Materials List for:

Use of LysoTracker to Detect Programmed Cell Death in Embryos and Differentiating Embryonic Stem Cells

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Materials

Name	Company	Catalog Number	Comments
LysoTracker Red DND-99	Invitrogen	#L-7528	
Hanks BSS	Invitrogen	14025-076	
Paraformaldehyde	EMD	EM-PX0055-3	
Vectashield	VECTOR	H-1200	
DMEM	Cellgro	10-013-CV	
Non-essential amino acids	Cellgro	25-025-CI	
Sodium pyruvate	Cellgro	25-000-CI	
FBS	Hyclone	SH30071.02	
Pen-Strep	Invitrogen	15140-122	
b-Mercapt–thanol, 50 mM	Invitrogen	21985-023	
LabTek-II Chamber slides (8-well)	Nalge Nunc International	154534	
0.1% Gelatin	Millipore	ES-006-B	
Dulbecco's PBS (D-PBS)	Cellgro	21-031-CV	

Solution Recipes

4% Paraformaldehyde

For 100 ml:

1. Mix 4 g paraformaldehyde, 90 ml H_2O, and NaOH (a drop of 2N NaOH). The paraformaldehyde will not go into solution until you have added some NaOH to increase the pH.
2. Stir and heat at 60 °C until all the powder is in solution (~10-20 min). **Do not overheat.**
3. Add ~10 ml 10x PBS to achieve a final volume of 100 ml.
4. Store at -20 °C in convenient (~10 ml or ~40 ml) aliquots.

WARNING: Paraformaldehyde in 'frill' form (compressed small pellets) is less powdery and can therefore be measured outside of a hood. However you should still wear a protective dust mask (N95 at least) during handling.

EB Culture Media

For 500 ml EB Media:

DMEM:	**404.5 ml**
FBS:	75.0 ml
L-Glutamine:	5.0 ml
Penicillin/Streptomycin:	5.0 ml
Non-essential amino acids:	5.0 ml
Sodium pyruvate:	5.0 ml
β-Mercapt–thanol:	500 µl

