**Penn Center for Musculoskeletal Disorders Histology Core**

**Protocol:** Safranin O and fast green staining

**Materials**
- Safranin O solution (0.2% aqueous)
- Fast green solution (0.02% aqueous)
- Acetic acid solution (1% aqueous)
- Mounting medium
- Citrisolve (or xylene)
- Ethanol

**Deparaffinize and Rehydrate (for paraffin sections)**
1. Citrisolve 5 mins
2. Citrisolve 5 mins
3. 100% ethanol 5 mins
4. 100% ethanol 5 mins
5. 95% ethanol 5 mins
6. 70% ethanol 5 mins
7. Deionized water 5 mins

**Staining**
1. Stain in fast green solution for 3 mins
2. Submerge in acetic acid solution for 10 secs
3. Stain in safranin O solution for 10 mins
4. Rinse in tap water for 5 mins

**Dehydration and Mounting**
1. 70% ethanol 2 mins
2. 95% alcohol 2 mins
3. 100% alcohol 2 mins
4. 100% alcohol 2 mins
5. Citrisolve 5 mins
6. Coverslip

**Expected Results:**
- Glycosaminoglycan-rich tissue: red/orange
- Other connective tissue: bluish green

**Suggested Controls:**
- Cartilage or intervertebral disc

**References**