

SOP-P014

Preparation of 10 mM Hydroethidine (HE)

Objective: To prepare a solution of hydroethidine (reduced ethidium bromide) in dimethyl sulfoxide (DMSO) for staining cells for the detection of superoxide release in functional assays using flow or image cytometry

Procedure:

1. Weigh out a quantity of hydroethidine close to 3.15 mg on a square of glassine paper.
2. Transfer the dye immediately to a 12 x 75 mm foil-wrapped test tube with stopper.
- 3a. Calculate the volume of DMSO required **in mls** by dividing the actual weight in mg by 3.15.
- 3b. Pipet the calculated volume of DMSO into the tube.
4. Stopper the tube and mix well. Check to see that the dye has dissolved.
5. Label the tube with content information, date, and your initials. Store the dye solution refrigerated at 0-5°C.

Created by: Gretchen Lawler

Verified by: _____ **Date:** _____
Print Name Sign Name