

## phosphate buffer

Information from [cshprotocols.org](http://cshprotocols.org):

Gomori buffers, the most commonly used phosphate buffers, consist of a mixture of monobasic dihydrogen phosphate and dibasic monohydrogen phosphate. By varying the amount of each salt, a range of buffers can be prepared that buffer well between pH 5.8 and pH 8.0 (please see the tables below). Phosphates have a very high buffering capacity and are highly soluble in water. However, they have a number of potential disadvantages:

- \* Phosphates inhibit many enzymatic reactions and procedures that are the foundation of molecular cloning, including cleavage of DNA by many restriction enzymes, ligation of DNA, and bacterial transformation.
- \* Because phosphates precipitate in ethanol, it is not possible to precipitate DNA and RNA from buffers that contain significant quantities of phosphate ions.
- \* Phosphates sequester divalent cations such as  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$ .

0.5L of 1M  $\text{K}_2\text{HPO}_4$  at  $174.18\text{g mol}^{-1} = 87.09\text{g}$

0.5L of 1M  $\text{KH}_2\text{PO}_4$  at  $136.09\text{g mol}^{-1} = 68.045\text{g}$

## preparation of 0.1 M potassium phosphate buffer at 25°C

### Preparation of 0.1 M Potassium Phosphate Buffer at 25°C

pH	VOLUME OF 1 M $\text{K}_2\text{HPO}_4$ (ml)	VOLUME OF 1 M $\text{KH}_2\text{PO}_4$ (ml)
5.8	8.5	91.5
6.0	13.2	86.8
6.2	19.2	80.8
6.4	27.8	72.2
6.6	38.1	61.9
6.8	49.7	50.3
7.0	61.5	38.5
7.2	71.7	28.3
7.4	80.2	19.8
7.6	86.6	13.4
7.8	90.8	9.2
8.0	94.0	6.0

Dilute the combined 1 M stock solutions to 1 liter with distilled  $\text{H}_2\text{O}$ . pH is calculated according to the Henderson-Hasselbalch equation:

$$\text{pH} = \text{pK} + \log \left\{ \frac{[\text{proton acceptor}]}{[\text{proton donor}]} \right\}$$

where  $\text{pK} = 6.86$  at 25°C.

# Potassium Phosphate Buffer Recipe

**Peipei Pang**



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