MILK COMPOSITION AND ITS CONSTITUENTS

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Milk, according to the PFA Rules, is the normal mammary secretion derived from the complete milking of healthy milch animal without either addition thereto or extraction there from. It shall be free from colostrums. Milk of different classes and of different designations shall conform to standards laid down in Table. It shall conform to both the parameters for milk fat and milk solids-not-fat (SNF), independently.

Milk is valued commercially for its two important parameters: (i) Milk fat (F) and (ii) Solids-Not-fat (SNF). The SNF largely consists of proteins, lactose and minerals. These solids are also referred to as 'serum solids'. These two parameters usually form the basis of payment to milk producers in India. The term 'total Solids' (TS) refers to the quantity of SNF plus fat present in milk. It may range from 12 to 16 per cent, depending on its source. For cow milk, TS is 12 per cent (3.5%F and 8.5% SNF) while for buffalo milk it ranges between 15 and 16 per cent (6-7% F and 9% SNF).

Table1 Average composition of milk of different animal species

Species	Water	Fat	Protein	Lactose	Ash
Friesian cow[1]	87.92	3.40	3.13	4.86	0.69
Sindhi cow[2]	86.07	4.90	3.42	4.91	0.70
Gir cow[2]	86.44	4.73	3.32	4.85	0.66
Tharparkar cow[2]	86.58	4.55	3.36	4.83	0.68
Sahiwal cow[2]	86.42	4.55	3.33	5.04	0.66
Crossbred cow[2]	86.54	4.50	3.37	4.92	0.67
Buffalo[3]	82.76	7.38	3.60	5.48	0.78
Goat[4]	87.10	4.25	3.52	4.27	0.86
Sheep[5]	81.00	7.90	5.80	4.50	0.80
Camel[6]	86.50	3.10	4.00	5.60	0.80

[1]Bos Taurus; [2]Bos indicuis; [3]Bos bubalis; [4]Capra hircus; [5]Ovis aries; [6]Camelus dromedarius.

Table 2 PFA standards for different classes and designations of milk in India

Class of milk	Designations	State and Union Territories	Minimum percentage	
			Milk fat	Milk solids-not-fat (SNF)
Buffalo milk	Raw, pasteurized, boiled, flavoured and sterilized	Assam; Bihar; Chandigarh; Delhi; Gujarat; Maharashtra; Haryana; Meghalaya; Punjab; Sikkim; Uttar Pradesh; West Bengal; Andaman & Nicobar; Andhra Pradesh; Arunachal Pradesh; Dadra & Nagar Haveli; Goa; Daman & Diu; Kerala; Himachal Pradesh; Jammu & Kashmir; Karnataka.	6.0	9.0
	-do-	Kerala; Lakshadweep; Madhya Pradesh; Manipur; Mizoram; Nagaland; Orissa; Pondicherry; Rajasthan; Tripura; Tamil Nadu.	5.0	9.0
Cow milk	-do-	Chandigharh; Haryana; Punjab.	4.0	8.5
	-do-	Andaman & Nicobar; Andhra Pradesh; Arunachal Pradesh; Assam; Bihar; Dadra & Nagar Haveli; Delhi;Goa; Daman & Diu; Gujarat; Himachal Pradesh; Jammu & Kashmir; Karnataka; Kerala; Lakshadweep; Madhya Pradesh; Maharashtra; Manipur; Meghalaya; Nagaland; Pondicherry; Rajasthan; Sikkim; Tamil Nadu; Tripura; Uttar Pradesh; West Bengal.	3.5	8.5
	-do-	Mizoram; Orissa;	3.0	8.5
Goat or sheep milk	-do-	Chandigharh; Haryana; Kerala; Madhya Pradesh; Maharashtra; Punjab; Uttar Pradesh.	3.5	9.0
	-do-	Andaman & Nicobar; Andhra Pradesh; Arunachal Pradesh; Assam; Bihar; Dadra & Nagar Haveli; Delhi;Goa; Daman & Diu; Gujarat; Himanchal Pradesh; Jammu & Kashmir; Karnataka; Kerala; Lakshadweep; Manipur; Meghalaya; Mizoram; Nagaland; Orissa; Pondicherry; Rajasthan; Sikkim; Tamil Nadu; Tripura; West Bengal.	3.0	9.0
Mixed milk	-do-	All India	4.5	8.5

Standardized milk	Pasteurized, flavoured and sterilized	All India	4.5	8.5
Recombined milk	-do-	All India	3.0	8.5
Toned milk	-do-	All India	3.0	8.5
Double toned milk	-do-	All India	1.5	9.0
Skimmed milk	-do-	All India	Not more than 0.5	8.7
Full cream milk	Pasteurized, and sterilized	All India	6.0	9.0

Constituents of Milk

Water- Water is the medium in which all the other components of milk (total solids) are dissolved or suspended. Water content varies from 83.18 per cent to 87.2 per cent in milk of different species of cows and buffaloes. In commercial milk, the range is 83.18 to 86.50 per cent. A small percentage of water in milk is hydrated to the lactose and salts, while some portion is bound with proteins.

Fat- Milk fat, though quite bland in taste, imparts richness/smoothness to fat-containing dairy products. In freshly secreted milk, it occurs as a microscopic globular emulsion of liquid fat in an aqueous phase of milk plasma. Fat is the most variable component of milk. The average size of fat globules in buffalo milk is larger (4.15 to 4.60 μm) than that of cow milk (3.36 to 4.15 μm.).

The fat globules are stabilized by a very thin membrane of 5-10 μm thickness, closely resembling plasma membrane. The membrane consists of proteins, lipids, lipoproteins, phospholipids, cerebrosides, nucleic acids, enzymes, trace elements and bound water. The membrane is important in keeping fat from separating as free oil when it is subjected to physical abrasion during handling/processing of milk. It also protects milk lipids against the action of enzymes, notably lipase, in development of rancidity. Certain enzymes such as alkaline phosphatase and xanthine oxidase as well as certain important minerals like iron and copper, are preferentially attached to the fat globule membrane.

Both buffalo and cow milk fats consist chiefly of the triglycerides of fatty acids, which make up 95-99 per cent of milk fat. The remaining portion of milk fat is composed of di-glycerides (about 4.1% in buffalo milk and 1.26-1.59% in cow milk), mono-glycerides (about 0.7% in buffalo milk and 0.016-0.038% in cow milk). High, medium and low molecular weight triglycerides in buffalo milk occur in the proportion of 42.5, 17.1 and 40.5 per cent, respectively. Corresponding values for cow milk fat are 52.9, 18.9 and 28.2 per cent, respectively. Free fatty acid content of buffalo milk fat is lower (0.22%) as compared to that in cow milk fat (0.33%). The functional properties of milk fat are attributed to its fatty acid make up. In general milk fat consists of 65% saturated, 32% mono-unsaturated and 3% poly-unsaturated fatty acids. It also contains 7% short chain fatty acids (C_4- C_8), 15-20% medium chain fatty acids (C_{10}- C_{14}) and 73-78% long chain fatty acids (C_{16} or higher)

Buffalo milk fat contains appreciably higher butyric acid in its triglycerides in comparison to cow milk fat. However, other short-chain fatty acids (caproic, caprylic, capric and myristic are lower in buffalo milk fat. The content of long-chain fatty acids (palmitic and stearic) is relatively higher in buffalo milk. The unsaturated fatty acid level of buffalo and cow milk is comparable. Milk fat also consists of varying quantities of other lipids such as phospholipids, sterols, carotenoids, vitamins A, D, E and K and some traces of free fatty acids.

Cholesterol- The cholesterol content of milk is significantly affected by the species, breed, and stage of lactation and season of the year. Generally, the cholesterol content is higher in the western breeds of cattle (317-413 mg/100 g fat), followed by zebu (humped like desi) cow (303-385mg/100g fat). It is lowest for buffalo (235-248 mg/100 g fat.

The cholesterol content, generally lowest at the beginning of the lactation period, progressively rises to reach its highest level towards the end.

Phospholipids- The total phospholipids content of buffalo milk fat averages 21.04 mg/100 ml of milk, whereas for cow milk the corresponding value is 33.71 mg/100 ml.

Proteins- Casein, the principal milk protein, makes up 80 per cent of the total protein content, and whey proteins the remaining 20 per cent. These fractions, shown to be heterogeneous, consist of many proteins.

Casein: Typical of milk proteins, caseins display a distinctive structure as well as physical, biological and nutritional properties. These exist in milk as particles called casein micelles, which are made up of calcium phosphate and casein compounds. A very small proportion of casein is also present in soluble form.

Caseins are heat stable. Heat has little or no effect on casein molecules since they exist naturally in an open and extended state (gain in water entropy) at higher temperatures. However, heating of milk at elevated temperatures for a long time may result in hydrolytic cleavage of peptide and phosphate bond, which affects the stability of the complex, contributing to coagulation of milk.

The ε - amino group of lysine present in casein interacts with the aldehyde group of lactose at elevated temperature, leading to the formation of brown pigments (Maillard reaction). This explains browning of heat desiccated products during storage.

As compared to cow milk, buffalo milk is richer in total proteins, particularly the casein and whey proteins. However, the proportion of various protein fractions is similar in milk of both species. Distinct differences exist in the physico-chemical make-up of casein in buffalo and cow milk. The proportion of micellar casein is nearly 100 percent in buffalo milk. While that of soluble casein is very low whereas in cow milk, the micellar casein is of the order of 90-95 percent. The voluminosity of the buffalo milk casein is 2.68-3.72 ml/g at 25-27 ˙C, while that of the cow milk casein is 4.18 ml/g. The hydration (salvation) of the buffalo micelle is lower92.60-2.90g water/G casein) as compared to 3.48 g water/ g casein for cow micelle. The particle size of the buffalo casein micellar is larger in diameter (70-160 nm). Than that of the cow casein micellar (70-110 nm). The buffalo casein micelle is more opaque (about three times, when suspended in a different medium) than the cow micelle. Buffalo casein has superior whitening as compared to that of cow casein due perhaps to a higher proportion of calcium present in it. The magnesium content of buffalo milk is also higher. However, the sialic acid and hexose content in buffalo milk is lower than that in cow milk.

Whey Proteins: Whey proteins consist of β - lactoglobulin and α - lactalbumin, bovine, serum albumin, immuno-globulins (mainly IgG1, IgG2 and IgM), lactoferrin, proteose-peptone, serum transferring, glycoprotein and enzymes.

The average range of whey proteins in buffalo milk is found to be between 0.70-0.74 per cent as compared to 0.50-0.53 per cent in cow milk. The average level of proteose-peptone in buffalo milk (330.5mg/100ml) is significantly higher than that in cow milk (240mg/100ml).

β – lactoglobulin : This is the major whey protein present in buffalo and cow milk. The buffalo milk β -lactoglobulin displays molecular weight of about 38500 daltons, as compared to that of 37600 daltons observed in the cow milk β – lactoglobulin. In milk of Indian cattle breeds, mainly two genetic variants (A and B) of β – lactoglobulin are found, whereas in the western breeds, C and D variants have also been reported. Milk of Murrah buffalo shows the pre-dominant presence of two genetic variants of β–lactoglobulin. Their electrophoretic mobility is similar to that of A and C variants of cow milk β – lactoglobulin. Buffalo milk also shows the presence of yet another variant of β– lactoglobulin, which has faster electrophoretic mobility than the A variant of cow milk β-lactoglobulin.

α-lactalbumins : The α-lactalbumins present in buffalo and cow milks display almost similar attributes. The molecular weight of buffalo α-lactalbumins has been observed to be 16,200 daltons and that of cow α-lactalbumins 14,176 daltons.There are two genetic variants of α-lactalbumins – variants A and B – both in cow and buffalo milk. They have almost similar electrophoretic mobility.

Immunoglobulins (Ig) : They are antibodies synthesized in response to stimulation by specific antigens. These offer non-specific humoral response to Gram-negative enteric and respiratory bacteria. There are five major classes of immunoglobulins, viz., IgA, IgD,IgE,IgG and IgM. The basic structure of all Igs is similar.These are composed of two identical light chain (23,000 daltons) and two identical heavy chains (53,000 daltons). Of the five- immunoglobulin classes IgG is the predominant fraction in buffalo milk and cow milk comprising about 90 per cent of the total colostral immunoglobulins. The average immunoglobulin content of buffalo milk is appreciablyhigher as compared to cow milk, being 76.2 μg/100 gm whey proteins in first milking post- partum. It decreases to 43.4 μg/100gm on the sixth day milking post-partum. The total concentration of immunoglobulins in Zebu cattle averages about 40. 1 μg/ml immediately after calving and it declines to 0.45 μg/ml on the eight day post-partum.

Lactoferrin : This glycoprotein displays a strong inhibitory effect towards Gram-negative enteropathogenic bacteria by virtue of its ability to bind free ionic iron, which is essentially required for the growth of enteropathogenic microorganisms. Apart from the antibacterial effect, a nutritional role in iron metabolism has also been ascrived to lactoferrin.The average lactoferrin content of buffalo milk is relatively higher at 0.32-0.5 mg/ml than the average of 0.15-0.32 mg/ml for cow milk.

Proteose-Peptone : The average level of Proteose-Peptone in buffalo milk at about 330.5 mg/100ml is significantly higher when compared to 240 mg/100ml in cow milk.

Lactose: Also called milk sugar, lactose in the major carbohydrate of milk. It is a disaccharide of one residue each of D- glucose and D-galactose. Buffalo milk usually contains lactose in the range of 4.7-5.0 per cent, while cow milk has slightly lower amounts of lactose in the range of 4.5-4.8 per cent. Crystallization of lactose, when milk is concentrated, is of importance in regard to the texture of end-products. Lactose has only 16-33 per cent of the sweetening power of sucrose. The application.Its use as icing toppings and various types of fillings can improve the quality of food formulation Being a reducing suger, it reacts with proteins to form a highly flavoured golden brown substance, commonly found on the the crust of baked foods. Lactose contributes significantly to the flavour, texture, appearance, shelf life and toasting qualities of baked food. The pharmaceutical industry has used lactose for many years for making tablets or pills. A compound formed from lactose in heated milk products is lactose. It stimulates the growth of *Bifidobacterium bifidum* and is thus beneficial for producing useful microflora in the gut.

Minerals: Average normal milk contains 0.70 per cent ash, and this amount represents a salt content of about 0.90 per cent. The percentage of salt and ash in milk varies with the breed, feed, season, and stage of lactation and disease. The white residue after incineration of a given weight of milk is used as a measure of the mineral content of milk.

The mineral make-up of milk is crucial to the stability of the the physico- chemical equilibrium. Mineral in milk are present in complex equilibria such as colloidal, soluble: ionic and non-ionic. Although their concentration is less than 1 per cent in milk, minerals significantly affect the technological behaviour of milk thus:

- Heat stability and alcohol coagulation of raw milk;

- Preparation quality and storage stability of products like concentrated/ heat desiccated milk products;
- Clumping of fat globules upon homogenization of cream; and
- The calcium content of milk influences the firmness of curd during paneer and chhana making.

Sodium potassium and chloride are to a large extent (95-96%) present in true solution and in ionic form and therefore diffuse freely across the membrane during ultra filtration, reverse osmosis and electro dialysis of milk and whey.

Buffalo milk contains more calcium and phosphorus (0.22% and 0.13% respectively) than cow milk (0.12% and 0.09% respectively). Moreover, the calcium / phosphorus ratio is higher in buffalo milk (2.26) than cow milk (1.96). Buffalo milk has low heat stability in concentrated form (2:1). Heat stability, expressed as heat coagulation time at 130 C, is 31.82 minutes for buffalo milk, while it is 32.28 minutes for cow milk. However, when milk is concentrated 2 to 1, their corresponding hat coagulation times are 2.89 and 6.58 minutes, respectively. The soluble forms of calcium, magnesium and citrate are lower in buffalo milk than in cow milk.

MINOR CONSTITUENTS

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Enzymes: Milk is a repository of a variety of enzymes. Over 20 enzymes have been isolated and characterized in cow milk. However, information on enzymes in buffalo milk is very limited. Several enzymes in milk are utilized for quality testing and control. Some important enzymes from the processing point of view are listed below:

Alkaline Phosphatase: This enzyme has assumed significance because of the relationship between the temperature at which it is inactivated and the temperature employed for pasteurization of milk. Buffalo milk on an average contains about two-thirds the activity of alkaline phosphates as that of cow milk. Alkaline phosphates are distributed through milk although its concentration is higher in the cream fraction.

Lipoprotein Lipase: Residual activity of lipase remaining in processed milk and milk products tends to reduce their shelf life.

Protease/ Plasmin: Residual proteolytic activity in processed milk and milk products reduces their shelf life.

Lactoperoxidase (LP): This enzyme is naturally present in milk. One of its unique biological functions is an antibacterial effect in the presence of hydrogen peroxide and thiocyanate. Both these substances are naturally present in milk in varying concentrations. This enzyme has gained significance in view of its supportive role in preserving raw milk under ambient conditions through the LP-system. Lactoperoxidase activity in buffalo milk ranges between 5.2-9.8 units/ml, which is on average higher than the corresponding value of 4.4-7.2 units/ml for cow milk.

To activate LPs in milk, add about 10 PPM (parts per million) of thiocyanate (preferably in powder from) to the raw milk. It increases the overall thiocyanate level to 15 PPM from the natural level of 5 PPM. The resultant solution is thoroughly mixed for 30 seconds and then an equimolar amount (8.5 PPM) of hydrogen peroxide is added (generally in the form of a granulated sodium carbonate peroxyhydrate). The LPs activation has a bacteriostatic effect on the raw milk, expending its shelf life by 7-8 hours under tropical conditions. This means the milk can be transported from the collection point to the processing plant without affecting its quality, thereby significantly increasing the income generated at the producer group level.

Lysozyme: This is a relatively small, single peptide chain protein. Its content in cow milk is about 13 μg/100 ml. recently emphasis has been focused on the antibacterial role of lysozyme as a natural defense in milk. During mastitis, lysozyme levels in milk tend to increase considerably, being in the range of 100 to 200 μg/100 ml.

Pigments: In cow milk, carotene is the major pigment and it is derived from feed. It gives cow milk a pale yellow appearance. However, buffalo has the ability to convert carotene into Vitamin A before passing it into milk and so it appears white. Milk also contains bile pigments. Among them, biliverdin predominates. Its concentration is 50.4 mg/ml of milk. It binds to caseins. In the course of souring of milk, biliverdin is detached from casein and it undergoes a rapid chemical to a fat-soluble yellow pigment called bilirubin. Butter fat from soured buffalo milk develops a greenish yellow colour, reaching its maximum in two hours. This is attributed in the presence of two pigments, non-carotenoid in nature. Of them, the predominant pigment is green and the minor component is blue.