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

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4% Paraformaldehye in .1M PB preparation

Monique Copeland[1]

[1]HHMI Janelia Research Campus

Monique Copeland
HHMI Janelia Research Campus

ABSTRACT

For perfusion solution.

MATERIALS

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Paraformaldehyde **Catalog #19200**

PROTOCOL MATERIALS

Paraformaldehyde **Catalog #19200** Materials, Step 4

Sodium Hydroxide, 10.0N **Thomas Scientific Catalog #C871W32** Step 3

Corning™ Disposable Vacuum Filter/Storage Systems **Fisher Scientific Catalog #09-761-5** Step 8

Falcon Tube (50 mL) **Fischer Scientific** Step 9

Sodium Phosphate Dibasic Dihydrate (USP/FCC/EP/BP), Fisher Chemical **Fisher Scientific Catalog #S472-500** Step 5.1

Sodium Phosphate Monobasic Anhydrous (Colorless-to-White Crystals), Fisher BioReagent **Fisher Scientific Catalog #BP329-500** Step 5.2

PROTOCOL integer ID:
26993

Funders Acknowledgement:

HHMI

Paraformaldehyde prep

1 Add 400 mL of MilliQ water to a 1000 mL beaker

Equipment

Milli-Q® Integral Water Purification System for Ultrapure Water	NAME
filter	TYPE
Millipore Sigma	BRAND
ZRXQ010WW	SKU
http://www.emdmillipore.com/US/en/product/Milli-Q-Integral-10-Water-Purification-System,MM_NF-ZRXQ010WW	LINK

2 Add stir bar in beaker and set on hot plate at 50 °C

3 Add 5 drops (500ul) of 10N NaOH (from 60ml dropper)

Sodium Hydroxide, 10.0N **Sigma Aldrich Catalog #C871W32**

(this makes the solution basic to help dissolve the paraformaldehyde)

4 Weigh 20 g of Paraformaldehyde and add to MilliQ water.

(Stir about 01:00:00 or until all parafromaldehyde is dissolved and solution is clear)

Paraformaldehyde **Sigma Aldrich Catalog #19200**

5 Make [M] 1 Molarity (M) Phosphate buffer at PH 7.4

5.1 Prepare [M] 1 Molarity (M) Soduim Phosphate Dibasic Dihydrate (Store stock at Room temperature .)

Sodium Phosphate Dibasic Dihydrate (USP/FCC/EP/BP), Fisher Chemical **Sigma Aldrich Catalog #S472-500**

5.2 Prepare [M] 1 Molarity (M) Sodium Phospate Monobasic Anyhydrous (store stock at Room temperature)

Sodium Phosphate Monobasic Anhydrous (Colorless-to-White Crystals), Fisher BioReagent **Sigma Aldrich Catalog #BP329-500**

5.3 To make 100 mL 1M PB at PH 7.4 add 19 mL Monobasic plus 81 mL Dibasic (store as stock at Room temperature)

5.4 Add 50 mL 1M PB PH 7.4 to paraformaldehyde solution.

6 Test PH and adjust solution to final desired pH using HCl or NaOH.

7 Top off to 500ml with MilliQ water.

8 Filter solution through .2um CA member filter.

Corning™ Disposable Vacuum Filter/Storage Systems **Sigma Aldrich Catalog #09-761-5**

9 Lable container with '4% paraformaldehyed in .1M PB' and date. Store at 4 °C for up to 1 week. (Can also be aliqouted into

Falcon Tube (50 mL) **Sigma Aldrich**

and stored at -20 °C for 3 months.)