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Creating Gelvatol Solution

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ABSTRACT

A gel-like mounting media (with an anti-fade ingredient) for fluorescence slides.

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MATERIALS TEXT

- DABCO (in small fridge below oven; Sigma #D-2522)
- Polyvinyl Alcohol (in chemical bin#11; Sigma #D-8136)
- Glycerol (in chemical bin #7; Sigma #G-9012)
- 0.2M Tris buffer @ pH 8.5 (12ml = 2.4ml 1M Tris + 9.6 ml double distilled water)

- 1.5 ml microcentrifuge tubes
- 90 ml centrifuge tubes (including tubes for balance)

SAFETY WARNINGS

Be careful when handling DABCO, as it is corrosive to skin (must be treated as hazardous dry or sharps waste).

Label all tubes with "Gelvatol" and date prepared (in case a batch turns bad or is incorrectly prepared).

BEFORE STARTING

- Add a small drop of Gelvatol to specimen.
- Carefully place coverslip on the drop(s) avoiding bubbles.
- The mounting media will set overnight at 4 degrees or at room temperature 3-6 hours.

1 Add 2.4 g of Polyvinyl Alcohol (Mol. Wt. 30,000-70,000) to 6ml of glycerol.

2 Stir well to mix.

3 Add 6ml of double distilled water and leave for at least 2 hours at room temperature (rotate overnight)

4 Add 12ml of 0.2M Tris (pH 8.5).

2.4 ml Tris 1M
9.6 ml H_2O

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5 Heat to 50° C for 10 minutes with occasional mixing.

6 After polyvinyl alcohol is dissolved, clarify by centrifugation (5000g = ~6800 rpm) for 15 minutes at 20° C.

7 Use the large tabletop 5810R Eppendorf centrifuge.

8 Collect supernatant liquid and measure amount collected.

9 Add DABCO to 2.5% as antifade medium:

2.5g DABCO / 100ml of liquid

10 pH to 8.6 with pH paper (the paper will disintegrate, so pipette onto paper rather than putting paper into container).

11 Aliquot in 0.5ml microcentrifuge tubes and store at -20° C.

12 Stocks of Gelvatol are supposed to be stable at room temperature for several weeks after thawing.

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