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Standard Operating Procedures: Mixing Vitrogen

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Mixing Vitrogen (PureCol)

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Required Equipment:

Clean glass jar – autoclaved if necessary
Vitrogen (PureCol) – stored at 4°C, ordered from Inamed, Cat# 5410
10x PBS – on shelf above sink in main lab
0.1M NaOH – either in fridge or under fumehood
disposable gloves
safety glasses
10mL disposable pipettes
pH papers or pH meter
0.1M HCl

Procedure:

Basic procedure for mixing is to have ratio of Purecol:NaOH:10xPBS of 8:1:1, though this actual ratio may vary slightly as ultimate mixture is determined by pH. pH should be 7.2 – 7.4. Please wear gloves and safety glasses at all times.

If you want your mixture to be sterile, all of the steps below should be carried out in the laminar flow hood, and aseptic technique should be followed.

1. Measure out 8mL of PurCol into jar.
2. Add 1mL of NaOH.
3. Add 1mL of 10x PBS
4. Measure pH with pH papers – it should be between 7.2 – 7.4.
5. In most cases, no further steps will be required. However, sometime the pH does not fall into this range. Adjust pH if necessary by adding NaOH if pH is too low, or HCl if pH is too high. **If you add 0.1M HCl, this should be done with a glass pipette.**
6. Store and label any extra in the fridge. Label should include your name, the date, that the vitrogen has been "mixed". Generally it should be used within 3 weeks of mixing.