Yogurt (yoghurt) is a form of bacterially fermented milk produced in most countries where fresh milk is available. Originally a means of preserving the food value of milk (the acidic pH of yogurt inhibits the growth of spoilage organisms), yogurt is now a very popular food product everywhere it is available.

Yogurt is produced commercially in the U.S. by the controlled fermentation of bovine (cow’s) milk by two species of bacteria, *Lactobacillus delbrueckii* subsp. *bulgaricus* (also known as *Lactobacillus bulgaricus*) and *Streptococcus salivarius* subsp. *thermophilus* (also known as *Streptococcus thermophilus*).

The disaccharide sugar in milk (lactose) is fermented to lactic and formic acids and the lowered pH causes the characteristic curd to form as proteins in the milk become denatured.

*Streptococcus thermophilus* brings the pH of the milk down to 5.5. *Lactobacillus bulgaricus* converts lactose to lactic acid. The total acid in yogurt is 59% lactic acid, 28% citric acid, 5.3% acetic acid, 2.4% formic acid, 2.3% succinic acid and small amounts of other acids.

The two bacteria have a mutually growth stimulating effect on one another. Proteolytic enzymes from *L. bulgaricus* break down milk proteins into short peptides and amino acids. These peptides stimulate the growth of *S. thermophilus* which in turn produces formic acid and carbon dioxide which are growth stimulants for *L. bulgaricus*. At the end of the incubation, pH may fall to as low as 4.0.

The low pH coagulates the remaining milk proteins, causing the yogurt to thicken (set). The characteristic component of yogurt aroma is acetaldehyde, a metabolic product of both bacterial species, others are acetone, ethanol, butan-2-one, diacetyl and ethyl acetate. Incubation takes 12 hours at 32°C (90°F) to reach the set point of natural yogurt.

Sugar, color and/or fruit pulp is often added to increase popularity of product. Some yogurts may receive additional heat treatment to kill off any living bacteria before or after packaging for the consumer.

![Diagram of bacterial actions in yogurt culturing](image-url)

**Figure 1. Bacterial actions in yogurt culturing**

**Processing Steps in Commercial Yogurt Production:**

1. **Adjust Milk Composition and Blend Ingredients**
   
   Milk composition may be adjusted to achieve the desired fat and solids content. Often dry milk is added to increase the amount of whey protein to provide a desirable texture. Ingredients such as stabilizers are added at this time.

2. **Pasteurize Milk**
   
   The milk mixture is pasteurized at 85°C (185°F) for 30 minutes or at 95°C (203°F) for 10 minutes. A high heat treatment is used to denature the whey (serum) proteins. This allows the proteins to form a
more stable gel, which prevents separation of the water during storage. The high heat treatment also further reduces the number of spoilage organisms in the milk to provide a better environment for the starter cultures to grow. Yogurt is pasteurized before the starter cultures are added to ensure that the cultures remain active in the yogurt after fermentation to act as probiotics; if the yogurt is pasteurized after fermentation the cultures will be inactivated.

3. Homogenize
   The blend is homogenized to mix all ingredients thoroughly and improve yogurt consistency.

4. Cool Milk
   The milk is cooled to 42°C (108°F) to bring the yogurt to the ideal growth temperature for the starter culture.

5. Inoculate with Starter Cultures
   The starter cultures are mixed into the cooled milk.

6. Hold
   The milk is held at 108°F (42°C) until pH 4.5 is reached. This allows the fermentation to progress to form a soft gel and the characteristic flavor of yogurt. This process can take several hours.

7. Cool
   The yogurt is cooled to 7°C (45°F) to stop the fermentation process.

8. Add Fruit & Flavors
   Fruit and flavors are added at different steps depending on the type of yogurt. For set style yogurt, the fruit is added in the bottom of the cup and then the inoculated yogurt is poured on top and the yogurt is fermented in the cup. For Swiss style yogurt the fruit is blended with the fermented, cooled yogurt prior to packaging.

9. Package
   The yogurt is pumped from the fermentation vat and packaged as desired.

Figure 2. Yogurt Processing
Previously, we learned about how microbes, such as bacteria and molds, contaminate our food supply (See: “Bacterial Contamination of foods”). We have already learned about the process of fermentation, an anaerobic alternative to the process of aerobic cellular respiration and we know that some microbes assist in the production of substances we use as food. Yeasts, for example, can be used for the production of bread and of certain types of fermented beverages, such as beer and wine. Some molds (a type of fungus) help produce cheeses, such as Camembert, blue cheese and Roquefort.

In this exercise, you will observe another example of how microorganisms can benefit instead of harm us. This lab procedure presents another positive way in which we can use microbes - for the production of yogurt, a product of the fermentation of lactose (milk sugar) by bacteria.

In the production of yogurt, two different species of bacteria ferment lactose: *Lactobacillus bulgaricus* and *Streptococcus thermophilus*.

Enzymes in the bacteria convert the disaccharide lactose into an acid called lactic acid. As the acid accumulates in the milk solution and the pH drops, proteins denature and the milk thickens and takes on a “sour” acidic taste. A variation in pH can make the yogurt sweeter and thinner (higher pH) or thicker and more sour (lower, more acidic pH). More milk protein coagulates (denatures) when the pH is low.

**PROCEDURE**

1. Add 250 mL (8 ounces) of milk into a clean beaker. We will use beakers reserved just for this process.

2. Heat the milk (cover the top of the beaker with a piece of foil) in a water bath until it reaches 85°C (185°F), but do NOT boil the solution. Stir frequently.

3. Remove the milk from the water bath and cool it to 46°C (115°F) while stirring.

4. Add 1.25 mL (1/4 teaspoon) of yogurt starter culture to the cooled milk. Stir constantly for three minutes.

5. Place a lid on the container, tape the container closed and be certain it is labeled with your name.

6. Place the container in the incubator. It will be kept there for 8-12 hours, and then we will move it to a refrigerator where it will be stored until our next class meeting.

7. The next time we meet you will get to check your product.

**NOTE:** If you are making yogurt at home, heat a gallon of milk to 96°C (205°F) in a stock pot or a large sauce pan, cool the milk to 46°C (115°F), add 20 grams of yogurt starter powder (or 4 teaspoons of actual homemade unsweetened live-culture yogurt) and incubate. You may incubate the yogurt by wrapping the lidded container in big towels to keep it warm.
Commercial Yogurt Production

Yogurt (also spelled yogourt or yoghurt) is a semi-solid fermented milk product which originated centuries ago in Bulgaria. Its popularity has grown and is now consumed in most parts of the world. Although the consistency, flavor and aroma may vary from one region to another, the basic ingredients and manufacturing are essentially consistent.

Ingredients

Although milk of various animals has been used for yogurt production in various parts of the world, most of the industrialized yogurt production uses bovine (dairy cow’s) milk. Whole milk, partially skimmed milk, skim milk or cream may be used. In order to ensure the development of the yogurt culture the following criteria for the raw milk must be met:

- low bacteria count
- free from antibiotics, sanitizing chemicals, mastitis milk, colostrum, and rancid milk
- no contamination by bacteriophages which are viruses that infect bacteria

Other yogurt ingredients may include some or all of the following:

Other Dairy Products: concentrated skim milk, nonfat dry milk, whey, lactose. These products are often used to increase the nonfat solids content

Sweeteners: glucose or sucrose, high-intensity sweeteners (e.g. aspartame)

Stabilizers: gelatin, carboxymethyl cellulose, locust bean guar, alginites, carrageenans, whey protein concentrate. These products modify the texture of the finished yogurt and prevent the separation of the liquid and solid components (increase shelf-life).

Flavors: including natural and artificial flavoring, color

Fruit Preparations:

Starter Culture

The starter culture for most yogurt production in North America is a symbiotic blend of Streptococcus salivarius subsp. thermophilus (ST) and Lactobacillus delbrueckii subsp. bulgaricus (LB). Although they can grow independently, the rate of acid production is much higher when used together than when either of the two organisms are grown individually. ST grows faster and produces both acid and carbon dioxide. The formate (formic acid) and carbon dioxide produced stimulates LB growth. On the other hand, the proteolytic activity of LB produces stimulatory peptides and amino acids for use by ST. These microorganisms are ultimately responsible for the formation of typical yogurt flavor and texture. The yogurt mixture coagulates during fermentation due to the drop in pH. The streptococci are responsible for the initial pH drop of the yogurt mix to approximately 5.0. The lactobacilli are responsible for a further decrease to pH 4.0. The following fermentation products contribute to flavor:

- lactic acid
- acetaldehyde
- acetic acid
- diacetyl

Manufacturing Method

The milk is clarified and separated into cream and skim milk, then standardized to achieve the desired fat content. The various ingredients are then blended together in a mix tank equipped with an agitation system. The mixture is then pasteurized using heat for 30 min at 85°C or 10 min at 95°C. These heat treatments, which are much more severe than fluid milk pasteurization, are necessary to achieve the following:

- produce a relatively sterile and conducive environment for the starter culture
- denature and coagulate whey proteins to enhance the viscosity and texture of the finished product

The mix is then homogenized using high pressure. Besides thoroughly mixing the stabilizers and other ingredients, homogenization also prevents creaming and wheying off during incubation and storage. Stability, consistency and body are enhanced by homogenization. Once the homogenized mix has cooled to an optimum growth temperature, the yogurt starter culture is added.
A ratio of 1:1, ST to LB, inoculant is added to the jacketed fermentation tank. A temperature of 43 °C is maintained for 4-6 h under quiescent (no agitation) conditions. This temperature is a compromise between the growth optimums for the two microorganisms (ST 39 °C; LB 45 °C). The acidity is carefully monitored until the pH reaches about 4. At this time the jacket is replaced with cool water and agitation begins, both of which stop the fermentation. The coagulated product is cooled to 5-22°C, depending on the product. Fruit and flavor may be incorporated at this time; then the product is packaged. The product is now cooled and stored at refrigeration temperatures (5°C) to slow down the physical, chemical and microbiological degradation.

**Yogurt Products**

- There are two types of plain yogurt:
  - Stirred style yogurt
  - Set style yogurt

The above description is essentially the manufacturing procedures for stirred style. In set style, the yogurt is packaged immediately after inoculation with the starter and is incubated in the packages.

Other yogurt products include:
- Fruit-on-the-bottom style: fruit mixture is layered at the bottom followed by inoculated yogurt, incubation occurs in the sealed cups
- Soft-serve and Hard Pack frozen yogurt
- Continental, French, and Swiss: stirred style yogurt with fruit preparation
- “Greek” and other strained, set style yogurts.

Modified from information found at: [http://www.foodsci.uoguelph.ca/dairyedu/yogurt.html](http://www.foodsci.uoguelph.ca/dairyedu/yogurt.html)

**Briefly describe how the procedures you followed in this laboratory exercise correspond to the manufacturing processes for the production of stirred style yogurt.**