



Visikol[®] TOX[™] Alizarin Staining Technique:

GD20/21 Rat Fetus



Introduction

This protocol is intended for use with fetal rats in developmental and reproductive toxicology (DART) studies. Visikol TOX greatly reduces the time required for staining skeletal systems for DART studies.

Protocol

1. Eviscerate fetuses.
2. Dehydrate in 95% ethanol (or isopropanol) for (at least) 24 hours.
3. Place into Visikol TOX Pre-Treatment Solution (400 mL for 8 rat fetuses) at room temperature for at least 4 hours (max. overnight).
4. After soaking in the Pre-Treatment Solution, briefly (~5 min) rinse the specimens thoroughly in deionized water. Be sure all of the Visikol TOX Pre-Treatment solution has been washed away with deionized water before proceeding with staining.
5. Prepare Alizarin Red S staining solution by dissolving 20 mg Alizarin in 500 mL of 1% KOH (Prepare fresh prior to use, use 400 mL for 8 rat fetuses). Stain fetuses for 24 hours.
6. Remove from stain solution and transfer directly into Visikol TOX Post-stain solution for 4 hours (400 mL for 8 rat fetuses). Animals may be left in this solution indefinitely.
7. Specimens are transferred to Visikol TOX Clearing solution until clear (400 mL for 8 rat fetuses, 2-12 hours, may be left overnight)
8. After clearing, specimens may be stored in Visikol TOX clearing solution indefinitely, or they may be transferred to aqueous glycerol solution containing thymol for storage.

Contact

For any questions on the Visikol TOX process reach out to info@visikol.com.