PPD Recipe (Antifade reagent)

- 1. Put 0.020 g p-phenylenediamine (Sigma p6001-50g) in a 15 ml plastic centrifuge tube
- 2. Add 1ml 1M tris-pH9.0 -this doesn't keep forever-we usually remake it every 2-3 months.
- 3. Add 2 ml H₂0.
- 4. Vortex until dissolved
- 5. Add 7ml glycerol (we use Sigma cat #49770-250ml).
- Vortex.
 Wrap in foil and store at -20.
- 8. Warm it before use—(It can last a while, but once it starts getting dark it is going bad.)
- 9. Seal slide with nailpolish (red glitter nail polish seals better than clear).
- 10. Store slides in -20.

Note: Do not use so much mounting solution that the coverslips are floating. Normally, 15 – 20 ul is sufficient for a 22 x 22 mm coverslip. 22 x 50 mm coverslips require about 40 - 50 ul.