FERMENTED MILK PRODUCTS

17.2.2005

Fermented milk products

Microorganisms play important role in manufacturing dairy products. A variety of milk products are produced with the selected microorganisms. Fermented milk products includes ~~as~~ butter, cheese, yoghurt, curd etc. In the manufacturing of such products use of microorganisms, mainly lactic acid bacteria are particularly important. The branch of microbiology deals with such products ~~are~~ is known as dairy microbiology.

Fermented milks are prepared by allowing developing the normal lactic acid producing microorganisms present. or by inoculating ~~fresh milk~~ (or pasteurised milk) with specific microorganism. The nature of products depends on (i) source of milk (cow, buffalo, goat, sheep etc

(ii) temperature to which milk is heated before inoculation.

(iii) the kinds of microorganisms present in the starter culture.

(iv) the incubation temperature.

- Starter culture

The known culture of microorganism(s) with which the pasteurised milk is inoculated to produce fermented milk products in the industry, is known as **starter culture**.

The market milk contains numerous species of microorganisms in varying numbers. Their activities uncontrolled and ~~their effect~~ therefore ~~in~~ uniformity of product is difficult to achieve. It is often necessary to carry out controlled fermentation with specific identified organisms to get the desired product. Principal organisms employed are of the order Lactobacillales produce lactic acid ~~as~~ as their major or sole fermentation product ~~are~~ and are sometimes called lactic

acid bacteria. They are non-sporing and usually non-motile. They lack cytochromes and obtain energy from substrate level phosphorylation rather than oxidative phosphorylation (ETC). They normally depend for sugar fermentation for energy. Many vitamins, amino a, purines and pyrimidines must be supplied because of their limited biosynthetic capacity. Lactic a bacteria are usually catagorised as facultative anaerobes but some classify them as aerotolerent anaerobes. The principle lactic a bacteria which are used in production of fermented milk products are Lactobacillus sp., Streptococcus sp., Lactococcus sp. etc.

Fermented milks :-

At least 400 different types of milks are produced in this world. These fermentations are carried out by mesophillic, thermophillic and therapeutic lactic acid bacteria, as well as by yeasts and molds.

Mesophillic :- Lactobacillus sp., and Lactococcus lactis are used in this process and incubation done at approx. 20-30°C and then microbial growth is stopped by cooling. Here acid production occurs through microbial activity causes protein denaturation. The organism L. lactis subspecies diacetilactis converts milk citrate to diacetyl, which gives a special buttery flavour of the finished product.

Thermophilic :- Thermophillic fermentations are carried at around 45°C.

Eg- Yogurt production by Streptococcus thermophilus and Lactobacillus bulgaricus (1:1)

Therapeutic:- Fermented milks may have beneficial therapeutic effects. **Acidophilus milk** is produced by **L. acidophilus**, which may modify the ~~non~~ lower microbial flora of the lower intestine, thus improving general health. The exact nature and nature of health benefits are still unclear but involve minimization of lactose intolerance lowering serum cholesterol.

Another interesting group used in milk fermentation are bifidobacteria. The genus Bifidobacterium contains irregular, non-sporing, gram +ve rods. They are non-motile, anaerobic and ferment lactose and other sugars to lactic a and acetic a. Bifidobacteria help to maintain the normal ~~vit~~ intestinal balance and improve lactose tolerance, sometimes present Rota viruses, a cause of diarrhea among children.

Yeast lactic :→

Yeast lactic fermentation include Kefir a product with a ethanol conc. upto 2%. Kefir products ~~to~~ tend to be foamy due to active CO_2 production.

Mold lactic:→ The milk is inoculated with a mixture of the fungus Geotrichium candidum and lactic a bacteria. Incubated at 18-20°C for 24 hrs. Lactic a reaches a conc. of about 0.9%.

When mixed cultures are used generally one of them is used to grow rapidly to make more lactic acid and more anaerobic condition to prevent the growth of other contaminated microorganisms if present as well as to prepare appropriate conditions for the other microorganisms of the starter culture. Even when single species is used it is better to use many varities of same species. To protect the culture from destruction by phages.

Cheese production

It is one of the oldest human foods (developed about 8000 years ago). About 2000 varieties of cheese are produced in the world. Cheese is a product made by separating casein of milk from the liquid or whey. The manufacturer of all cheeses depend upon the activities of selected microorganisms. Often cheeses are classified based on texture or hardness. — soft cheese, hard cheese, very hard cheese etc. Milk from cows, goats and other animals have been used for making cheese. Four steps are generally followed in preparation of cheese. —

(i) Inoculation of milk with starter culture and curdling of milk.
(ii) processing of curd.
(iii) Salting.
(iv) Ripening.

(i) **Inoculation of milk with starter culture and curdling of milk:-**

Milk inoculated with starter culture ferments lactose with acid production. The acids reacts with calcium to form calcium lactate. When the content is reduced to certain low point the casein precipitate with the formation of firm curd. Starter culture consists of Lacto streptococcus lactis and Lacto streptococcus cremoris if milk is held below 38°C. For higher

temp. (50°C) the starter culture consist of Streptococcus thermophillus combined with Lactobacillus lactis, L bulgaricus etc.

[[One of the greatest problem is the destruction of starter culture by phage infection. To solve this problem aseptic techniques and phage resistant pure cultures (may be genetically engineered) is used. The most stable and dependable cultures, called P starter culture contain in the bacteriophages in a pseudolysogenic state. Antisense RNA is now being used in attempt to provide an agent bacteriophage genes.]]

(ii) Processing of curd:—

The watery fluid in which separates out during the formation of curd is called whey. It contains 93% of water, 5% lactose and smaller amounts of vitamins, minerals, fats etc. Draining of whey without pressure results in the production of soft cheese. Hard cheese contain low moisture. This is achieved by heat and pressure.

(iii) Salting:— Salt is applied to practically to all variety of cheese.

(iv) Ripening:— It's a very complex process and is highly variable, depending upon the kind of cheese being made. In the young cheese, all nitrogen is present in the form of insoluble protein but as ripening proceeds, the protein is progressively cleaved into soluble peptides, ultimately to free amino acids. In addition to changes in protein components ripening involves considerably hydrolysis of the fats present in young cheese. During ripening the flavour, aroma, composition and physical properties of cheese undergo changes due to the action of microbial enzymes.

The final hardness of cheese is partially a function of duration of ripening. Soft cheese ripened for 1-5 months. Hard cheese ripened for 3-12 months and very hard cheese ripened for 12-16 months. Hard cheese are ripened largely by lactic a bacteria which grow through out the cheese, die, autolyse and release hydrolytic enzymes. Soft cheese are ripened by enzymes from yeasts and other fungi that grow on surface. Some microorganisms play highly specific role in the ripening process. The blue colour and unique flavour of Roquefort cheese is a consequence of the growth of a blue coloured mold, P. Penicillium roqueforti. In some countries like USA a white mutant of P. roqueforti is used for the people who like the flavour but not the colour. Swiss cheese with characteristic holes is produced by CO_2, a product of the fermentation of lactic acid by Propionebacterium

MILK (Lactose, fat protein, β-carotene)

CURD FORMATION Ca^{2+}

1. Bacterial culture: Produces lactic acid from lactose, lowers pH to 4-6; 6 hrs, 31°C
2. Rennet - digests K-casein; leads to precipitation of α and β casein; 30 min 32°C
3. Lipase (before rennet is added) - Lipid digestion.

↓

COAGULUM (lactose, fat, proteins, β-carotene)

COOKING 37°C Inactivates rennet

↓

SEPARATION → whey.

↓

CURD

↓

SALTING

↓

PROTEASES and/or LIPASES and/or MICROORGANISMS

1. Protein hydrolysis - flavour production.
2. Lipid hydrolysis - Strong flavours due to butyric acid.

↓

PRESSING

↓

RIPENING 2-10°C, 2-12 Months

↓

CHEESE (Texture, flavour, aroma, Colour due to β-Carotene)

Fig: A schematic representation of the various steps in cheese production.

MICROORGANISMS / ENZYMES USED FOR CHEESE PRODUCTION AND THEIR FUNCTIONS

PROCESS	ORGANISM / ENZYME	FUNCTION
CURD formation.	1. Bacterial culture :- *Streptococcus lactis*, *S. cremoris* or, *S. thermophilus* Plus *Lactobacillus lactis*, *L. bulgaricus*	Produces lactic acid from lactose, lowers pH to 4.6 and leads to coagulation of milk
	2. Rennet (calf or *Mucor michei*)	Digests K-casein, which leads to coagulation of α- and β-caseins; results in curd formation. It cleaves the peptide bond between Phe 105 and Met 106 of K-casein.
	3. Lipase (before rennet is added; from *Mucor michei* or *Aspergillus-niger*	lipid digestion (modest) and accumulation of butyric acid; Produces strong flavours of Italian cheese
RIPENING.	1. Proteases (from *Bacillus amyloliquefaciens*) or, 2. *Penicillium roqueforti* *Penicillium camembertii*	Flavour production, e.g., in **cheddar** cheese by Proteolysis. 1. Peptides with terminal acidic amino acid residues — **meaty appetising flavour** 2. Non-terminal hydrophobic amino acid residues; a. Medium-sized peptides – bitter flavour b. Intensity of bitter flavour decreases with decrease in peptide size. c. Bitter flavour disappears with longer peptides.