Fermented Milk Products
ACIDOPHILUS MILK

- Milk fermented by live cultures of the bacterium acidophilus and consumed especially to promote intestinal health.
- This cultured product is usually low in fat and has a longer shelf life than ordinary milk.
- Acidophilus milk is usually not recommended for people who have immune system problems.
- *Lactobacillus acidophilus* used as a starter culture.
- Lactic acid – 1.0% (0.6-0.7 for medicinal use) Lactobacillus counts 2000-3000 million/ml
Manufacture of Acidophilus Milk

1. Receiving milk (skim/whole/defatted)
2. Filtration/Clarification (35-40 °C)
3. Homogenization
4. Sterilization (115 °C/15 min)
5. Cooling (38 – 40 °C)
6. Inoculation (3-5 %)
7. Incubation (38-40 °C/12-16h)
8. Coagulation
9. Break-up of coagulum
10. Cooling (10 °C)
11. Packaging & storage (5 °C)
Therapeutic value of Acidophilus Milk

- Constant use of acidophilus milk helps in combating intestinal disorders.
- It exerts its effects in patients suffering from constipation and colonic ulcerative colitis.
- It was shown that feeding \textit{L. acidophilus} milk to rats for 4 weeks significantly lowered the serum cholesterol level as compared to rats fed on either milk or water.

For a longer storage, 0.65\% acidity and 16\degree C storage temperature are recommended. The quality acidophilus milk should be creamy with a mildly acid taste with no abnormal flavours and odours.
Acidophilus Based Products

Sour Acidophilus Milk

- Acidophilus milk is a sour product that has been allowed to ferment under conditions that favour the growth and development of a large number of *Lactobacillus acidophilus* organisms.
- This acidophilus milk is considered as a probiotic since it aids in the well-being of the consumer.
- Acidophilus milk differs from Indian dahi or curd in body, texture, consistency, flavour, chemical composition and in antibacterial activities.
Sweet Acidophilus Milk

- As natural fermented acidophilus milk was sour and having medicinal type of flavour, it was thought appropriate to sell as non-fermented milk. This gave birth to sweet acidophilus milk.

- It is probiotic dairy product based on unfermented milk. It is produced by adding concentrated probiotic bacteria to intensively heat treated and chilled milk.

- Heat treatment is necessary to achieve sufficient microbiological stability during storage of the final product.

- In some cases, it is also prepared by adding concentrated cells of *L. acidophilus* in chilled pasteurized milk.

Acidophilin

- Originated in USSR, obtained from cow‘s milk which is set or stirred, flavor may vary from mild to acid depending on the quality of starter culture.

- The principle of processing is mixed acid and alcoholic fermentation involving *L. acidophilus, L. lactis* subsp. *lactis* or *L. delbruckii* subsp *bulgaricus* and Kefir culture) at the rate of 6-9 %.
Acidophilus Yoghurt:

• Many probiotic products have been developed taking yoghurt as a base. In some products, acidophilus or other probiotic bacteria are added as a supplement or in other cases, one of the yoghurt culture is replaced by *L. acidophilus*.

• The acidophilus yoghurt produced from cow milk is popular in Germany, USA, Scandinavia, Australia, and many other countries.

• It is believed that human intestinal strains of acidophilus culture increase the beneficial value of the yoghurt made with them. The starter culture consists of yoghurt culture (*S. thermophilus* and *L. delbruckii* subsp. *bulgaricus*) and *L. acidophilus*.

• A product known as ACO-Yoghurt is also reported from Switzerland and U.K with improved dietetic and therapeutic values of the yoghurt. The starter cultures consist of yoghurt culture and culture of an intestinal strain of *L. acidophilus*. 
Acidophilus Bifidus Yoghurt:

- This product is popular in Germany, US, Japan and several other countries, obtained from cow milk. It was first manufactured in Germany in order to improve the nutritive and therapeutic value of yoghurt.
- The products involve three groups of bacteria, yoghurt culture (\textit{S. thermophilus} and \textit{L. delbruckii} subsp. \textit{bulgaricus}), strains of \textit{L. acidophilus} and \textit{Bifidobacterium bifidum} or \textit{B. longum}.
- The final product is expected to contain $10^7$ per ml each of \textit{L. acidophilus} and \textit{B. bifidum} and large numbers of yoghurt organisms.

Acidophilus Cream:

- The product has originated from Czechoslovakia, obtained from cream of cow milk. The cultures involved is \textit{L. acidophilus}.
- The process involves cream with 40-45% fat, which is homogenized, pasteurized, added with sugar syrup and then inoculated with the culture of \textit{L. acidophilus}.
Acidophilus Ice Cream

- The acidophilus ice cream finds its origin from USA. It is prepared with a mix added with concentrated *L. acidophilus* culture (10⁹ per ml) and frozen.
- Study of viability of *L. acidophilus* in ice cream has been studied. Recently, Amul launched ‘Prolife’, a probiotic ice cream containing lactobacilli and bifidobacteria.

Acidophilus Yeast Milk

- The manufacture of the product using the culture *L. acidophilus* and lactose fermenting yeast targeted mainly for the therapy of the gastrointestinal disorders and tuberculosis.
- The viability of the acidophilus bacteria is expected to improve when they are grown together with yeasts.
- The product has a final acidity of 0.8-1.0% and contains about 0.5% ethanol in addition to carbon dioxide.
Bifidus Products

Bifidus Milk:

• The product is originated from Germany.
• Bifidobacteria sp. are the predominant intestinal flora and the major components of large intestine of human adults.
• Bifidus milk is produced in small quantities in some of the European countries. The consumption is linked to the dietetic and therapeutic values rather than the sensory properties.
• However, as Bifidobacteria ferment milk producing more of acetic acid than lactic acid, it has a vinegar like flavour and people do not enjoy it as a food. It remained a product to be used for therapeutic purposes.
Bifidus Baby Foods

• Several formulations containing Bifidobacteria are in the market intended for use as baby foods or infant food formulae. The product aims to increase the population of bifidobacteria in intestinal tract.

• Some of the commercial baby foods are The dried formulae product called Lactana-B, containing lactulose and viable *B. bifidum*, produced from modified milk. The liquid formulae product called Bifiline, containing viable Bifidobacteria, first developed by the Russians in 1982. It is made by using milk formulae called Malutka (Acidophilus baby food) and selected strain of bifidobacteria.
The dried formulae product called Femilect, containing viable Bifidobacteria, developed in Czechoslovakia in 1984, made by fermenting heat treated cream (12% fat) with mixed culture consisting of *B. bifidum*, *L. acidophilus* and *P. acidilactici*. The final product when reconstituted contains $10^8-10^9$/ml viable culture bacteria.

**Bifighurt:** This is a commercial product from Germany. The starter culture used in this product is *B. longum* CKL 1969 and *S. thermophilus*. The final product contains 95% L (+) lactic acid and about $10^7$ bifidobacteria per ml.

**BiogardeR:** This product from Germany is known for its mild acidic taste and flavor. The BiogardeR culture contains *L. acidophilus*, *B. longum* and *S. thermophilus*. Biogarde R produces fermented milk as well as icecream.

**Biokys:** Developed in Czechoslovakia, where the product is a cultured sour cream like beverage obtained by fermenting milk with a mixed culture of bifidobacteria, acidophilus bacteria, and *Pediococcus acidilactici*. 
<table>
<thead>
<tr>
<th>Name</th>
<th>Physical Type</th>
<th>Country of Origin</th>
<th>Microflora</th>
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</thead>
<tbody>
<tr>
<td>Fermented acidophilus</td>
<td>Gel</td>
<td>Most Countries</td>
<td><em>Lactobacillus acidophilus</em></td>
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<tr>
<td>Sweet acidophilus</td>
<td>Liquid</td>
<td>USA</td>
<td><em>Lactobacillus acidophilus</em></td>
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<tr>
<td>Nu Trish a/B</td>
<td>Liquid</td>
<td>USA</td>
<td><em>Lb. acidophilus</em> + Bifidobacteria species</td>
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<td>A-38 Fermented milk</td>
<td>Liquid</td>
<td>Denmark</td>
<td><em>Lb. acidophilus</em> + Mesophilic lactic culture</td>
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<td>Acidophilus Yoghurt</td>
<td>Gel</td>
<td>Most Countries</td>
<td><em>Lb. acidophilus</em> + <em>S. thermophilus</em> and <em>Lb. bulgaricus</em></td>
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<tr>
<td>ACO-Yoghurt</td>
<td>Gel</td>
<td>Switzerland</td>
<td><em>Lb. acidophilus</em> + <em>S. thermophilus</em> and <em>Lb. bulgaricus</em></td>
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<td>Cultura</td>
<td>Liquid</td>
<td>Denmark</td>
<td><em>Lb. acidophilus</em> + <em>B. bifidum</em></td>
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<td>AB-Yoghurt</td>
<td>Gel</td>
<td>Denmark</td>
<td><em>Lb. acidophilus</em> + <em>B. bifidum</em> + <em>Yoghurt culture</em></td>
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<td>Biograde</td>
<td>Liquid</td>
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<td><em>Lb. acidophilus</em> + <em>B. bifidum, S. thermophilus</em></td>
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<tr>
<td>Bifighurt</td>
<td>Gel</td>
<td>FRG</td>
<td><em>Lb. acidophilus</em> + <em>S. thermophilus</em> and <em>B. bifidum</em></td>
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<td>Mil-mil E</td>
<td>Gel</td>
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<td><em>Lb. acidophilus</em> + <em>B. bifidum</em> + <em>Yoghurt culture</em></td>
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<td>Miru-Miru</td>
<td>Liquid</td>
<td>Japan</td>
<td><em>Lb. acidophilus</em> + <em>L. Casei</em>, <em>B. breve</em>, <em>L. casei</em></td>
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<td>Yakault</td>
<td>Liquid</td>
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<td><em>L. casei</em></td>
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<td>Smetara</td>
<td>Liquid/Gel</td>
<td>Eastern Europe</td>
<td><em>Lb. acidophilus</em> + <em>S. lactis subsp. diacetylactis</em></td>
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<td>Zdorov E.(+ malt extract)</td>
<td>Liquid</td>
<td>USSR</td>
<td>Kefir culture</td>
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<td>Kefir</td>
<td>Liquid</td>
<td>Most Countries</td>
<td><em>Lb. delbruckii subsp. bulgaricus</em>, <em>S. thermophilus</em>, <em>S. lactis</em>, <em>Leuconostoc sp</em>, <em>S. cerevisiae</em></td>
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<td>Biogarde Ice Cream</td>
<td>Frozen</td>
<td>FRG</td>
<td><em>Lb. acidophilus</em> + <em>B. bifidum</em></td>
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<td>Big M (Cheese Whey)</td>
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<td>FRG</td>
<td><em>Lb. acidophilus</em></td>
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<td>Product</td>
<td>Form</td>
<td>Origin</td>
<td>Species</td>
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<tr>
<td>Soy yo</td>
<td>Gel</td>
<td>USA</td>
<td><em>Lb. acidophilus</em> + <em>Lb. delbruckii subsp. bulgaricus, S. thermophilus</em></td>
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<td>Lupinseed</td>
<td>Gel</td>
<td>Korea</td>
<td><em>Lb. acidophilus</em> + <em>S. thermophilus, Leuconostoc mesenteroides subsp. mesenteroides, Lb. casei, L. lactis</em></td>
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<td>Matsoni</td>
<td>Liquid /Gel</td>
<td>Eastern European</td>
<td><em>Lb. acidophilus</em> + <em>S. thermophilus</em></td>
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<td>Smetana</td>
<td>Liquid /Gel</td>
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<td><em>Lb. acidophilus</em> + <em>S. thermophilus</em></td>
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<td>Tvorog</td>
<td>Liquid /Gel</td>
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<td><em>Lb. acidophilus</em> + <em>S. faecalis, L. fermentum</em></td>
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<td>Suluguni Cheese</td>
<td>Solid</td>
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<td><em>Lb. acidophilus</em> + <em>S. thermophilus</em></td>
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<td>Prostokoasha</td>
<td>Liquid /Gel</td>
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<td><em>Lb. acidophilus</em> + <em>S. thermophilus</em></td>
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<td>Ryazhenka</td>
<td>Liquid /Gel</td>
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<td><em>Lb. acidophilus</em> + <em>S. thermophilus, Lb. delbruckii subsp. lactis</em></td>
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<td>Sourdough</td>
<td>Solid</td>
<td>FRG</td>
<td><em>Lb. acidophilus</em> + <em>Lb. farcininis, yeas species mixture</em></td>
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<td>Recombined milk (NFDM + anhydrous milk fat)</td>
<td>Liquid /Gel</td>
<td>UK</td>
<td><em>Lb. acidophilus</em></td>
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<td>Soy protein concentrate</td>
<td>Liquid /Gel</td>
<td>Korea</td>
<td><em>Lb. acidophilus</em> + Yoghurt culture</td>
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<td>Soy flour plus WPC</td>
<td>Liquid /Gel</td>
<td>Italy</td>
<td><em>Lb. acidophilus</em> + Yoghurt culture, B. bifidum</td>
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<td>Strain of the Organism</td>
<td>Company</td>
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<td>Chr. Hansen</td>
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<td><em>Lb. acidophilus</em> NCFM</td>
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<td><em>Lb. casei</em> ShirotA</td>
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<td><em>Lb. plantarum</em> 299v</td>
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<td><em>Lb. rhamnosus</em> GG</td>
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<td><em>B. longum</em> BB-536</td>
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<td><em>B. longum</em> SBT-2928</td>
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<td><em>B. breve</em></td>
<td>Yakult</td>
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<td><em>B. lactis</em> LaftiTM B94</td>
<td>DSM</td>
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<td><em>B. longum</em> UCC35624</td>
<td>UCC Cork</td>
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<tr>
<td><em>B. lactis</em> DR-10/Howaru</td>
<td>Danisco</td>
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**Therapeutic Acidophilus/Bifidus products used as probiotics in market**
**Viili**

- Viili is a Finnish cultured dairy food noted for its viscous, gelatinous and almost ropey consistency.
- Longer the strands produced, the more treasured the viili; indeed, some ropes of viili have been reported to reach upwards of one foot in length.
- Viili’s characteristic texture is the result of naturally present, beneficial yeasts and lactic-acid producing bacteria.

It is mildly sour, and in many respects faintly sweet by comparison to other cultured dairy foods and yoghurt, making it a good option for small children.
• Viili is a mesophilic fermented product of Finland that may be either plain or flavored with fruit.

• The fat content may vary from 2-12%, depending on classification (such as low fat, full fat).

• Milk is heated to a high temperature (83°C for 20–25 min), tempered to the incubation temperature of 20°C, and inoculated with 4% starter culture consisting of *L. lactis* subsp. *lactis and diacetylactis* culture, *L. mesenteroides* subsp. *cremoris*, and *Geotrichum candidum*. Incubation occurs in consumer cups at 20°C for 24 h (final acidity of 0.9%).

• The fermentation of viili starters is characterized by a rapid increase in the number of LAB from an initial value of $10^6$ to $10^9$ CFU/g after the first 20 h of fermentation.
• The purpose of incubation in consumer cups is to allow fat to rise to the surface during incubation where the *Geotrichum* will grow and contribute to the typical musty aroma.

• These microorganisms are being used to produce exopolysaccharides which gives a ropy, gelatinous consistency and a pleasantly mild taste. Apart from *Geotrichum candidum*, there are *Kluyveromyces marxianus, Saccharomyces unisporus*, and *Pichia fermentans* has been identified in viili starter cultures at levels of 58%, 11%, and 31%, respectively. Viili yeasts produce carbon dioxide and alcohol, thus, contributing to the typical yeast flavor.

• In addition to its ability to provide a thick body, viili has many of the functional activities including antioxidant, immunomodulatory, ACE Inhibitory and antitumor activity that make it useful in the treatment of cancer, inflammation, diabetes, accelerated ageing, hypertension and heart diseases
Yakult
• Yakult is a Japanese probiotic milk-like product made by fermenting a mixture of skimmed milk with a special strain of the bacterium *Lactobacillus casei Shirota*.

• It was created by Minoru Shirota who graduated from the Medical School of Kyoto University in 1930.

• Yakult has also introduced a line of beverages for the Japanese market that contain *Bifidobacterium breve* bacteria.
• After its introduction in Japan and Taiwan, Yakult was first sold in Brazil in 1966, due to the large number of Japanese immigrants in the country, before it was marketed elsewhere.

• Today, Yakult is sold in 32 countries, although its bacteria cultures are provided from a mother strain from Japan regardless of production location.

Yakult Central Institute for Microbiological Research, Tokyo, Japan
Technology of Yakult

1. Mixing of Raw Ingredients: Live *Lactobacillus casei* Shirota strain is cultured in a 'seed tank' in laboratory. Skim milk powder is mixed with sugar, glucose and filtered, sterilized water to make a sweet milky solution.

2. Sterilization: The sweet, milky solution is sterilized at a high temperature for a short time, destroying any bacteria that may be present. The solution is then transferred to a 6,500-litre culture tank via a closed system of pipes and valves.

3. Culture Tank: The temperature of the tank is reduced to 37°C and live *Lactobacillus casei* Shirota strain is added. The solution is allowed to ferment for about one week until the number of *Lactobacillus casei* bacteria reaches an ideal concentration.
4. Mixing and Storage Tank: The concentrate is transferred to a 12,000-litre mixing and storage tank. The tank is chilled to around 2° C. Sterilized flavours, syrup solution, vitamins and calcium are added to the concentrate. Prior to bottling, the concentrate is diluted with filtered, sterile water.

5. Injection Blow Moulding Machine: The plastic bottles are produced on site. The bottles are made from polystyrene.

6. Bottling and Packaging: The bottles are wrapped with individual bottle labels. Then, they are filled with Yakult, capped with a foil lid, sealed and transferred along the conveyor belt to the packaging facility. Single bottles of Yakult are sorted into groups of five and shrink-wrapped in polypropylene film.

7. Refrigeration Room: Finished products are kept refrigerated before delivery to stores.

8. Yakult Quality Assurance: Yakult maintains a comprehensive quality assurance program in order to ensure that product is of the highest quality. For this reason, samples are collected for laboratory analysis throughout the production process to confirm that the quality assurance measures in place have been effective.
Yakult in India

- Yakult Danone India Pvt. Ltd. is a 50:50 joint venture between Yakult Honsha of Japan and Groupe Danone of France.
- Yakult Honsha is a world leader in probiotics while Groupe Danone is a leader in dairy products.
- The two companies have joined hands to launch Yakult, in India. Yakult is a leading probiotic drink globally. It is being manufactured in a state-of-the-art facility at Rai, Sonepat (Haryana).
- Today Yakult is available in Delhi, NCR, Chandigarh, Punjab, Jaipur, Mumbai, Pune and Bangalore.
LEBEN

- Leben is used across the Arab World (Middle East and North Africa), to refer to a food or beverage of fermented milk. Generally, there are two main products known as leben: in the Levant region, yogurt; and in Arabia and North Africa (Maghreb), buttermilk.
- There is a mixed microflora consisting of *S. lactis*, *S. thermophilus*, *L. bulgaricus* and lactose fermenting yeasts.
- Labneh has 7–10% fat
It is a concentrated yoghurt like or kefir like product. The condensation is brought about by hanging the fermented curd in a cloth bag which allows the whey to drain out.

In some countries like Turkey, a goat or sheep skin bag is used whereas in Egypt an earthenware porous vat is used through which moisture is removed.
Milk (in earthenware containers)
\[\downarrow\]
Inoculation with sample from previous batch
\[\downarrow\]
Incubation at ambient temperature until firm coagulum production
\[\downarrow\]
Churning with gradual addition of warm water

Addition of Fenugreek seeds
\[\leftarrow\]
Heating
Butter
\[\rightarrow\]
Butter oil
Leben
\[\downarrow\]
Cooling
\[\downarrow\]
Packing
BULGARIAN SOUR MILK

- Bulgarian sour milk comes under the general category of yogurts which contain live bacteria. It is a wonderful probiotic food.
- It is prepared – using a combination of the two strains: Lactobacillus Bulgaricus and Streptococcus Thermophilus.
Manufacture of Bulgarian Sour Milk

- Originated from Traki’s tradition who came from Asia to Bulgaria bringing the tradition of sour milk preparation.
- The commercial production may involve only *L. bulgaricus* or mix of both cultures.
- Acidity may reach as high as 4%.
KISHK

• Kishk is a typical wheat-milk dried mixture fermented food popular in Egypt and most of the Arab World.

• It consists of small, round or irregular pieces, yellowish brown in colour, which have a rough surface and hard texture. When moistened with water, it becomes white and breaks up after a short time.

• Kishk is rich in nutritive constituents, possible source of many vitamins and growth factors associated with the microbial fermentative processes.

• Kishk is naturally fermented product and aerobic spore formers like *B. subtilis* and *B. megaterium* are high in numbers in initial stage, but inhibited later on due to high acid.

• The most predominant microflora responsible for fermentation are homo and heterofermentative lactobacilli, mainly *L. plantarum, L. casei* and *L. brevis* are predominate bacteria. During the later stage of fermentation, yeasts develops, which produce gas and improve porosity and also enrich product with vitamins.
Manufacture of Kishk

• Preparation of Par-boiled wheat: Wheat grains, are placed in large cooking pan, covered with water and heated slowly to boiling and simmered until soft. The cooked wheat is then washed with cold water, spread on straw mats and left to dry in air. The dry material which is hard in texture is coarsely ground in stone mills, then sieved and seed coats are removed.

• Preparation of Laban Zeer: Laban-zeer is sour coagulated milk. Traditionally, the milk was churned in skin bag and left out fluid is called as sour milk. In hot weather, milk coagulates spontaneously and is then often considered unsuitable for cheese making. Such sour milk is stored in earthenware containers known as zeer. A suitable quantity of salt is added and the contents of the zeer are mixed. The whey oozes through the walls and thus consistency of sour milk becomes considerably thicker. Accumulated laban zeer is especially used in summer, when wheat are plentiful in the farmers store for making kishk. The most dominant microflora of laban zeer compose of *L. casei*, *L. plantarum* and *L. brevis*. 
Preparation of hamma (kishk mix): The coarser powder of the parboiled wheat is placed in large pots and moistened with slightly salted boiling water. Then raw milk or laban zeer diluted with water is added and thoroughly mixed to obtain a homogenous paste, called hamma. After 24h, the hamma, which has meanwhile increased in volume as a result of fermentation, is kneaded by hand. Later on Laban zeer, twice the volume added before is diluted with water or milk with syrupy consistency, is added in portions to the hamma and left for a further 24h. Subsequently, the fermented mix is thoroughly remixed, cut in small balls and placed on straw mats to dry in the open. The dried product is kishk. Some times spices like cumin or pepper are added for enhancing taste. Complete drying of product takes 5 -7 days and during this period also fermentation goes on gradually at reducing rates.