Vital Staining of Chromosomes with Hoechst 33258

Materials

1. Hoechst 33258, 10 mg/ml stock in DMSO.

2. Culture medium.

3. 15 ml sterile conical tube.

Procedure

1. In a 15 ml conical tube, add 1 volume Hoechst 33258 stock to 1000 volumes of medium. Need 3 ml for each coverlisp dish. Mix thoroughly.

2. Remove all medium from the culture dish, replace with the dye-containing medium.

3. Incubate for 30 min.

4. Remove dye-containing medium. Rinse dish twice with regular medium.

5. Add regular medium to the dish. Fluorescence can be observed with Zeiss 02 UV filter set and quartz-halogen lamp. Mercury arc lamps yield a much higher intensity but could cause damange to live cells.

Note: the same protocol can be used with ethidium bromide. Replace Hoechst stock solution with 10 mg/ml aqueous stock solution of ethidium bromide.
