

Protocol for Safranin O Staining for Micromass Cell Culture
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Safranin O stains proteoglycans and glycosaminoglycans. Is considered a cationic dye, so also can bind nucleic acids.

- 1) Fix cells with 0.1% glutaraldehyde in PBS (or lab's fixative solution for Alkaline phosphatase staining) for 20min at room temperature.
- 2) rinse 3 times with 1x PBS
- 3) rinse quickly with 1% acetic acid for 10 seconds
- 4) stain in 0.1% Safranin O solution for 5 minutes
- 5) rinse twice with 1x PBS
- 6) keep in ddH₂O or PBS

Staining Results:

Nuclei.....black
Cytoplasm.....gray green
Cartilage, mucin, mast cell granules.....orange to red