

## Fermentation - Distillation

### Background

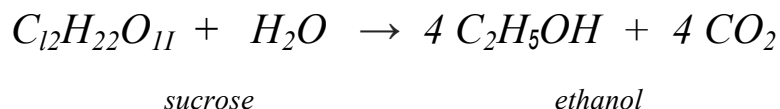
In this experiment you will be working with your lab partner(s) to prepare a sugar solution, allow it to ferment for three weeks, do a simple distillation, collect all of the alcohol, and finally determine the density, percent alcohol, and percent yield.

Fermentation is the chemical process by which organic compounds are converted into new compounds by microorganisms or by enzymes obtained from microorganisms. Enzymes in yeasts and bacteria catalyze the conversion of carbohydrates (sugars) to ethyl alcohol (ethanol) and carbon dioxide.



A 3900-year-old clay tablet, which was found in Iraq between the Tigris and Euphrates rivers, had a Sumerian poem (<http://www.piney.com/BabNinkasi.html>) honoring Ninkasi, the patron goddess of brewing. It contains the oldest surviving beer recipe, describing the fermentation of the carbohydrates found in bread, *bappir*, made from barley, honey, dates and sweet aromatic herbs. The global availability of carbohydrates and native microbes (yeasts) has led to the production of many different types of beers, ales, wines, and fruit based alcoholic beverages in many countries throughout the world. [The bottle on the left was found in Eugene, Oregon, ... But, it dates only to 2016.]

In this experiment you will ferment a carbohydrate, sucrose (table sugar), using bakers yeast. The reaction is:



This process will require about two weeks or more to get maximum alcohol production.

When the reaction is complete, you will perform a simple distillation on the reaction mixture. In a simple distillation, the mixture is placed in a flask and boiled. The gases produced are directed through a condenser. A condenser is a device made of glass with an inner tube surrounded by an outer jacket through which cold water is circulated. When the vapor from your boiled solution enters the inner tube of the condenser, it will condense to liquid, called the distillate, which will be collected. Simple distillation is not a very efficient way to separate all liquids from one another, but it very nicely separates them from dissolved or suspended solids. The distillate that you collect will contain all the alcohol produced in the fermentation and most of the water in which you originally dissolved the sugar. This is similar in content to some wines, ale or beer. Alcohol inhibits the activity of the yeast, which acts as a catalyst, until the alcohol reaches a high enough level that the yeast no longer works (~15% by volume).

You will determine the density of your distillate and calculate its percent alcohol.

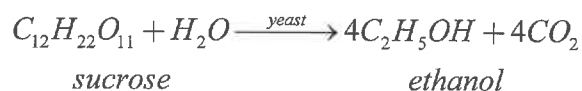
# Fermentation – Distillation

## Background

In this experiment you will prepare a sugar solution, allow it to ferment for two weeks and then do a simple distillation to collect all the alcohol and finally a fractional distillation to concentrate the solution. Following each distillation the density and percent alcohol will be determined. Finally the percent yield will be calculated. Students will work in pairs on this experiment.

Fermentation is the chemical process by which organic compounds are converted into new compounds by microorganisms or by enzymes obtained from microorganisms. Enzymes in yeasts will catalyze the conversion of carbohydrates to ethyl alcohol (ethanol) and carbon dioxide. Alcoholic fermentations have been known and used since prehistoric times. The widespread availability of both carbohydrates and yeasts has led to the production of many different alcoholic beverages.

In this experiment you will cause the fermentation of the carbohydrate sucrose (table sugar) using bakers yeast. The reaction is:



This process will require about two weeks to get maximum alcohol production.

When the reaction is complete, you will perform a simple distillation on the reaction mixture. In a simple distillation the mixture is placed in a flask and boiled. The gases produced are directed through a condenser. A condenser is a device made of glass with an inner tube surrounded by an outer jacket through which cold water is circulated. When the vapor from your boiled solution enters the inner tube of the condenser, it will condense to liquid, called the distillate, which will be collected. A simple distillation is not an efficient way to separate liquids from one another, but it very nicely separates liquids from dissolved or suspended solids. The distillate that you collect will contain all the alcohol produced in the fermentation and most of the water in which you originally dissolved the sugar.

You will determine the density of your distillate and using the table provided find its percent alcohol.

You will use a volumetric pipet to measure an exact volume of distillate for the density determinations. A volumetric pipet is a long glass tube with a bulge in the middle. The stem above the bulge has a line etched around it. The liquid to be used is drawn into the pipet using the suction from a pipet bulb. When the meniscus of the liquid is above the line on the stem, the bulb is replaced by your forefinger that is used to control the flow of liquid from the pipet. When the meniscus of the liquid is exactly on the line on the stem, the pipet is positioned over a receiving vessel and the finger removed. The liquid will flow out of the pipet. Do not blow out the last drop. A pipet will deliver the volume written on it good to three significant figures.

## Equipment

From the stockroom:

Fermentation tube – Preparation of the Solution

Thermometer – Simple Distillation

Kem Kit and tubing – Simple Distillation

20 or 25 mL pipet – Density

Pipet bulb – Density

From the common drawer for Simple Distillation:

ring stand and ring

2 utility clamps

Bunsen burner

From your drawer

500 mL Erlenmeyer flask – Preparation of the Solution

250 mL Erlenmeyer flask – Preparation of the Solution

50 or 100 mL graduated cylinder – Simple Distillation

## Procedure

### Preparation of the Solution

Weigh out about 25 g of sucrose (table sugar) and record the mass to 0.01g. Dissolve it in about 200 mL of deionized water in your 500 mL flask, add a pinch of yeast nutrient and some yeast. The exact amount of yeast is not important; one good sized scoop on the end of your scoopula is about right. Use a fermentation tube provided by the stockroom. Stopper the flask with a one hole stopper. Put about 150 mL of deionized water into your 250 mL flask. Insert the longer piece of plastic tubing through one hole of the two-hole stopper so that when the flask is stoppered the plastic tubing almost touches the bottom of the flask. See Figure 1. Store the apparatus as directed by your instructor to ferment for two weeks.

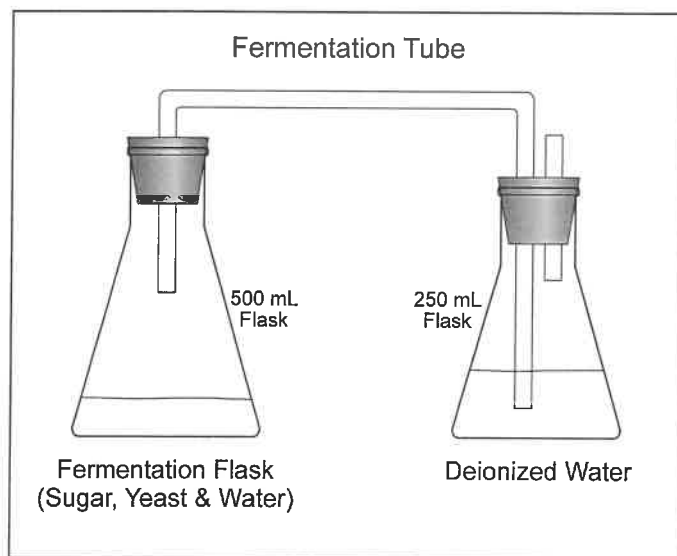
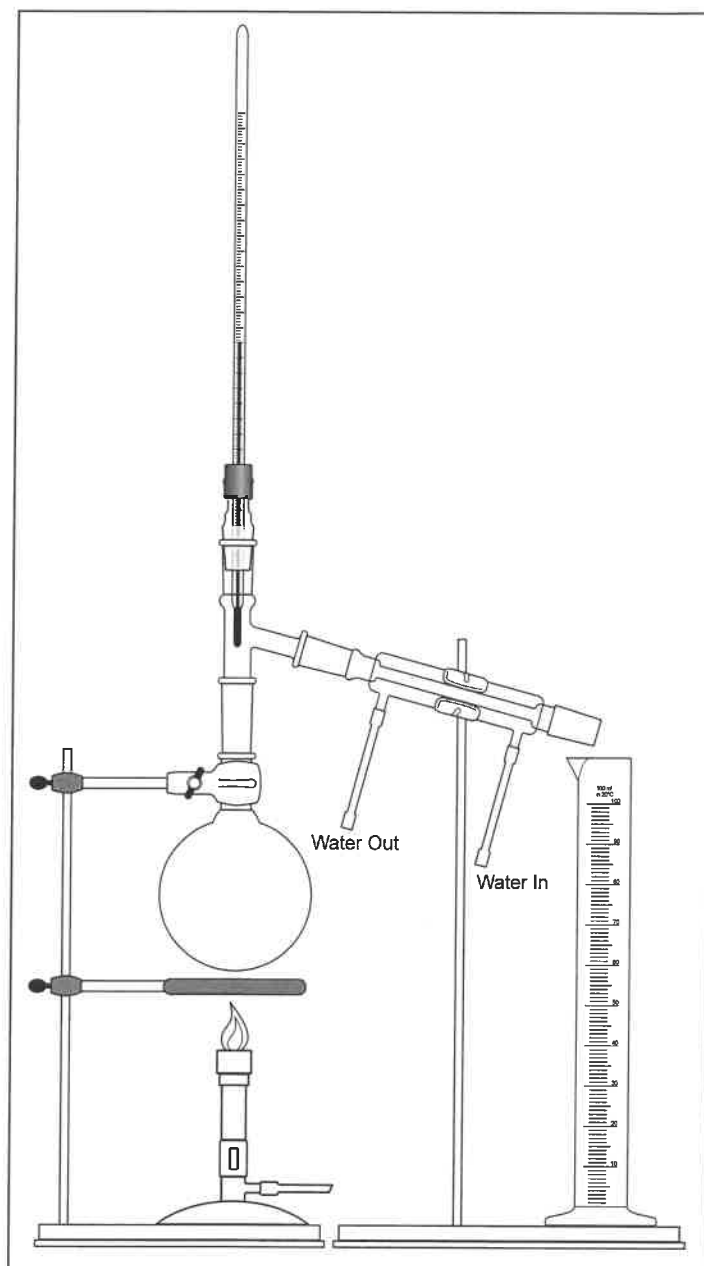


Figure 1–Fermentation Setup

## Simple Distillation

After the solution has fermented for two weeks, a simple distillation is to be performed. Obtain a Kem Kit from the stockroom. Transfer the liquid from the fermentation flask to the 500 mL distilling flask which you will find in the Kem Kit. Add 2–3 boiling chips and spray the solution with anti-foam spray (one squirt is enough). Assemble a distillation apparatus as demonstrated by your instructor. (See Figure 2.) Heat the solution gently to prevent frothing. Record the temperature when the distillate first begins to run through the condenser. If the distillate comes through hot or if steam comes out of the condenser, reduce the heat under the distillation flask. Collect about 50 mL of clear distillate in a graduated cylinder. Record the temperature when the last drop of distillate is collected. Measure and record the volume of distillate collected in a graduated cylinder. Save the distillate in a clean 125 mL flask for use in the next parts of the experiment. The residue in the distilling flask and the water solution may be discarded in the sink.



*Figure 2–Simple Distillation*

### Density and Percent Alcohol of the Distillate

While the distillate is cooling weigh a clean dry vial and cap or small beaker and record the mass. Obtain a 20 or 25 mL pipet (each partner should have a vial or small beaker and each partner must use a different volume) and rinse it thoroughly with deionized water. Draw distillate into the pipet until the bulb is about one-third full. Rinse all parts of the pipet with the distillate and return the it to the distillate in the flask. You don't want to discard any alcohol at this point. Rinse the pipet a second time with distillate and return it to the flask. Pipet 20 or 25 mL of distillate (depending on the volume of your pipet) into the weighed vial and cap the vial or into a small beaker. Weigh the vial or beaker and contents and record the mass. If your density and your partner's don't agree within 0.005 g/mL repeat the procedure. When you have two densities that agree, record your partner's density and average them. Determine the percent alcohol to 0.1 % from the table of densities.

### Determination of Percent Yield

From the density, volume, and percent alcohol of the distillate, calculate the actual yield in grams of ethanol. From the balanced equation for the reaction, given in the background, and the mass of sucrose fermented, calculate the theoretical yield. Finally, calculate the percent that the actual yield is of the theoretical.

### PERCENT ETHANOL FOR VARIOUS DENSITIES

% ethanol by mass	Density (g/mL)	% ethanol by mass	Density (g/mL)	% ethanol by mass	Density (g/mL)
0.0	0.998	35.0	0.945	69.0	0.870
1.0	0.996	36.0	0.943	70.0	0.868
2.0	0.995	37.0	0.941	71.0	0.865
3.0	0.993	38.0	0.939	72.0	0.863
4.0	0.991	39.0	0.937	73.0	0.860
5.0	0.989	40.0	0.935	74.0	0.858
6.0	0.988	41.0	0.933	75.0	0.856
7.0	0.986	42.0	0.931	76.0	0.853
8.0	0.985	43.0	0.929	77.0	0.851
9.0	0.983	44.0	0.927	78.0	0.848
10.0	0.982	45.0	0.925	79.0	0.846
11.0	0.980	46.0	0.923	80.0	0.843
12.0	0.979	47.0	0.920	81.0	0.841
13.0	0.978	48.0	0.918	82.0	0.838
14.0	0.976	49.0	0.916	83.0	0.836
15.0	0.975	50.0	0.914	84.0	0.833
16.0	0.974	51.0	0.912	85.0	0.831
17.0	0.973	52.0	0.909	86.0	0.828
18.0	0.971	53.0	0.907	87.0	0.826
19.0	0.970	54.0	0.905	88.0	0.823
20.0	0.969	55.0	0.903	89.0	0.821
21.0	0.967	56.0	0.900	90.0	0.818
22.0	0.966	57.0	0.898	91.0	0.815
23.0	0.965	58.0	0.896	92.0	0.813
24.0	0.963	59.0	0.893	93.0	0.810
25.0	0.962	60.0	0.891	94.0	0.807
26.0	0.960	61.0	0.889	95.0	0.804
27.0	0.959	62.0	0.887	96.0	0.801
28.0	0.957	63.0	0.884	97.0	0.798
29.0	0.955	64.0	0.882	98.0	0.795
30.0	0.954	65.0	0.879	99.0	0.792
31.0	0.952	66.0	0.877	100.0	0.789
32.0	0.950	67.0	0.875		
33.0	0.949	68.0	0.872		
34.0	0.947				