Lab Skills: Mixing and Centrifuging Solutions:

B3 Summer Science Camp at Olympic High School 2016

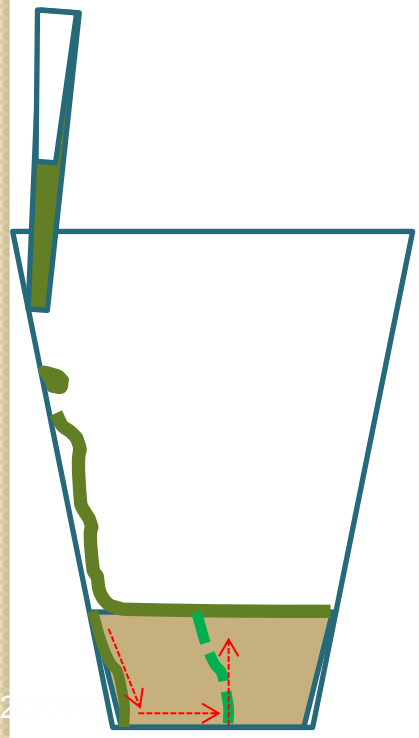
Introduction

- When you make a solution from several components, each is diluted by the others, plus any solvent you add (like water) - they may not mix very well
  - Some are denser than others
  - Some are ‘stickier’ than others
- Why does this matter?
  - If you pull some solution from part of an unmixed solution
    - Your reaction conditions will each be different
    - Some reactions may not work at all

Introduction

- Mixing can be done in a number of ways
  - Cap and invert (shake – slower mixing))
  - Pipette up and down a number of times, using a setting that is close to the total volume
  - Cap and Vortex (fast mixing)
- Once you are done mixing some of the solution may be splashed around on the inside of the cap or sides of the tube – to collect it all in one place
  - Centrifuge at low speed to pull it to the bottom of the tube
  - ** Don’t forget to balance tubes at 180° from each other

Dense and Sticky components - problems



If I run my salt-in-water solution down the side from the lip of the cup it will slide down until it gets to the layer with the closest similar density and across its surface - little mixing occurs

If I put the solution in from below, it will make a column and rise up to the top – little mixing.

It the solution is very sticky and I run it down the side, droplets may stick to the side instead of flowing down with the rest – little mixing.

When you freeze a solution sometimes one part freezes first – crystals may float or sink.

Mixing over time – or diffusion

Over time diffusion will occur-
→ very slow mixing the edges are starting to blur on the right)

Removing solution from a poorly mixed stock.

If the pipette tip is placed just below the surface, you will get the yellow part of the solution.

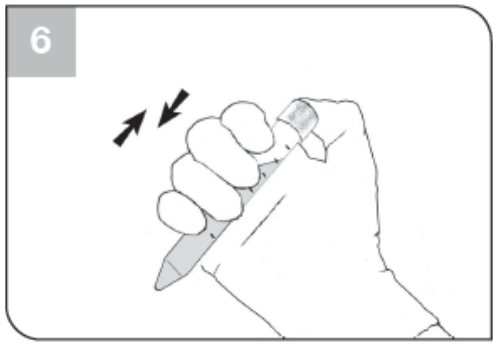
If the pipette tip is placed towards the bottom of the tube you will get the blue part of the solution.

If your reaction needs the right proportions of all 4 components, there is nowhere you can pipette out of this tube to get what you need.

Most components are clear and you can't actually see whether mixing has happened.

Mixing Modes

A.

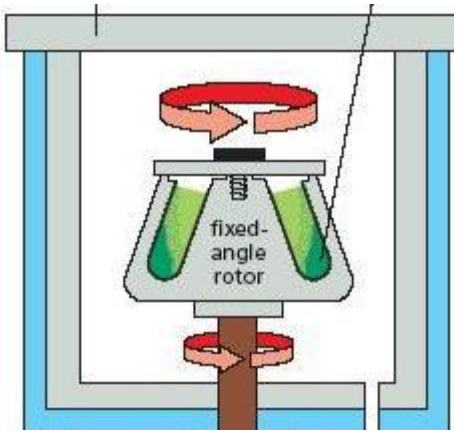


B.

C.

Protein will ‘whip’

Centrifugation to collect solution



If the tubes across from each other not balanced (by mass) the rotor will start to rock, and can eventually break.

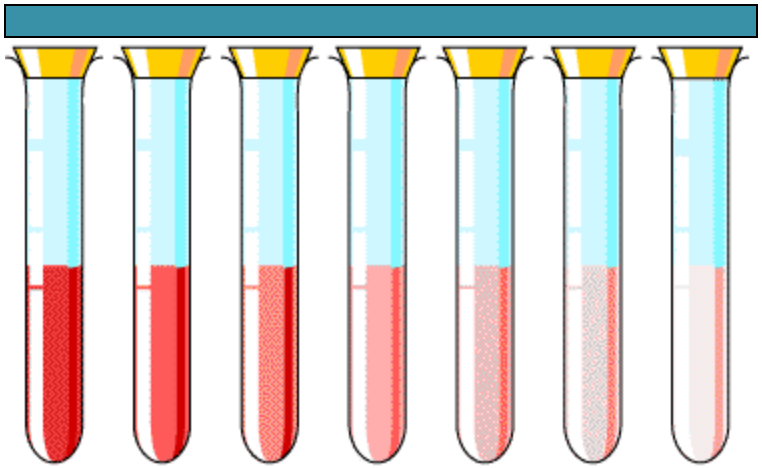
Denser material will collect at the bottom of the tube.

Making Stock solutions

- A stock solution is made up of one or more dissolved chemicals that are at a higher concentration than you need for your processes.
- There are different ways to express

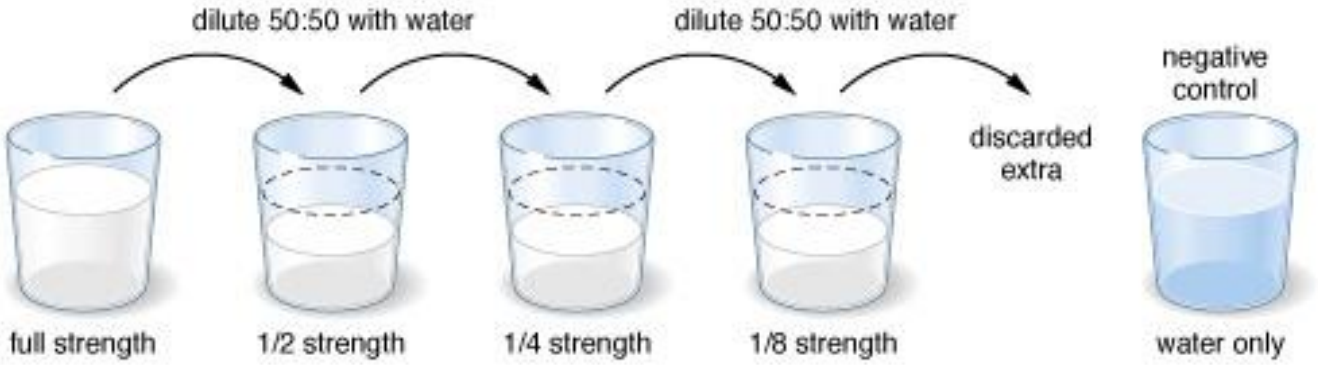
Dilution using Stock Solutions

- Solutions are uniform mixtures
- Stock solutions –
  - Usually a concentrated solution
  - Easier/less expensive to store and ship
- Dilute to the needed amount



Serial Dilution

Hypothesis: If the original bleach solution is diluted repeatedly with water, the bleaching effect will lessen as concentration decreases.



Final dilution factor (DF) = DF1 * DF2 * DF3

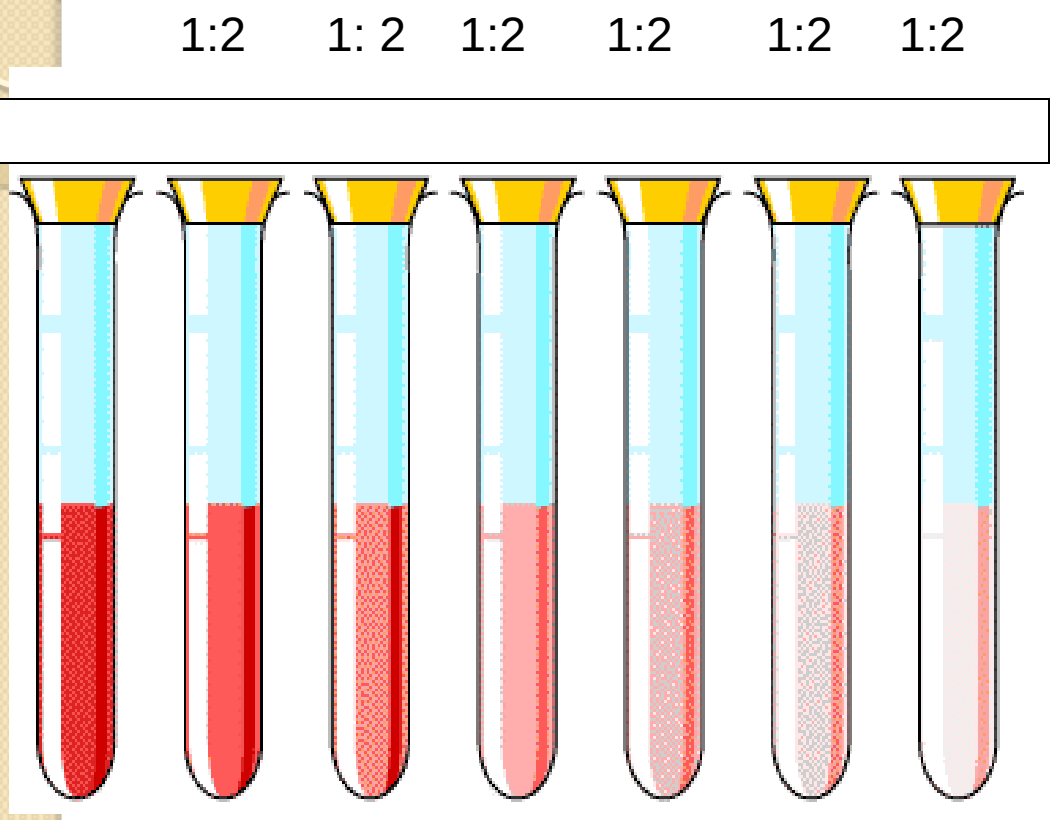
Dilution Factor

- Original Solution to Diluted Solution

  1: 2 = 1 to 2 dilution

- Combine 1 volume of original solution + 1 volumes of the solvent
- 1 + 1 = 2 dilution factor

Serial Dilution Example



Final Dilution

Final dilution factor: 2 x 2 x 2 x 2 x 2 x 2 = ?

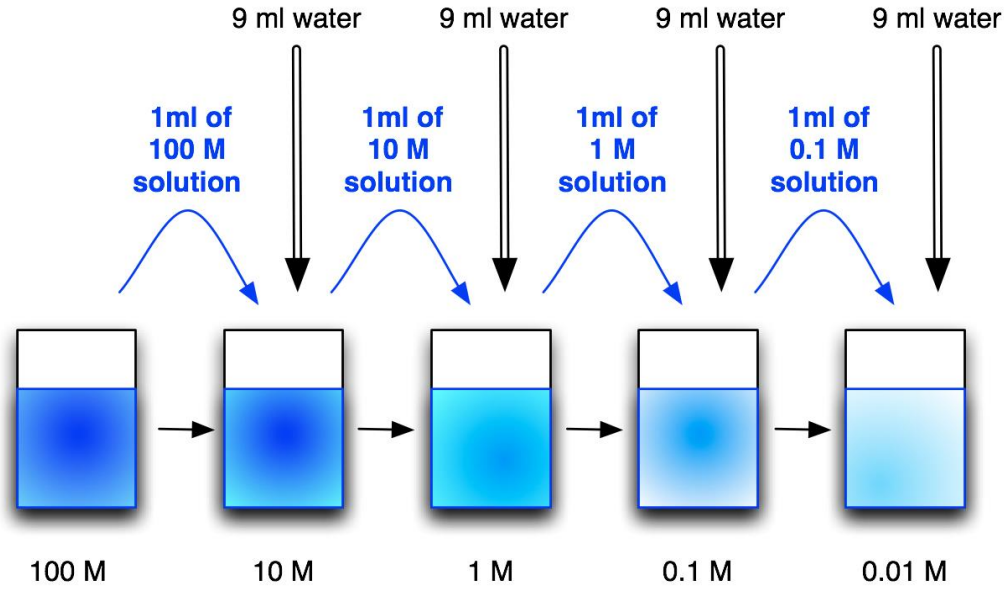
Dilution Factor

- Original Solution to Diluted Solution

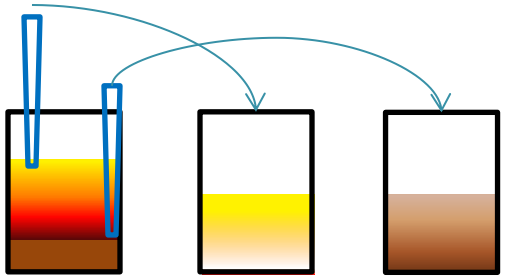
  1: 10 = 1 to 10 dilution

- Combine 1 volume of original solution + 9 volumes of the solvent
- 1 + 9 = 10 dilution factor

Diluting/dispensing from mixed solutions



If each solution is completely mixed before taking out 1ml to make the next solution in the series then your set will differ by a factor of 10 each time.



If the solution is not mixed then you get different amounts of the components depending on where the tip goes into the starting solution – all of the resulting samples will be different, and in a series you won't know what factor they differ by.