



DEC 11, 2023

OPEN ACCESS



DOI:
dx.doi.org/10.17504/protocols.io.6qpvr3qbbvmk/v1

Protocol Citation: thnasko 2023. Preparation of fresh paraformaldehyde for mouse perfusion and tissue fixation. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.6qpvr3qbbvmk/v1>

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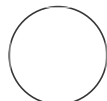
Protocol status: Working
 We use this protocol and it's working

Created: Dec 11, 2023

🌐 Preparation of fresh paraformaldehyde for mouse perfusion and tissue fixation

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ABSTRACT

Hnasko lab - preparation of fresh PFA for ms perfusion and tissue fixation



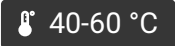


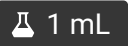



MATERIALS


paraformaldehyde (PFA, EMS, cat# 19210)
 10X PBS (Fisher bioreagents, cat# BP399-1)
 vacuum filtration flask (ThermoScientific, cat# 567-0020).


SAFETY WARNINGS



Caution: Paraformaldehyde is toxic. Please read the MSDS before working with this chemical. Gloves, mask and safety glasses should be worn and solutions made inside a fume hood.

- 1 1. For  1 L of 4% paraformaldehyde, warm up  700 mL of ddH₂O in a glass beaker using microwave until  40-60 °C .
- 2 2. Place on heated stir plate in a chemical fume hood, add a magnetic stir bar and start stirring and insert thermometer into solution.
- 3 3. Weigh  40 g of granular paraformaldehyde (PFA, EMS, cat# 19210).
- 4 4. Add PFA to the heated ddH₂O solution and place on a stir plate in a chemical fume hood. Insert thermometer. Stir and heat to approximately  60 °C (paraformaldehyde flash point = 71°C). Avoid overheating.
- 5 5. The powder will not immediately dissolve into solution. Slowly raise the pH by adding about  1 mL NaOH 10N until the solution clears.
- 6 6. Once the paraformaldehyde is dissolved, add  100 mL 10X PBS (Fisher bioreagents, cat# BP399-1).
- 7 7. Cool solution to  Room temperature before adjusting pH, can place on ice to speed cooling.
- 8 8. Adjust pH to ~  7.4 with small amounts of 12N HCl.

9 9. Adjust the volume of the solution to  1 L with ddH₂O.

10 10. Filter the solution using a vacuum filtration flask (ThermoScientific, cat# 567-0020). Label and store in sealed container at  4 °C .