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Phantom Dyes Viability Staining Protocol

Buffers: 1X PBS (azide-free, protein/serum free) | Stain Buffer - 1X PBS with 2% FBS, 0.09% Sodium Azide

1. **Remove Phantom Dye vial from freezer and thaw at room temperature.**
2. **Spin Phantom Dye vial down to collect contents at bottom before opening.**
3. **Prepare Stain Buffer for labeling procedure.**
4. **Wash cells twice in 1-2 mL 1X PBS. Spin at 300-400 x g for 5 minutes at room temperature. Afterwards, decant supernatant.**
5. **Resuspend to 1-10 x 10^6 cells /mL in 1X PBS.**
6. **Add 1 uL of Phantom Dye solution for each 1 mL of cell suspension and vortex immediately.**
7. **Incubate for 30 minutes at 2-8°C in the dark to prevent photobleaching.**
8. **Wash cells 1-2 times with 1-2 mL Stain Buffer to remove unreacted dye.**
9. **Cells can be subsequently stained, fixed and permeabilized according to user protocol.**

Note: Cells labeled with Phantom Dyes can be cryopreserved for later use or used in intracellular staining protocols without any loss of fluorescence intensity.