Background: DNA Extraction from fruit

Read this before doing your lab to learn in more detail what each chemical does in the DNA extraction process

Introduction

The goal of this lab is to extract DNA from a tissue such that you can actually see the isolated DNA. There are three basic steps in DNA extraction. First, the cell must be broken open (lysed). Next, the nucleus must be broken open to release the DNA. Last, the DNA must be precipitated out of solution. The reagents necessary for the extraction procedure are **detergent**, **salt and alcohol**.

Cells and their membranes are composed primarily of phospholipids and proteins. Phospholipids, like other lipids, are "oily". If you have ever washed dishes, you know that detergents "cut" oil (grease). That is, detergents **emulsify** oil. If **detergent** comes into contact with a cell, it will emulsify its lipid-based membranes.

How does detergent work? Phospholipids have two parts, a polar end that is hydrophilic and a non-polar end that is hydrophobic. In water, these molecules will spontaneously arrange themselves in a double layer (phospholipid bi-layer) with the polar ends facing out. Likewise, the cell membrane consists of a double layer of phospholipids, with the polar ends facing out and the non-polar tails forming an oily middle layer. Detergent molecules have tails that are attracted to the phospholipid tail, and heads that repel other detergent molecules. They break up and isolate lipid molecules (Figure 1).

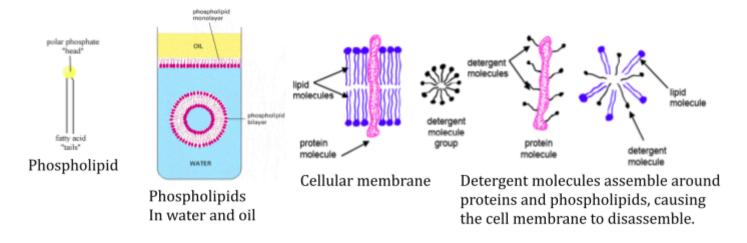


Figure 1. The cell membrane and how detergent disrupts it.

The **detergent** that we will use is sodium dodecylsulfate (SDS). SDS is also called sodium lauryl sulfate, and is an ingredient in dish detergent and shampoo. In the video, they used dish detergent.

As the cell and nuclear membranes emulsify and fall apart, the DNA is released from the cell into the surrounding liquid. DNA is soluble in water, but not in alcohol. When alcohol is added to a solution of water and DNA, the DNA precipitates from the water and becomes visible in a boundary layer between the water and alcohol. The alcohol added should be cold, for two reasons: most things are less soluble when cold, so more likely to precipitate, and second, because DNase enzymes, which chew up DNA, are less active when cold. In the video, they used rubbing alcohol (isopropanol), while we will use lab-grade ethanol. Both alcohols cause DNA to precipitate, and are commonly used in DNA extractions.

Salt (sodium chloride, NaCl) provides DNA with a favorable environment for precipitation in alcohol by contributing positively charged atoms that neutralize the normal negative charge of the DNA. This allows the DNA to clump together.