

## Preparation Paraformaldehyde PAF 4%

To prepare the PFA: watch out it is not easy, **you have to use the chemical hood** because and heat the solution otherwise it will not dissolve and you will kill yourself by inhaling this stuff!!!...

Morevoer, it takes a little while so prepare a lot and aliquot in 5ml so you'll have enough for all year long. I keep my aloquots at -20°C and never use them more than twice, and never keep them at 4oC.

1. Place 450 ml of dH<sub>2</sub>O in a glass beaker. Heat to 60°C using a hot plate with stirring.
2. While stirring, add 20g of paraformaldehyde powder [Fisher #04042] to the heated water. Cover and maintain at 60°C.
3. Add 5 drops of 2N NaOH (1 drop per 100 ml). The solution should clear within a couple of minutes. pH 10 dissolves the paraformaldehyde. There will be some fine particles that will not go away. **Do not heat solution too much above 60°C.** PFA will dissociates and break down at temperature above 60°C.
4. Remove from heat and add 50 ml of 10X PBS. Adjust pH to 7.2; you may have to add some HCl. Final volume will be 500 ml. Place on ice, cover with foil to protect from the light. Sterilize through a 0.22 micron filter.
5. Use immediately, or aliquot may be frozen at -20°C and thawed as needed.