Oil Red O Staining Solution

Introduction

Oil Red O is a lipid-soluble azo dye. When lipids in a sample (such as lipid droplets and triglycerides) come into contact with Oil Red O, the dye molecules diffuse physically into the lipids and form noncovalent interactions with them, causing the lipids to appear bright red. Oil Red O shows selectivity in binding to lipids, with a strong affinity for neutral fats but weaker binding to polar lipids such as phospholipids. Therefore, it stains lipid droplets but not biological membranes.

Product Information

Oil Red O Staining Solution	
Ingredient	Oil Red O
CAS	1320-06-5
Conc.	0.5%
Solvent	Isopropanol

Features

1. Strong specificity for lipid droplet labeling.
2. Easy to use.
3. Good staining performance.
4. High cost-effectiveness.

Application

Adipocyte differentiation assay;
Hepatocyte lipid metabolism model;
Immune cell lipid phagocytosis assay;
Atherosclerotic plaque detection

Preparation of Working Solution

Dilute the Oil Red O stock solution with ddH_2O at a volume ratio of 3:2, then filter through a 0.45 μm membrane to prepare the staining working solution. The freshly prepared working solution should appear burgundy in color without any precipitate. It is recommended to prepare the working solution freshly before use to avoid precipitation caused by prolonged storage.

Instructions

1. Staining of Adherent Cells

(1) Remove the cell culture medium, wash once with PBS, and add an appropriate amount of 4% paraformaldehyde solution or 10% formaldehyde solution to fix for 10 min.
(2) Discard the fixative, wash twice with PBS, and incubate with 60% isopropanol for 20-30 s.
(3) Remove the 60% isopropanol, add freshly prepared Oil Red O working solution, and stain for 10-20 min.
(4) Discard the staining solution, rinse with 60% isopropanol for 10–20 s until the background is clear. Wash with ddH2O 2–5 times until no excess stain leaches out.
(5) Add hematoxylin solution to counterstain the nuclei for 1-3 min. Remove the stain and rinse gently with running water (avoid direct water flow onto the cells during operation).
(6) Cover the cells with ddH2O, transfer them to a microscope, and take photographs.

2. Staining of Cell Smears

(1) Prepare fresh bone marrow or blood smears and allow them to air dry naturally at room temperature. Fix with 4% paraformaldehyde for 10-15 minutes, then rinse twice with ddH2O.

(2) Add freshly prepared Oil Red O working solution and stain for 10-20 minutes.

(3) Remove the staining solution, rinse with 60% isopropanol for 10-20 seconds, and then wash with ddH2O 2-5 times.

(4) Add hematoxylin staining solution to counterstain the nuclei for 1-3 minutes. After discarding the staining solution, gently rinse with running water (avoid direct flushing of the cells).

(5) (Optional) Mount the smear with an aqueous mounting medium, then observe and photograph under a microscope.

Storage

Store at room temperature, protected from light. Valid for one year.

Precautions

1. The staining time for Oil Red O is flexible.
2. Oil Red O solution is a saturated solution. When stored at room temperature or 4ºC, a small amount of precipitation or insoluble material adhering to the bottle wall may occur, which does not affect its use.
3. During Oil Red O staining, evaporation of the staining solution should be avoided, as it may cause precipitation and lead to background staining.
4. Frozen sections for Oil Red O staining sould be relatively thick, generally 10–15 μm.
5. Oil Red O staining results cannot be preserved for a long time; samples should be observed promptly.
6. The product is for R&D use only, not for diagnostic procedures, food, drug, household or other uses.
7. Please wear a lab coat and disposable gloves.