

冰冻切片制片实验报告

一、实验器材及试剂

1、实验器材

名称	厂家	型号
冰冻切片机	Thermo	CRYOSTAR NX50
切片刀	上海徕卡仪器有限公司	LEICA 819
防脱载玻片	Wanwu	

2、主要实验试剂

试剂	厂家	货号
OCT 包埋剂	Sakura	4583
固定液	Wanwu	G1101
蔗糖	国药集团化学试剂有限公司	57-50-1

二、冰冻切片制作

- 组织固定：**需要固定的新鲜组织于固定液中固定 24h 以上。而低温运输存放的新鲜直接切片的组织则跳过固定以及脱水的步骤，进行 OCT 包埋并切片。
- 脱水：**将组织从固定液取出放于 15%的蔗糖溶液内 4℃冰箱脱水沉底后转入 30%的蔗糖溶液内 4℃冰箱脱水沉底。
- OCT 包埋：**将脱好水的组织取出用滤纸将表面水稍吸干后，用手术刀将目的部位组织修平整，切面朝上放于样本托上，组织周围滴上 OCT 包埋剂，将样本托放在冰冻切片机的速冻台上速冻包埋，OCT 变白变硬后即可进行切片。
- 切片：**将样本托固定于切片机上，先粗切将组织面修切平整后即可开始切片，切片厚度 8-10 μ m，将干净的载玻片平放于切出的组织片上方即可将组织贴于载玻片上。片子写好标签后-20℃保存备用。

Frozen slides experimental report

1. Experimental equipments and reagents

1.1 Experimental equipments

Name	Manufacturer	Model
Frozen section machine	Thermo	CRYOSTAR NX50
Slicer	Shanghai Leica Instrument Co., Ltd	LEICA 819
Adhesion microscope slide	Wanwu	

1.2 Main experimental reagents

Reagent	Manufacturer	Article number
OCT embedding agent	Wanwu	G6059-110ML
Fixative	Wanwu	G1101
Sucrose	Sinopharm Group Chemical Reagent Co. LTD	57-50-1

2. Frozen slides making

2.1 Tissue fixation: Fresh tissues that need to be fixed were fixed in the fixative for more than 24 hours. However, the fresh and directly sliced tissues stored in low temperature transportation skip the steps of fixation and dehydration, and are embedded and sliced by Oct.

2.2 Dehydration: The tissue was removed from the fixed solution and placed in 15% sucrose solution for 4 °C refrigerator dehydration, then transferred into 30% sucrose solution for 4 °C refrigerator dehydration.

2.3 OCT embedding: Take out the dehydrated tissue, drain the surface water with filter paper, smooth the target tissue with scalpel, put the section face up on the specimen chuck, drop OCT embedding agent around the tissue, put the specimen chuck on the quick freezing table of the frozen section machine for quick freezing and embedding. When OCT becomes white and hard, it can be sectioned.

2.4 Section: The specimen chuck was fixed on the slicer, the tissue surface was rough cut, and then sliced. The slice thickness was 8-10 μ M. the clean slide was placed on the top of the cut tissue slide, and the tissue was pasted on the slide.

After the film is labeled, it should be stored at - 20 °C for future use.