Hoechst Staining protocol for cells

Hoechst 33528 stain: Sigma 14530
Make 1 mg/ml solution in ddH2O
Store protected from light at 4 deg C
On day of assay, dilute to 20 ug/mL in buffer TNE (10 mM Tris, 1 mM EDTA, 2M NaCl, pH 7.4, stored at RT)
After assay, remove media from cell plate by overturning over paper towels
Freeze plate at -80deg C
On day of assay, remove plate and add 100 ul ddH2O per well; incubate at RT for 1 hour
Freeze plate for >20 mins at -80 deg C
Remove plate and allow to thaw to room temperature
Add 100 ul of 20ug/mL Hoechst in TNE buffer for final concentration of 10 ug/mL
Measure fluorescence: Excite 350 nm, measure emission at 460 nm.

For questions, contact Harini Sampath, sampath.harini@gmail.com