MILK CHEMISTRY

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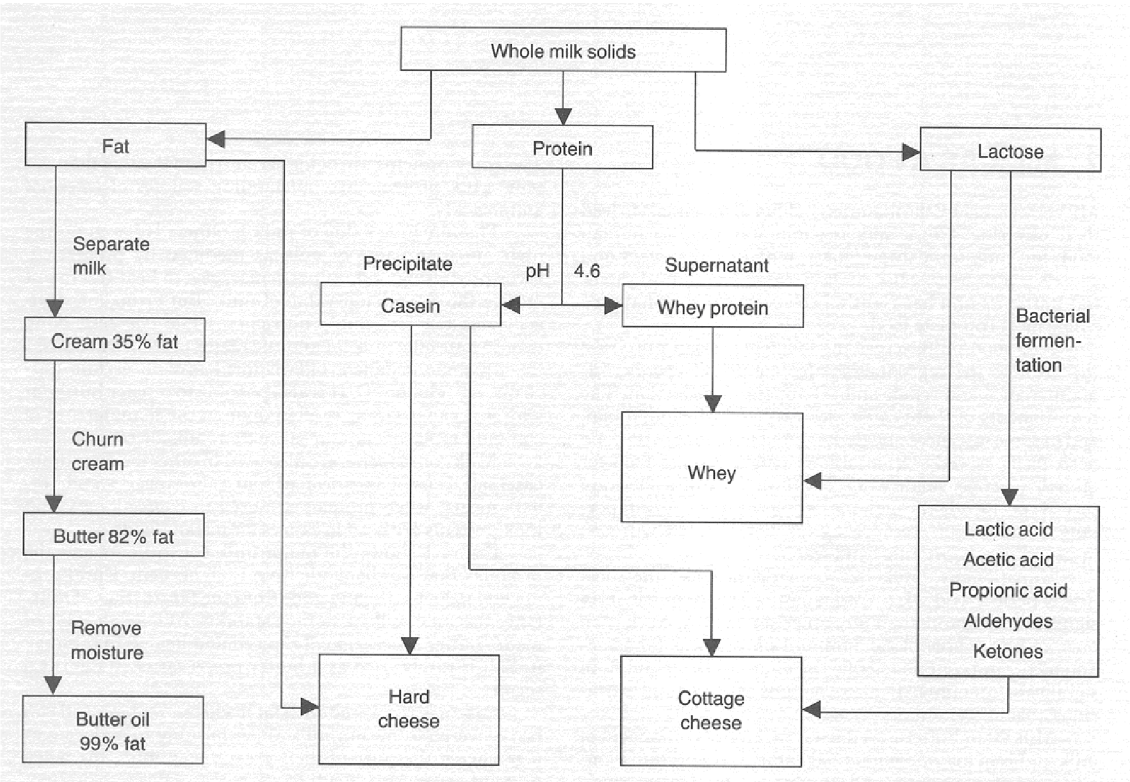
Milk and milk products since prehistoric times have been used by humans to help in growth, reproduction, supply of energy, maintenance, repairs and to satisfy appetite. Milk is secreted by the mammary gland of mammals to feed their offspring. Milk (Cow or Buffalo) is defined as the whole, fresh, clean, lacteal secretion obtained by complete milking of one or more healthy milch animals, excluding that obtained within 15 days before or after calving or such periods as may be necessary to render the milk practically colostrum free and containing the minimum prescribed percentages of milk fat (MF) and milk solids not fat (SNF).

Species	Percentage of Composition				
	Water	Fat	Protein	Lactose	Ash
Ass	90.0	1.3	1.7	6.5	0.5
Buffalo	84.2	6.6	3.9	5.2	0.8
Camel	86.5	3.1	4.0	5.6	0.8
Cow	86.6	4.6	3.4	4.9	0.7
Ewe	79.4	8.6	6.7	4.3	1.0
Goat	86.5	4.5	3.5	4.7	0.8
Human	87.7	3.6	1.8	6.8	0.1
Mare	89.1	1.6	2.7	6.1	0.5

Cow and buffalo milk are commonly used as human food, one can also use milk from ewe, goat, yak, mare, ass and camels. In addition to MF and SNF (contents that determines the price) milk also contains large amounts of essential nutrients and is recognised as nature's single most complete and perfect food for the young, aged and convalescents alike as per their varied individual requirements.

It is rich in carbohydrate primarily **lactose, protein** mainly **casein** and **fat**. Minerals like calcium, phosphorous, sodium; potassium and magnesium are also present in appreciable quantities. Milk provides considerable quantity of Vitamin C, thiamine, riboflavin, niacin, pyridoxine, pantothenic acid, biotin, folic acid, and Vitamin B_{12}. In case of human milk the new born is also nourished by the mammary gland secretion called **colostrum** which is very rich in immunoglobulin, fat, protein and minerals. Human colostrum contains more of vitamin A, vitamin D, iron, calcium, magnesium, chlorides and phosphorus than normal milk from cows and buffalos. In bovine milk and colostrum the principle immunoglobulins are **IgG** and **IgA**.

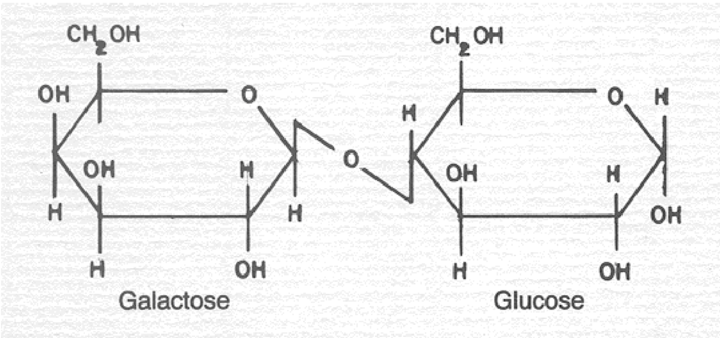


Major milk solid fractions in milk products

MILK CARBOHYDRATES

Lactose or milk sugar unique to milk is the principal carbohydrate disaccharide, composed of two molecules of monosacharides; glucose, and galactose. In addition, milk also contains traces of glucose, galactose and oligosaccharides.

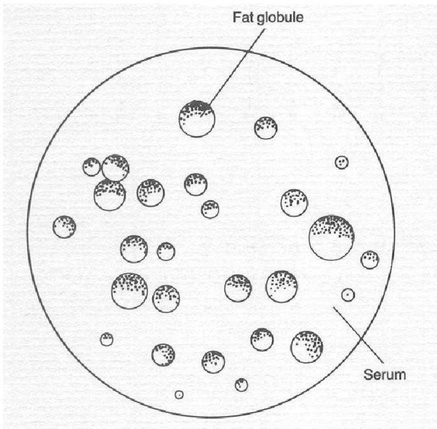
Lactose the source of energy in milk is less soluble in water than sucrose. It is also less sweet and easily breaks down to glucose and galactose by enzyme galactosidase present due to bacterial action and then gets fermented to lactic acid especially when milk goes sour. Under controlled conditions they can also be fermented to other acids to give a desired flavour, such as propionic acid fermentation in Swiss-cheese manufacture. Heating milk to above 100°C causes lactose to combine irreversibly with the milk proteins reducing the nutritional value of the milk and turning it brown.



People unable to metabolise lactose suffer from an allergy called lactose intolerance. To overcome this difficulty pre-treatment of milk with lactase enzyme to break down lactose becomes necessary.

Lactose is readily fermented by lactic acid fermenting bacteria to produce lactic acid. It exists in two water soluble isomeric forms, α and β of which the β form is more soluble. The lactose content in milk is inversely proportional to its ash content.

MILK FAT



If milk is left to stand, a layer of cream forms on the surface that differs considerably in appearance from the lower layer of skim milk. When seen under a microscope cream consists of a large number of floating spheres of varying sizes. Each sphere is surrounded by a thin skin or the fat globule membrane acting as the emulsifying agent for the suspended fat. This membrane protects the fat from enzymes and prevents the globules coalescing into butter grains.

About 98% of milk fat is a mixture of triacyl glycerides. In addition neutral lipids, fat-soluble vitamins and pigments (e.g. carotene, which gives butter its yellow colour), sterols and waxes are also present. Milk fats gets oxidised and supply a concentrated source of energy of about 9 calories / gm to our body. It also acts as a solvent for the fat-soluble vitamins A, D, E and K and is the source of essential fatty acids. (viz., linoleic, linolenic and arachidonic).

A fatty acid molecule comprises a hydrocarbon chain and a carboxyl group (-COOH). In saturated fatty acids the carbon atoms are linked in a chain by single bonds. In single unsaturated fatty acids there is one double bond and in poly-unsaturated fatty acids there is more than one double bond. Fatty acids vary in chain length from 4 carbon atoms, as in butyric acid (found only in butterfat), to 20 carbon atoms, as in arachidonic acid. Nearly all the fatty acids in milk contain an even number of carbon atoms. Fatty acids can also vary in degree of unsaturation, e.g. C18:0 stearic (saturated), C18:1 oleic (one double bond), C18:2 linoleic (two double bonds), C18:3 linolenic (three double bonds).

However milk fat present as an oil-in-water emulsion can be broken down by shaking or any such mechanical action.

Principal fatty acids found in milk triglycerides.

	Molecular formula	Chain length	Melting point
Butyric	**$CH_3(CH_2)_{2COOH}$**	**C_4**	**–8°C**
Caproic	$CH_3(CH_2)_{4COOH}$	C_6	–2°C
Caprylic	$CH_2(CH_2)_{6COOH}$	C_8	16°C
Capric	$CH_3(CH_2)_{8COOH}$	C_{10}	31.5°C
Lauric	$CH_3(CH_2)_{10COOH}$	C_{12}	44°C
Myristic	$CH_3(CH_2)_{12COOH}$	C_{14}	58°C
Palmitic	$CH_3(CH_2)_{14COOH}$	C_{16}	64°C
Stearic	$CH_3(CH_2)_{16COOH}$	C_{18}	70°C
Arichidonic	$CH_3(CH_2)_{18COOH}$	C_{20}	-49.5 (?)
Oleic	$CH_3(CH_2)_{7CH}{=}CH(CH_2)_{7COOH}$	$C_{18:\,1}$	13°C
Linoleic	$CH_3(CH_2)_4(CH{=}CH.CH_2)_2(CH_2)_{6COOH}$	$C_{18:\,2}$	–5°C
Linolenic	$CH_3.CH_2(CH{=}CH.CH_2)_3(CH_2)_{6COOH}$	$C_{18:\,3}$	NA

The melting point and hardness of the fatty acid is affected by the length of the carbon chain, and the degree of unsaturation. As chain length increases, melting point increases. As the degree of unsaturation increases, the melting point decreases. Fats composed of short-chain, unsaturated fatty acids have low melting points and are liquid at room temperature, i.e. oils. Fats high in long-chain saturated fatty acids have high melting points and are solid at room temperature.

Butterfat is a mixture of fatty acids with different melting points, and therefore does not have a distinct melting point. Since butterfat melts gradually over the temperature range of 0–40°C, some of the fat is liquid and some solid at temperatures between 16 and 25°C. The ratio of solid to liquid fat at the time of churning influences the rate of churning and the yield and quality of butter.

Fats in foods are subject to two types of deterioration that affect the flavour of food products.

Hydrolytic rancidity*:* In hydrolytic rancidity, fatty acids are broken off from the glycerol molecule by lipase enzymes produced by milk bacteria. The resulting free fatty acids are volatile and contribute significantly to the flavour of the product.

Oxidative rancidity*:* Oxidative rancidity occurs when fatty acids are oxidised. In milk products it causes tallowy flavours. Oxidative rancidity of dry butterfat causes off-flavours in recombined milk.

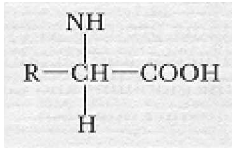
Milk fat is endowed with short and medium chain fatty acids and exists in an aqueous emulsion phase. It is easily digested because of the ability of the lipases to split the ester bonds and gets absorbed in our body through the intestinal tract unlike vegetable oils that comprises of long chain fatty acids and has to be emulsified with bile salts, enzymes from pancreas and fat splitting lipases.

Supplementing of milk fat in our diet increases energy density. When compared to human milk, cow milk is low in essential fatty acids such as linoleic and linolenic acids. Short and medium chain fatty acids with 4-12 carbon atoms, present in milk fat, are reported to have antibacterial and fungistatic activity and play an important role in preventing tooth decay by forming a protective coat over tooth enamel.

Recent experimental models devised to elicit the active properties, of the conjugated linoleic acid (CLA), a natural constituent of milk and other dairy products have shown that CLA possess anticarcinogenic property in the fore stomach of mouse, prevention of mammary cancer in rats, anti carcinogenic activity in the colon of rat, anti carcinogenic activity in skin, anti diabetic activity in rats, reduction in body fat and anti atherogenic activity in rabbits and immuno modulation in rats.

Apart from conjugated linoleic acid, milk fat has several bioactive and bio-protective molecules such as sphingomyelins, butyric acid, myristic acid, beta carotenes and fat soluble vitamins that show promise in anti-carcinogenic effect and immuno-stimulatory effect. Butyric acid is known for its anticancer properties and in improving immunity by activating macrophages.

MILK PROTEINS



Proteins are an extremely important class of naturally occurring compounds that are essential to all life processes. They perform a variety of functions in living organisms ranging from providing structure to reproduction. Milk proteins represent one of the greatest contributions of milk to human nutrition. Proteins are polymers of amino acids and have the general structure where R represents the organic radical. Each amino acid has a different radical and its content and the sequence in a protein molecule affect its properties. Twenty different amino acids occur regularly in proteins.

Proteins that contain substances other than amino acids are called conjugated proteins.

Phosphoproteins: Phosphate is linked chemically to these proteins—examples include casein in milk and phosphoproteins in egg yolk.

Lipoproteins: These combinations of lipid and protein are excellent emulsifying agents. Lipoproteins are found in milk and egg yolk.

Chromoproteins: These are proteins with a coloured prosthetic group and include haemoglobin and myoglobin.

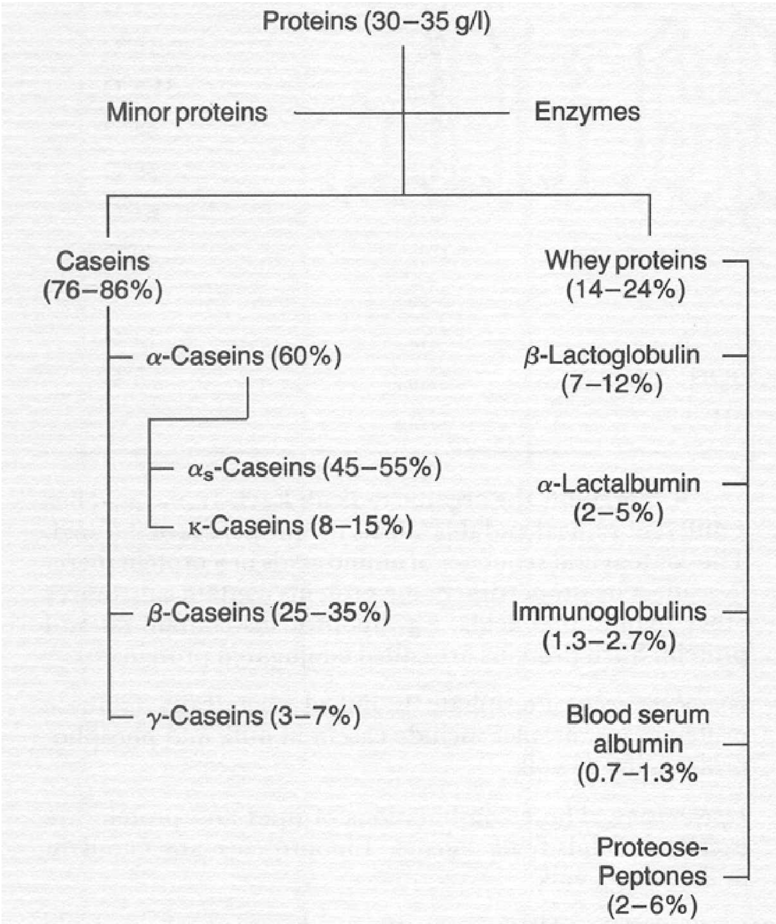
Milk protein is a rich source of essential amino acids. Milk contains a number of protein components, which differ in composition. They are the principal nitrogenous constituents of milk, made up of several amino acids bound together by a peptide bond. In milk they are partly in true solution and mostly as colloidal suspension. The main proteins of milk are caseins, alpha lactalbumin and beta lactoglobulins.

Casein

Casein is a distinct heterogeneous protein made up of a number of fractions. Casein precipitates at pH 4.6 and is exclusive to milk. It is present in spherical bodies as micelles, which vary in size with a negative surface charge. The micelles are stabilised by the K-casein. Caseins are hydrophobic but K-casein contains a hydrophilic portion known as the glycomacropeptide and it is this that stabilises the micelles. The caseins of milk may be sub-divided into five main classes, α, αs, β, γ, and κ-caseins. In milk, casein is present in combination with calcium in the form of calcium caseinate or more precisely calcium hydrogen caseinate. The structure of the micelles is not fully understood.

When the pH of milk is changed, the acidic or basic groups of the proteins get neutralised. When the positive charge on a protein exactly equals the negative charge, the net total charge of the protein is zero. This pH is called the isoelectric point of the protein (pH 4.6 for casein). If an acid is added to milk, or if acid-producing bacteria are allowed to grow in milk, the pH falls low. As the pH falls the charge on casein falls and it precipitates out causing curdling of milk and becoming sour.

WHEY PROTEINS



After the fat and casein have been removed from milk, one is left with whey. Whey contains soluble milk salts, milk sugar and the remainder of the milk proteins. Whey proteins are made up of a number of distinct proteins fractions namely α-lactoglobulin and β-lactoglobulin, in addition to globulin and albumin fractions. β-lactoglobulin accounts for about 50% of the whey proteins, and has a high content of essential amino acids. It gets coagulated by heat and forms a complex with K-casein when milk is heated to more than 75°C, affecting the functional properties of milk, the most important being the change in flavour.

Sulphur containing important essential amino acids, are also found in higher concentration in whey protein than in casein. Similarly α-lactalbumin, whose biological value, net protein utilization and protein efficiency ratio is considered superior to casein is also found in whey protein. Other whey protein fractions include α-lactalbumin, β-lactoglobulin, immunoglobulins, lactoferrin, transferrin, proteose-peptone fractions etc.

Most of these are globular proteins and get denatured by heat. α-lactalbumin exists in A and B forms and is susceptible to denaturation by unfolding of the tertiary structure. β-lactoglobulin is identical to blood globulin is insoluble in water and responsible for the transfer of antibodies. Normal milk contains 0.1% β-lactoglobulin whereas colostrum contains 6%. An important point to note is, normally double the quantum of vegetable protein is required to meet the daily requirement of essential amino acids when compared to that obtained from the milk proteins.

Whey proteins can be used to increase the net protein utilization of the entire food consumed. Whey proteins have better digestibility when compared to casein. The buffering capacity of milk proteins is very high and hence is very useful in the treatment of inflammation of mucous lining of the stomach in case of stomach ulcers. They are also used in the treatment of hyperuricaemia. The higher biological value of milk proteins naturally makes it as an ideal choice for persons suffering from renal impairment and fitness diets.

OTHER MILK PROTEINS

In addition to the major protein fractions outlined above, milk also contains a number of enzymes. The main enzymes present are lipases, which cause rancidity, particularly in homogenised milk, and phosphatase enzymes, which catalyse the hydrolysis of organic phosphates. Peroxidase enzymes, which catalyse the breakdown of hydrogen peroxide to water and oxygen, are also present.

Lactoperoxidase is activated thus making it inactive to alkaline phosphatase during pasteurisation and preservation of milk. Milk also contains protease enzymes, which catalyse the hydrolysis of proteins, lactalbumin, bovine serum albumin, the immune globulins and lactoferrin, which protects the young calf against infection. Milk derived peptides like β-casomorphins from β-casein, exorphin from α-casein, β-lactostensin from lactogobulin and serorphin from serum albumin have opium like sleep inducing functional properties

MILK SALTS

Milk salts are mainly chlorides, phosphates and citrates of sodium, calcium and magnesium. Although salts comprise less than 1 % of the milk they influence its rate of coagulation and other functional properties.

Calcium, Magnesium, Phosphorous and Citrate are distributed between the soluble and colloidal phases. Salt constituents in milk exist in colloidal and soluble forms. Colloidal minerals in milk are found attached with the casein micelles while soluble minerals are found dissolved in serum.

Major minerals in milk (mg/100ml) Elements	Cow	Buffalo
Calcium	129.4	176.88
Phosphorus	87.08	112
Sodium	-	-
Potassium	-	-
Magnesium	12.87	17.74
Chloride	-	-
Carbonates (as CO2)	-	-
Citrate (Citric Acid)	271.04	220

In general, about 33% of Ca, 33% of P, 75 % of Mg and 90% of citrates of milk are present in dissolved state. The colloidal salts are in equilibrium with dissolved salt; and heating causes shift of minerals from soluble to colloidal form while souring of milk carries the reverse effect.

If the ratio of soluble Ca and Mg to citrates and phosphates (salt balance ratio) is disturbed the stability of milk also changes. Their equilibria are altered by heating, cooling and by any pH change.

In addition to the major salts, milk also contains trace elements like Copper, Iron, Cobalt, Manganese, Silica and Zinc with some of the elements coming from animal feeds, milking utensils, equipments, etc.

MILK VITAMINS

Various vitamins present in milk are as follows. Fat Soluble Vitamins - include vitamins A, D, E and K. Water-soluble vitamins are the, B1 (thiamine), B2 (riboflavin), B6 (pyridoxine), Biotin, Niacin (nicotinic acid), Pantothenic Acid, Para-amino benzoic acid, Inositol, Choline, Folic acid, B12, and Ascorbic acid. Fat rich milk products contain large quantities of fat soluble vitamins, whereas whole milk, skim milk, buttermilk and whey are a good source of water soluble vitamins. Vitamins are unstable in nature and milk processing can reduce the effectiveness of vitamins present in milk.

PHOSPHOLIPIDS

Principally milk phospholipids are the Lecithin, Cephalin and Sphingomyelin. These contain phosphorus in their molecules in addition to the fatty acids, glycerol and a nitrogenous base. Though fat-soluble, they are hydrophilic and imbibe large quantity of water and swell. They are useful antioxidants in fat rich dairy products.

MINOR MILK CONSTITUENTS

In addition to the major constituents discussed above, milk also contains a number of organic and inorganic compounds in small or trace amounts some of which affect both the processing and nutritional properties of milk.