Biological Chemistry Laboratory
Biology 3515/Chemistry 3515
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Lecture 2:
Units, Concentrations and Dilutions

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Clicker Question #1:

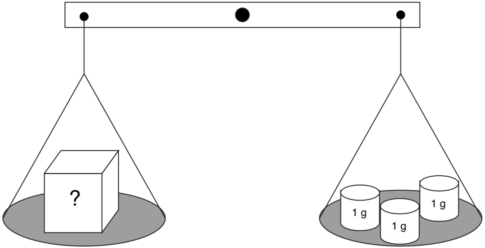
What does the kinetic sculpture in the atrium represent?

1. The mitochondrial ATP synthase.
2. Planets orbiting around a star.
3. Mechanical resonance.
4. Cells dividing.
5. A tree.
6. An upside-down tree.
7. What sculpture?

All answers count for now!

A Bit on Units and Conversion Factors

- All measurements are comparisons.



- Units of measurement are defined by the reference for comparison.
- Suppose that we want to use other units, such as mg or ounces? (what kind of ounces?)

Conversion Factors

- As instructions:
  - To convert from g to mg, multiply by 1,000
  - To convert from g to ounces (U.S.), multiply by 0.035274
- As equations:
  - $1\,\text{g} = 1,000\,\text{mg}$
  - $1\,\text{g} = 0.035274\,\text{ounce}$
- As ratios equal to 1:

$$\frac{1\,\text{g}}{1,000\,\text{mg}} = 1 \qquad \frac{1,000\,\text{mg}}{1\,\text{g}} = 1$$

$$\frac{1\,\text{g}}{0.035274\,\text{ounce}} = 1 \qquad \frac{0.035274\,\text{ounce}}{1\,\text{g}} = 1$$

We can always multiply or divide by 1 without changing anything!

Conversion by Multiplication (or Division?)

- Convert 3 g to ounces

  - Multiply

$$3\,\mathrm{g} \times \frac{0.035274\,\mathsf{ounce}}{1\,\mathrm{g}} = 0.1\,\mathsf{ounce}$$

  - or divide?

$$3\,\mathrm{g} \div \frac{0.035274\,\mathsf{ounce}}{1\,\mathrm{g}} = 85\,\mathrm{g}^2/\mathsf{ounce}$$

- Units can be treated as algebraic entities (and should cancel sensibly in the result!)
- “Dimensional analysis” or “unit factor analysis”

Clicker Question #2

"English" units for mass:

- 1 lb = 16 oz (avoirdupois)
- 1 oz = 28.349523125 g
- 1 stone = 14 Lb

If someone weighs 11 stone, what is that person's mass in kg?

1. ∼ 50 kg
2. ∼ 70 kg
3. ∼ 90 kg
4. ∼ 110 kg

Stones to kg

$$11\,\text{stone} \times 14\,\text{lb/stone} = 154\,\text{lb}$$

$$154\,\text{lb} \times 16\,\text{oz/lb} = 2.46{\times}10^3\,\text{oz}$$

$$2.46{\times}10^3\,\text{oz} \times 28.35\,\text{g/oz} = 6.98{\times}10^4\,\text{g}$$

$$6.98{\times}10^4\,\text{g} \div 1000\,\text{g/kg} \approx 70\,\text{kg}$$

$$1\,\text{kg} \approx 2.2\,\text{lb}$$

Units of Concentration

- Most convenient: amount of solute per volume of solution
  - g/L (= mg/mL): 1 g solute in 1 L final volume of solution
  - molar (M) = mole/L: 1 mole of solute in 1 L final volume of solution

    1 mole = amount of a substance containing the number of atoms or molecules equal to the number of atoms in 12 g of ^{12}C.

    Number of atoms or molecules in 1 mole of a substance is called Avogadro's number, $N_A \approx 6.02 \times 10^{23}$
- Some less convenient (for purposes of calculation) units of concentration
  - molal: 1 mole of solute dissolved in 1 kg solvent
  - 1%(m/v): 1 g solute in 100 mL final volume of solution
  - 1%(v/v): 1 mL pure liquid in 100 mL final volume of solution

A Source of Confusion: Units for “Molecular Weight”

- Molecular weight or molecular mass:
  - The mass of a single molecule
  - Units: atomic mass unit (u or amu) *or* dalton (Da) or kilodalton (kDa)

    1 amu = 1 Da = mass of one atom of $^{12}C \div 12$
  - Units are often not included, because it is a relative mass, M_r.
  - amu is commonly used in mass spectrometry
  - Da and kDa are very commonly used in biochemistry and molecular biology, especially for proteins and other macromolecules.
- Molar mass:
  - Mass of one mole of a compound
  - Units: g/mol (which doesn’t completely make sense)
- Molecular mass of 100 Da $\longrightarrow$ molar mass of 100 g/mol

To Calculate the Amount of Solute in a Solution

- The number of grams in 53 mL of a 5 g/L solution:

$$53\,\text{mL} \times 0.001\,\text{L}/\text{mL} = 0.053\,\text{L}$$
$$0.053\,\text{L} \times 5\,\text{g}/\text{L} = 0.26\,\text{g}$$

- The number of moles in 1.3 L of a 15 mM solution ($1\,\text{mM} = 0.001\,\text{M}$):

$$15\,\text{mM} \times 0.001\,\text{M}/\text{mM} = 0.015\,\text{M} = 0.015\,\text{mol}/\text{L}$$
$$1.3\,\text{L} \times 0.015\,\text{mol}/\text{L} = 0.0195\,\text{mol}$$

- The number of molecules in 1.3 L of a 15 mM solution:

$$1\,\text{mol} = 6.02\times10^{23}\,\text{molecules}$$
$$0.0195\,\text{mol} \times 6.02\times10^{23}\,\text{molecules}/\text{mol} = 1.17\times10^{22}\,\text{molecules}$$

Clicker Question #4

How many moles of water molecules ($M_r = 18$) are in 1 L?

1. ~ 10
2. ~ 30
3. ~ 50
4. ~ 70

How Many Moles of Water in 1 L?

- Liters to grams:

$$1\,\mathsf{L} \times 1,000\,\mathsf{ml/L} = 1,000\,\mathsf{mL}$$

$$1,000\,\mathsf{mL} \times 1\,\mathrm{g}/\mathsf{mL} = 1,000\,\mathrm{g}$$

- Grams to moles:

$$\begin{aligned} 1,000\,\mathrm{g} \div 18\,\mathrm{g}/\mathsf{mol} &= 1,000\,\mathrm{g} \times 1\,\mathsf{mol}/(18\,\mathrm{g}) \\ &= 56\,\mathsf{mol} \end{aligned}$$

Other Units of Concentration Commonly Used in Biochemistry

- Based on molar units:
  - $1\ \text{mM} = 1\times10^{-3}\ \text{M}$
  - $1\ \mu\text{M} = 1\times10^{-6}\ \text{M} = 1\times10^{-3}\ \text{mM}$
  - $1\ \text{nM} = 1\times10^{-9}\ \text{M} = 1\times10^{-3}\ \mu\text{M}$
  - $1\ \text{pM} = 1\times10^{-12}\ \text{M} = 1\times10^{-3}\ \text{nM}$

- Based on mass units:
  - $1\ \text{mg/mL} = 1\ \text{g/L}$
  - $1\ \mu\text{g/mL} = 1\times10^{-3}\ \text{mg/mL} = 1\times10^{-3}\ \text{g/L}$
  - $1\ \mu\text{g}/\mu\text{L} = 1\ \text{mg/mL} = 1\ \text{g/L}$

Calculating Dilutions

- Solutions used in biochemical experiments are often rather complicated, with multiple solutes at different concentrations.
- A solution for an experiment in this course might contain:

  0.1 M Tris-Cl buffer
  20 mM $CaCl_2$
  0.05 μg/mL Enzyme
  125 μM Substrate

- A common practice:
  - Make stock solutions at concentrations higher than the compounds will be used.
  - Dilute and mix stock solutions to make specific solutions for different experiments.

Calculating Dilutions

- Suppose that have a 2 mM stock of substrate solution, and we want to use it to make 0.8 mL of a solution that has a substrate concentration of 125 μM.
  How much of the stock solution do we use?
- A back-to-basics calculation:
  1. Calculate the number of moles in the final solution:

     moles = volume (L) × concentration (moles/L)

     $\text{volume} = 0.8\,\text{mL} \times 10^{-3}\,\text{L/mL} = 8{\times}10^{-4}\,\text{L}$

     $\text{concentration} = 125\,\mu\text{moles/L} \times 10^{-6}\,\text{moles}/\mu\text{moles} = 1.25{\times}10^{-4}\,\text{moles/L}$

     $\text{moles} = 8{\times}10^{-4}\,\text{L} \times 1.25{\times}10^{-4}\,\text{moles/L} = 10^{-7}\,\text{moles} = 0.1\,\mu\text{moles}$
  2. Calculate the volume of stock solution that contains $1{\times}10^{-7}$ moles

     volume (L) = moles ÷ concentration (moles/L)

     $= 10^{-7}\,\text{moles} \div 2{\times}10^{-3}\,\text{moles/L} = 5{\times}10^{-5}\,\text{L}$

     $= 5{\times}10^{-5}\,\text{L} \times 10^{6}\,\mu\text{L/L} = 50\,\mu\text{L}$

Another Way to Calculate Dilutions

- An equation:

$$C_1 V_1 = C_2 V_2$$

C_1 = concentration of stock solution

V_1 = volume of the stock solution to be used

C_2 = concentration of the dilute solution

V_2 = volume of the dilute solution.

Why this equation works: The number of moles is the same in the volume of stock solution used and in the final dilute solution.

- The equation rearranged:

$$V_1 = \frac{C_2 V_2}{C_1}$$

Another Way to Calculate Dilutions

- The same example: What volume of a 2 mM stock solution of substrate do we use to make 0.8 mL of a solution that has a substrate concentration of 125 μM?

$$V_1 = \frac{C_2 V_2}{C_1}$$

$$C_1 = 2\,\text{mM} = 2,000\,\mu\text{M}$$

$$C_2 = 125\,\mu\text{M}$$

$$V_2 = 0.8\,\text{mL} = 800\,\mu\text{L}$$

$$V_1 = \frac{C_2 V_2}{C_1} = \frac{125\,\mu\text{M} \times 800\,\mu\text{L}}{2,000\,\mu\text{M}}$$

$$= 50\,\mu\text{L}$$

- This method just skips the step of calculating the number of moles.
- Keep track of the units and make sure that the result makes sense!