Calculating final, or working, concentrations for journal-style scientific papers.

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When you read a Material and Methods section, you will notice that authors use two types of notations when referring to the concentration of reagents they are using: a *final concentration* in moles or in weight/volume (i.e. grams/ml), or, in percent (%). When reporting the formulations of complex solutions, authors may use both methods to report final concentrations depending on the particular reagent used and how its concentration is most usually reported.

Furthermore, the concentrations you read are the final, or working, concentrations of reagents used in the experiments, NOT the starting, or stock, concentrations stored in the laboratory. You do not need to report the starting, or stock, concentrations because they do not matter as long as the final concentration is given. You may trust that another scientist wishing to repeat your method will be able to do so from the final concentrations you report.

<u>Example 1</u>: You set up an experiment that requires a final concentration of 0.1 M Tris in your tubes, in a total volume of 2 ml. You have a 1 M stock solution in your laboratory. How much of the stock solution will you need in each tube?

There are several ways to solve this problem, but they all use basic algebra. One method is this:

 $C_1V_1 = C_2V_2$

 C_1 = concentration of the stock solution = 1 M

V₁ = the volume of the stock solution you need; this is your unknown that you solve for

 $C_2 = 0.1 M$ Tris, the final concentration

 $V_2 = 2$ ml, the volume of the reaction

 $V_1 = (0.1 \text{ M})*(2 \text{ ml}) / 1 \text{ M}$

 $V_1 = 0.2 \text{ ml}$

Each reaction tube will contain 0.2 ml of 1 M Tris + 1.8 ml of other soluble reagents = 2 ml total. The final concentration of the Tris in this volume will be 0.1 M.

Now, if your partner has a stock solution of 0.5 M Tris instead of 1 M Tris and makes the tubes, he would still want the final concentration of Tris to be 0.1 M Tris, but would use 0.4 ml of 0.5 M Tris + 1.6 ml of other soluble reactions in each tube. It will not matter whether you used 1 M or 0.5 M Tris to accomplish the correct final concentration; the final concentration is the only concentration reported.

The same approach applies if you are using a reagent with concentration units of weight/volume, such as mg/ml.

<u>Example 2</u>: You are given a solution of amylose-azure at 2 mg/ml. You want your final concentration to be 0.5 mg/ml in a 5 ml reaction. How much volume of the stock amylose-azure solution will you use for that 5 ml reaction?

 $C_1 = 2 \text{ mg/ml stock solution}$

 V_1 = volume of stock solution you need; this is your unknown you solve for

 $C_2 = 0.5 \text{ mg/ml}$

 $V_2 = 5 \text{ ml}$

 $V_1 = (0.5 \text{ mg/ml})*(5 \text{ ml}) / 2 \text{ mg/ml}$

 $V_1 = 1.25 \text{ ml}$

Each 5 ml reaction will contain 1.25 ml of 2 mg/ml amylose-azure solution + 3.75 ml of other liquids. If you started with a stock solution of 5 mg/ml amylose-azure, the volume of stock solution you would use would change, while the final concentration and final volume would not. The final concentration is the only concentration reported.

What if I am given a stock concentration and the volumes; how can I figure out the final concentration?

Example 3: You have a 2 mg/ml stock solution of BSA, and you use 0.2 ml of that BSA in a total volume of 1 ml.

Again, a simple algebraic approach works here. Identify what you know and what you need.

You know the stock concentration: $2 \text{ mg/ml} = C_1$

You know the volume of stock solution that you use: $0.2 \text{ ml} = V_1$

You know the final volume of the sample: $1 \text{ ml} = V_2$

You do not know the final concentration of the sample: C_2 = unknown

 $(0.2 \text{ ml})*(2 \text{ mg/ml}) = (1 \text{ ml})*C_2$

 $C_2 = (0.2 \text{ ml})*(2 \text{ mg/ml}) / 1 \text{ ml}$

 $C_2 = 0.4 \text{ mg/ml}$

This is the final concentration of BSA, 0.4 mg/ml, that you would use in your report.

What if I am given a reagent without concentration units? How do I report how much I used?

Here you need to report the proportion of the reagent you used as part of the total reaction. The best way to do this is as a final %, which is not a concentration unit but does give the reader an idea of how much you used.

<u>Example 4</u>: You have 1 cup of orange juice concentrate. You add 4 cups of water, for a total volume of 5 cups. What percent of the mixture is orange juice?

1 cup orange juice concentrate/ 5 cups of solution with orange juice = 0.2

0.2 * 100 = 20% orange juice in the final solution

NOTE: Notice there are no units here; they cancel out in the equation.

<u>Example 5</u>: You use 1 ml of blood in a final volume of 10 ml of heparin solution. What is the concentration of blood in the final solution?

1 ml blood / 10 ml final volume = 0.1

0.1 * 100 = 10% of the solution is blood

In summary, when you are reporting the concentrations of reagents you will:

- Report the final concentrations used, as you calculate them. You do *not* report the starting stock concentrations.
- If your stock reagent is given in a concentration unit such as M (moles/liter) or weight/volume (i.e. grams/liter), the final concentration is reported in similar units.
- If your stock reagent has no concentration given, you will report the final amount used as a proportion, most commonly reported as a percent (%).
- You WILL NOT use a concentration unit AND % together to report the final amount used of a single reagent. For example, you *will NOT* report using 10% of a 5 mg/ml solution.