

SI Unit Prefixes

Milli-	(m)	10^{-3}
Micro-	(μ)	10^{-6}
Nano-	(n)	10^{-9}
Pico-	(p)	10^{-12}
Femto-	(f)	10^{-15}
Atto-	(a)	10^{-18}

Dilution equation:

$$\text{Concentration}_{\text{Initial}} \times \text{Volume}_{\text{Initial}} = \text{Concentration}_{\text{Final}} \times \text{Volume}_{\text{Final}}$$

Example: If you wish to dilute a 1M NaCl stock solution to 10 mM solution with a final volume of 100 mL you would substitute the following numbers into the equation:

$$(C_I : 1M) * (V_I : \underline{\quad} \text{mL}) = (C_F : 10\text{mM} = 0.01M) * (V_F : 100 \text{ mL})$$

$$(V_I : \underline{\quad} \text{mL}) = \frac{0.01M * 100 \text{ mL}}{1M} = 1 \text{ mL}$$

Working with Moles and Molarity

First off, a mole is defined as being equivalent to the sum of the atomic masses in grams of the constituent atoms. Hydrogen has an atomic mass unit of 1 so a mole of hydrogen measures 1 gram.

It is common in molecular biology to use units of moles and molarity. It is important to distinguish the unit of mass (moles) from the unit of concentration (molarity). Moles are simply a unit of mass and are abbreviated as mol. Molarity is designated by an upper case “M” and is defined as moles per liter. For instance, a 1M solution of MgCl (MW = 59.7) is at a concentration of 59.7g per 1 liter of water. If you had $\frac{1}{2}$ of a liter of this solution the concentration of MgCl would still be 1M, but the total amount of MgCl in the $\frac{1}{2}$ liter would be $\frac{1}{2}$ mol, or 29.85g.

When setting up many reactions small molar amounts and small volumes of materials are required. When dealing with these small amounts it is often easier to convert from molarity (moles per liter) to smaller units such as milligrams or micrograms per milliliter or microliter. This is done by reducing the each of the units (moles and liters) by the same number of SI prefixes, as follows:

$$1 \text{ M MgCl} = 1 \text{ mol/L MgCl} = 1 \text{ mmol/mL MgCl} = 1 \text{ }\mu\text{mol}/\mu\text{LMgCl}$$

In many PCRs 5 pmol of a primer is required. Often primers are stored at a concentration of 10mM. To find out what volume of primers are needed in the reaction, the units can be decreased from micromoles per liter to picomoles per microliter as shown below:

$$10 \text{ }\mu\text{M} = 10 \text{ }\mu\text{mol/L} = \text{nmol/mL} = 10 \text{ pmol}/\mu\text{L}$$

Knowing that each microliter contains 10 pmol of primer we know that we need to add $\frac{1}{2}$ microliter of the primer to the reaction to have the desired 5 pmol per reaction.

Standard Reaction Strengths “working concentrations”

Commonly used solutions such as buffers are stored as a high concentration stock, such as a 10X solution. This indicates the material needs to be diluted to a concentration 10 times as dilute as the stock to reach the “working concentration” (1X). This is done by adding 9 volumes of liquid to 1 volume of the 10X stock solution.

Rec. DNA Tech.

Example problems

Convert a 100 mg/ml solution to the following units:

a. $\mu\text{g/ml}$

b. $\text{ug}/\mu\text{l}$

$$\text{a. } \frac{100 \text{ mg}}{1 \text{ ml}} = \frac{1000 \mu\text{g}}{1 \text{ mg}} = \frac{\mathbf{100,000 \text{ ug}}}{\mathbf{1 \text{ ml}}}$$

$$\text{b. } \frac{100,000 \mu\text{g}}{1 \text{ ml}} = \frac{1 \text{ ml}}{1000 \mu\text{l}} = \frac{100,000 \mu\text{g}}{1000 \mu\text{l}} = \mathbf{100 \mu\text{g}/\mu\text{l}}$$

You need a concentration of genomic DNA to be 50 ng/ μl in a total volume of 200 μl , but after you extract the DNA you find you have a concentration of 192.4 ng/ μl how much of this solution should you add?

$$V_i * C_i = V_f * C_f$$
$$? * 192.4 \text{ ng}/\mu\text{l} = 200 \mu\text{l} * 50 \text{ ng}/\mu\text{l}$$

$$\frac{200 \mu\text{l} * 50 \text{ ng}/\mu\text{l}}{192.4 \text{ ng}/\mu\text{l}} = \mathbf{51.97 \mu\text{l}}$$

You add 5 μl of loading dye to 20 μl of a linearized vector sample, and load 5 μl of this on an agarose gel. You also load 10 μl of a 0.05 $\mu\text{g}/\mu\text{l}$ of a Hind III digest of lambda DNA into an adjacent lane. After staining the gel, the fluorescence of the unknown DNA fragment is the same as the fluorescence of the 2.03 kb lambda fragment. What is the concentration of the linearized vector DNA? If you added 4.0 μl of a vector stock to the 20 μl reaction, what was the concentration of the stock?

5 μl of loading dye added to 20 μl sample, 5 μl of this loaded on to gel

- 5 μl of 25 μl total volume = $\frac{1}{5}$ of total sample
- $\frac{1}{5}$ of total sample is equal to the same amount of DNA as the 2.03 kb lambda fragment
- 2.03 kb lambda fragment has how much DNA in it?
 - Total lambda DNA = 10 μl \times 0.05 $\mu\text{g}/\mu\text{l}$ = 0.5 μg DNA
 - The 2.03 kb fragment of lambda is $\frac{2.03}{49}$ of the total mass of all of the lambda fragments
 - $\frac{2.03}{49} = 0.041$, or 4.1% of the total mass
 - total mass of lambda = 0.5 μg , so the 2.03 kb fragment has 0.5 μg \times 0.041 = 0.0205 μg = 20.5 ng of DNA
 - The vector has the same amount of DNA as the lambda fragment, so we know there is 20.5 ng of vector DNA in the gel
 - because we only loaded $\frac{1}{5}$ of our vector in the gel, the total amount of vector is 20.5 ng \times 5 = 102.5 ng
- The concentration of the linearized vector in the 20 μl reaction would be $\frac{102.5 \text{ ng}}{20 \mu\text{l}} = 5.1 \text{ ng}/\mu\text{l}$
- If the DNA was originally added to the reaction in a volume of 4 μl , the stock concentration of the DNA was $\frac{102.5 \text{ ng}}{4 \mu\text{l}} = 25.6 \text{ ng}/\mu\text{l}$

