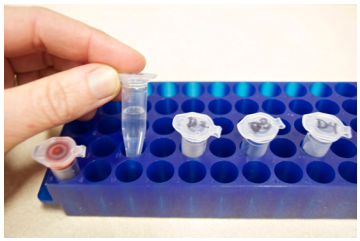
ASSET: How to Make a Serial Dilution

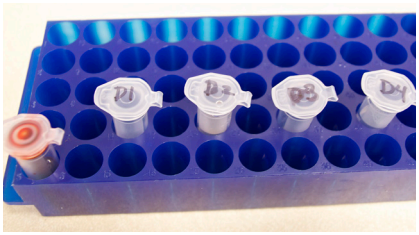
Serial dilutions are a step by step dilution of a solution. The following example is for a tenfold dilution where the dilution factor is 1:10 at each dilution.

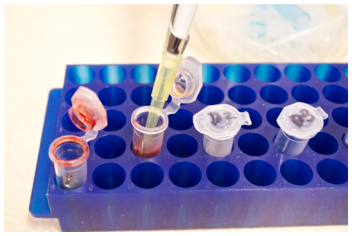
1. First, before beginning your serial dilution, make sure you have all your lab materials organized at your work space. Most dilutions will require a microfuge rack, microfuge tubes, *diluent (the diluting agent)*, micropipette, micropipette tips and a tip disposal container.

2. Next, label the dilution tubes that you will be making: D1, D2, D3, D4, etc...up to the maximum number of dilutions required. In this example we'll be making four dilutions.



3. Fill the D1, D2, D3, and D4 microfuge tubes with 180 µl of the diluent you will be using (distilled water, glucose solution etc...). For this step you will require a micropipette to assure that you have the correct volumes of diluent in each microfuge tube. Notice that all the microfuge tubes are closed except for the one(s) you are working with. This is important to minimize contamination.

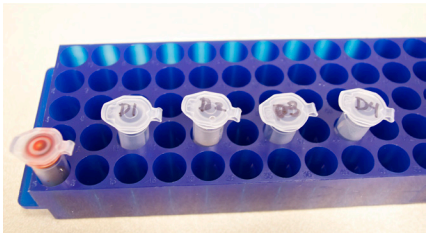




4. Open the original sample you are diluting and pipette 20 µl into your D1 tube. This makes the total volume in the D1 tube 200 µl, of which 20 µl (1/10) is the original sample. This is a tenfold dilution, or a 1:10 dilution.

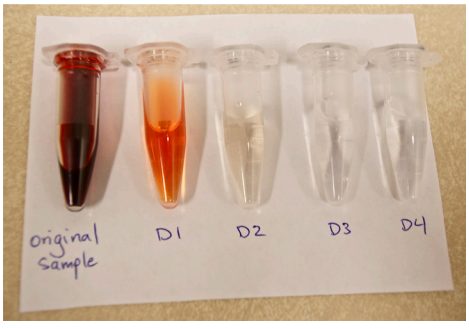
5. In most serial dilutions, you do not need to change the pipette tip between dilutions. However, you do want to ensure that you mix your solutions. With the tip submerged in your D1 tube, **GENTLY** press the plunger up and down a few times to thoroughly mix the solution. Each time you compete a dilution follow this procedure to mix the solutions.

6. After you complete D1, compare the original sample to your D1 tube. In this example, the original sample is a dye, so you can see how the dilution of 1:10 (10^{-1}) looks. However, your sample may or may not have color or cloudiness, in which case you won't be able to see the diluted contents.



7. Pipette 20 µl of the D1 solution into your D2 tube. Gently press the plunger on the pipette up and down to mix. Now you have a dilution of 1:100 (10^{-2}) of the original sample.

8. Pipette 20 µl of the D2 solution into your D3 tube and mix. Now you have a dilution of 1:1,000 (10^{-3}). Complete your last dilution by taking 20 µl from the D3 tube and transfering it into the D4 tube and mix. Your final solution is 1:10,000(10^{-4}) of your original concentrated sample.



9. If your samples are colored or cloudy and you compare them side by side, you will notice how much less concentrated your D4 tube looks compared to your original sample.

10. You now have your serial dilution complete and can proceed to the next step in your lab protocol.

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