

4% formaldehyde in 0.1 M PB pH 7.4

PPE: safety glasses or visor, lab coat, gloves. Carry out all work in the fume hood. Paraformaldehyde is a known carcinogen.

Precautions: Use dedicated glassware and thermometer. Maintain a separate bottle of dH₂O so that formaldehyde-contaminated glassware does not come into close proximity with the water purification system. Dispose of formaldehyde waste and formaldehyde/paraformaldehyde-contaminated solids (eg filter paper) through the Office of Research Safety; do not pour formaldehyde down the sink.

1. Make up 0.2 M Phosphate Buffer (PB):
 - PB solution A: 0.2 M Na₂HPO₄·7H₂O (MW: 268.07; 53.614 g/L)
 - PB solution B: 0.2 M NaH₂PO₄·H₂O (MW: 137.99; 27.598 g/L)
 - Mix A & B at ~4:1. Add B gradually to A until the pH is 7.4 (this usually takes less of B than a 4:1 ratio would suggest). 0.2 M PB can be stored at 4 °C for up to 2 weeks.
2. Make up 8% formaldehyde in dH₂O:
 - In 500 mL flask, heat 200 mL of dH₂O to ~50 °C (do not go above 53 °C)
 - Add 20 g paraformaldehyde
 - Add 1 M NaOH with a glass pipette until solution is clear (~30 drops)
 - Let solution cool and filter with glass funnel and filter paper
 - Make solution up to 250mL with dH₂O in graduated cylinder
3. Combine 250 mL of 8% formaldehyde and 250 mL of 0.2 M PB pH 7.4 to make 500 mL of 4% formaldehyde in 0.1 M PB pH 7.4. Check pH with pH paper. Store at 4 °C, use within a few days of making.