 EDTA 脱钙液继续脱钙，每周更换一次直至终点。
5. 用蒸馏水冲洗数次。
6. 常规脱水、包埋。

注意事项：

1. 厚度 5mm 的骨组织块脱钙时间一般脱钙 10~30 天即可。
2. 长期（间隔 1 个月以上）不用建议放置阴凉避光处保存。
3. 适当加温能加快脱钙的速度，一般维持在 37～40℃，温度过高容易使骨组织松散解体，尤其不可大于 60℃。
4. 脱钙应彻底，防止脱钙不足或过度。脱钙程度应控制在不影响组织切片的同时尽量缩短脱钙时间，以免脱钙过长引起组织损害。
5. 脱钙用具避免使用金属容器，尽量使用玻璃容器。
6. 骨组织脱钙应先固定后脱钙或脱钙固定同时进行，不应先脱钙后固定，以便减少组织的损伤程度。
7. 每隔一段时间检测一次脱钙程度，脱钙过度会增加组织的损伤程度，影响染色结果。
8. 为了您的安全和健康，请穿实验服并戴一次性手套操作。

附录：

脱钙终点的测定(物理法)：采用针刺、手掐、钳夹等方法，当骨组织变软或针刺时没有阻力感即可终止脱钙。物理检测法会对组织结构有一定的损害， 尽量避免用力过大或反复检测。

Formalin-EDTA Decalcifying Solution

Cat: G2520
Size: 500mL
Storage: RT, valid for 1 year.

Introduction

In the process of tissue sectioning, when some tissues contain bone or calcification, the tissue containing calcium should not be directly embedded in paraffin. This is because the density between calcium and paraffin is different, it is difficult to cut a complete section. It is better to fix the calcium containing tissue before decalcification or conduct both at the same time. Then continue operations such as dehydration, transparency, wax immersion, embedding and slicing. There are many decalcification reagents, including organic acid, inorganic acid, EDTA and electrolytic decalcification. EDTA is a relatively good chelating decalcifying solution, which has the least influence on the tissue structure and can better preserve some enzymes of the tissue. The tissue after decalcification by EDTA can be stained by immunohistochemistry and in situ hybridization. However, the speed of decalcification is too slow, and it usually takes weeks to months.

Formalin-EDTA Decalcifying Solution is mainly composed of formalin, EDTA and so on. The pH value is 7.2-7.4. Compared with the conventional EDTA decalcification solution, the solution has less damage to the tissue structure after decalcification, but the decalcification speed is slower. The advantages of this method are: ① the result of staining is good after EDTA decalcification; ② the damage to tissue structure is smaller than that of conventional EDTA decalcification solution. The disadvantages are: ① the speed of decalcification is very slow, which is not suitable for the use of conventional specimen decalcification; ② the tissue will be slightly hard after decalcification; ③ it is not suitable to determine the end point of decalcification by chemical method.

Protocol(*for reference only*)

1. When the bone tissue is decalcified, pick up the material avoiding too thick, generally about 5mm.
2. If it is not used for a long time (interval of more than 1 month). It is recommended to store it in a cool and dark place.
3. After fixing the tissue, wash with PBS for three times and each time for 20 min.
4. Wash the tissue with distilled water for three times and each time for 20 min.
5. Transfer the tissue to 20-30 times volume of Formalin-EDTA Decalcifying Solution to decalcify for 10-30 days or more. If you want to speed up the decalcification, you can put it at 37 ℃. If necessary, replace with new Formalin-EDTA Decalcifying Solution to continue decalcification once a week until the end point.
6. Rinse several times with distilled water.
7. Conventional dehydration and embedding.

Note

1. The decalcification time of 5 mm thick bone tissue block is generally 10-30 days.
2. Proper heating can speed up decalcification, generally maintain at 37 ~ 40 ℃, too high temperature is easy to cause bone tissue loose disintegration, especially avoid over 60 ℃.
3. Decalcification should be thorough to prevent insufficient or excessive decalcification. The degree of decalcification should be controlled to shorten the decalcification time as much as possible without affecting the tissue section, so as to avoid tissue damage caused by too long decalcification.
4. Avoid using metal containers for decalcification appliances, and try to use glass containers.
5. It is better to fix the calcium containing tissue before decalcification or conduct both at the same time in order to reduce the degree of tissue damage.
6. The degree of decalcification should be detected in a while. Excessive decalcification will increase the degree of tissue damage and affect the staining results.
7. For your safety and health, pl ease wear experimental clothes and disposable gloves.

Appendix:

Determination of the end point of decalcification (physical method): acupuncture, hand pinching, clamp and other methods are used to stop decalcification when the bone tissue becomes soft or there is no sense of resistance during acupuncture. Physical detection will damage the tissue structure to some extent, and try to avoid excessive force or repeated detection.