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A review on examination of breast milk in Ayurveda and its physicochemical properties with its component

Review Article

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Abstract

Background: Breast milk is the only and complete nutrition supply for infants breastfeeding contains all the necessary elements for healthy baby growth and development. Breast milk contains bioactive agents that help in the proper functioning of the gastrointestinal system which affects the circulatory system. Objectives: The objective is to review recent findings on the different content and components of breast milk from premature and term infants, examination protocol of breast milk, the physicochemical properties of mature breast milk, recent knowledge of the prebiotic and probiotic effect of breast milk through different articles, classical books of Ayurveda then summarized the evidence available. Materials and methods: The information is collected from articles which are from PUBMED, SCOPUS, GOOGLE SCHOLAR, and only classical books of Ayurveda. Observation: The composition of breast milk including protein, carbohydrate, fat, minerals, vitamin, and immunoglobulin contribute to the nutrition and immunity action of breast milk. The composition of breast milk i.e. macronutrients and micronutrients differ at different stages of breast milk including colostrums, transitional and mature milk. The result of physicochemical properties differs according to the methods used for the evaluation of the different physicochemical properties of breast milk but the mean value is taken into consideration. Results: The different parameters in the examination of breast milk as per Ayurveda. The physicochemical properties of breast milk include pH, specific gravity, electrical conductivity, viscosity, water activity, osmolarity, refractive index, freezing point, and surface tension. Conclusion: This review emphasis on the importance of breast milk examination on the basis of Ayurveda and modern methods to ensure its purity. It is a complete and essential nutrition for an infant so it must be pure and its examination is necessary for not only the prevention of infant diseases but also to diagnose the morbidity of the mother.

Key Words: Breast milk, Examination, Physicochemical, Microbiome, Composition, Colostrum.

Introduction

The world health organization advises continuing exclusive breastfeeding for the first 6 months and beyond 2 years of age (1). Breast milk is the only complete nutrition supply for breastfed infants, containing all the components necessary for the infant's healthy growth and development. It provides nutritional as well as health benefits to infants and reduces the risk of later chronic disease (2). Macro- and micronutrients are essential for infant growth and development, and also have a significant impact on neural development, metabolic processes, oxygen transport, and DNA synthesis. (3). Lack of micronutrients and vitamins can cause diseases including rickets, hemolytic anemia, xeropthalmia, etc. (4). Maternal diet greatly influences the composition of macro and micronutrient and vitamins content of the breast milk. Many studies have been conducted about breast milk composition with the maternal diet and some controversies are also seen (5-8). The physicochemical characteristics such as specific gravity, osmotic pressure, water activity, freezing point, refractive index, surface tension, pH value, electrical conductivity, and viscosity are significant parameters when investigating the quality of milk. Physical properties are strongly dependent on their chemical composition. In Ayurveda, breast milk is called *Stanya*, and a description of *Stanya* was given in almost all Samhita. *Shuddha Stanya* (good quality breast milk) (Table 1), *Stanya Dushti* (vitiated breast milk), and its type with its characteristic features and the diseases which they cause are well described by different Acharya (Table 2).

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Material and methods

A systematic review was done by computerized search strategies through databases like Pub med, Google Scholar, and Scopus in august 2022 searching keywords such as 'breast milk, physiological of breast milk, the composition of breast milk, Physico-chemical

Table 1: *Shuddha Stanya* according to different Acharya

As per Shusruta (9)	As per Charaka (10)	As per Kasyapa (11)
Shankhapramaneshvetam (color of shell i.e. White or light yellow	*Prakrta varna*(normal color)	*Avyahata Bala* (unhampered strength)
Madhura (sweet)	*Prakriti gandha*(normal smell)	*Avyahataanga*(unhamper body parts)
Laghu (light nature)	*Prakrta rasa* (normal taste)	*Arogya*(healthy)
Pathyakara (good for health),	*Prakrta sparsha* (normal touch)	*Vardhatesukham*(growth and development)
Deepana (increase digestive fire)	Gets mixed evenly with water	Trouble-free to child
Jeevanam(promoting life)	*Pushtikara* (nourished the child)	Trouble-free to wet-nurse
Snehana (unctuous)	*Arogyakara* (prevent diseases)	
Brumhaniya (increase body size),		
Shareeropachaya (development of the body)		
Balarudhirakara (stiffness to *Bala*),		
Pushtikara (nourishes the body),		
Arogyakara (prevent diseases)		

Table 2: *Dushta Stanya* with its cause, type, features, and diseases it causes

Dosha	Cause (12)	Type (13)	Features (14,15)	Diseases (16)
Vata	Breast milk gets vitiated by different factors like *Ahara* (food), *Vihara* (activity), *Manasika* (activity of mind), etc. *Aharaja* includes food that intake *Lavana* (salty), *Katu* (spicy), *Amla* (sour), *Ksara* (alkaline taste), *Asatmya* (unconducive), and *Virudha* (contrary to body). *Viharaja* includes carrying out *Vegodharana* (stopping natural urges), *Divasvapna* (day sleep), etc. *Manasika* is *Krodha* (anger), *Shoka* (grief), and *Chinta* (overthinking). Kashyapa described *Grahas* (seizing force) also as the cause of *Stanya Dushti*	*Vairasya*	*Syava* (blackish) or *Aruna* (reddish) in color, *Kashaya* (astringent) in *Anurasa* (after taste), *Vishada* (clear), *Analakshya Gandha* (absence of any conspicuous smell), *Ruksha* (dry), *Drava* (liquid), *Phenila* (frothy), *Laghu* (light), *Atruptikara* (not satisfying), and causing *Karshanam* (emaciation)	*Durbala* (weakness)
		Phenasanghata		*Vrudhi* (growth)
		Ruksha		*Swara Kshinata* (low voice)
				Mala Avarodha (stool obstruction)
				Mutra (urine) *Avarodha*
				Vayu (eructation) *Avarodha*
				Shirashool (headache)
				Peenasa (cold)
Pitta		*Vaivarnya*	*Krushna* (blackish), *Nila* (bluish), *Peeta* (yellowish) or *Tamra* (coppery) in color: *Tikta* (bitter), *Amla* (sour) or *Katu* (pungent) in *Anurasa* (aftertaste), *Kunapa Rudhira Gandhi* (having smell like that of a dead body or blood), *Brusha ushna* (excessively hot)	*Swedaadhikya* (sweating more)
		Vaigandhya		*Trishna* (thirst)
		Paichilyam		*Dravamlapravrutti* (loose stool)
				Shareerasparshushna (hot in touch)
				Pandu (anemia)
				Kamala (jaundice)
Kaphaja		*Gauravam*	*Atyartashukla* (exceedingly white), *Atimadhu* (excessively sweet) and having *Lavana* (saline) *Anurasa* (tastes) having the smell of *Ghrita* (ghee), *Taila* (Oil), and *Vasaa* (muscle fat) and *Majja* (bone marrows), *Picchila* (slimy) and thready settling down at the bottom when poured into a vessel containing water	*Chardi* (vomiting)
		Atisneham		*Lalasrava* (salvation)
				Kasa (cough)
				Swasa (breathlessness)
				Tamakaswasa (asthma)
				Lalasrava (salivation)
				Mukha Netrapradeshi Shotha (swelling of face and peri-orbital)
				Hrudroga (heart disease)

of breast milk. We include studies which are on the physiology of breast milk. The articles are screened by the reviewers and those articles which met the potential criteria were reviewed. The data were extracted and summarised in table form. In the studies in which the full texts were not available, a language other than Sanskrit and English were excluded.

Result

The composition of breast milk including protein, carbohydrate, fat, minerals, vitamin, and immunoglobulin contribute to the nutrition and immunity action of breast milk. The composition of breast milk i.e. macronutrients and micronutrients differ at different stages of breast milk including colostrum's, transitional and mature milk. The different parameters in the examination of breast milk as per Ayurveda. The

result of physicochemical properties differs according to the methods used for the evaluation of the different physicochemical properties of breast milk but the mean value is taken into consideration.

Discussion

Composition of breast milk

Acharya Charaka described human milk as *Jivanam* (promote life), *Bruhanam* (nourishes), *Satmya* (wholesome), and *Snehanam* (unctuous) (17). Breast milk composition is dynamic and varies according to the need for feeding, maternal factor, and environmental factor. Breast milk of mothers who deliver prematurely has a high content of protein, bioactive components, and minerals like free amino acids, sodium, zinc, and copper (18). Breast milk contents differ from the beginning till the end of feeding i.e. foremilk and hindmilk; also differ in the diurnal pattern of feeding. The difference may occur by the differences in the method applied for the estimation of this content. The fat content is relatively low in foremilk as compared to hind milk.

Macronutrient

Protein: The proteins of breast milk are casein and whey, the abundant proteins in breast milk are casein, lactoferrin, immunoglobulin A, albumin, and lysozyme. Casein is not easily digested and is present in the stomach in the form of clots (19). The whey and casein ratios in colostrum and mature milk are 90:10 and 60:40 respectively. The levels of protein in breast milk reduce after the first 4-6 weeks (20). Casein in breast milk, in the form of loose micelles and softer curds, is readily digested by carboxypeptidases (21). The protein concentration is largely unaffected by the intake of the mother but is affected by the maternal weight for a particular height (19). **Fat:** Human milk fatty acids are predominantly triglycerides (approximately 95% to 98%) and essential fatty acids (22). Fat in the form of long-chain polyunsaturated fatty acid (PUFA) like DHA has a positive impact on the infant's brain development. Fat is in more concentration in hind milk as compared to the foremilk. A short-chain fatty acid reduced the pH value of the gut hence inhibiting the growth of potentially harmful gut microflora (23). Fat in breast milk is more readily digested and absorbed attributed to the availability of bile salt-catalyst lipase which enhances pancreatic lipase (24). When breastfeeding mothers consume foods such as bread, snacks, fast foods, and sweets, trans fats can be found in breast milk for up to 7.7% of total fat (25). Arachidonic acid, EPA, and DHA are also correlated with arachidonic acid, etc. rich dietary intake in lactating mothers (26). Vegetarians, therefore, have very low DHA, and others are attributed to the shortage of fish and other non-vegetarian foods in their daily diets (27). **Carbohydrate:** The main carbohydrate present in breast milk is lactose and is seen in high concentrations in those who produce a high quantity of milk. Lactose is digested by lactase-phlorizin hydrolase available in the small intestinal. Lactose is well-digested by all infants except those having lactase deficiency in their small intestines (28). The other significant carbohydrate is Oligosaccharide; it has a role in the immune booster through its prebiotic effects. In a study (Dogaru et al), it was found that breastfeeding reduced the risk of asthma by 22%, with the greatest effect observed during early childhood. Our increasing lifestyles of hygiene and exaggerated consumption of antibiotics have led to certain bacteria depletion from the modern human microbiome (29). HMOs are only produced by the mammary glands during lactation and cannot be added to infant formula, although various types of oligosaccharides have recently been added to the formula (30). In Ayurveda to increase the effect of prebiotics, the mother can be advised with *Triphala, Yashtimadhu, and Ulmus Rubra* (slippery elm) (31).

Table 3: Macronutrient in preterm infants and post term infants. (32-35)

Macro-nutrient	Pre term		Term	
	Colostrum	Mature milk	Colostrum	Mature milk
Energy	45-75 kcal/L	61-94 kcal/L	50-60 kcal/100ml	57-83 kcal/L
Protein	2.5 g/dL	1.3-3.3 g/dL	14-16 g/L	0.9-1.5 g/dL
Lipid	2.1 g/dL	2.6-6.2 g/dL	15-20 g/L	2.2-5.0 g/dL
Lactose	54 g/L	6.4-8.8 g/dL	20-30 g/L	7.2-7.7 g/dL

Micronutrient

Minerals are less in breast milk, but no additional supplementation is necessary during exclusive breastfeeding due to their bioavailability which fulfilled the body's requirement. Minerals are abundant in colostrum and decrease as lactation progresses. Iron in breast milk binds to the lipid and low-molecular-weight fractions, and a lesser extent to lactoferrin. Iron is approximately 0.5-1.0 mg/L and 0.3-0.7 mg/L in colostrum and mature milk respectively (36). In exclusive breastfeeding infants, generally, it is not advised for iron supplementation but iron-enriched food is recommended after 6 months. Copper is detected primarily in the non-fat part of breast milk, but a significant quantity is also seen in the fat part of breast milk. It is account for 0.5-0.8 µg/L and 0.1-0.3 µg/L in colostrum and mature milk respectively (37). Minerals including iron, calcium, phosphorus, magnesium, etc. are not abundant but sufficient to meet the requirement of infants. **Vitamins:** Vitamins D and K may be inadequate to meet infant needs and supplementation may be required. Vitamin D is affected by both sun exposure and maternal diet. Vitamin K is given to newborns to prevent bleeding disorders (38). Vitamin D supplementation of 200 to 400 IU/day as a maintenance dose and 2,000 IU/day as a deficit is recommended for lactating women (39). In general, mothers on an improper diet may have vitamins B6, B12, and folic acid deficiency, but can have relatively adequate levels of thiamine and riboflavin (40).

Hormone and growth factors

Hormones and growth factors in breast milk include epidermal, nerve cell, brain-derived neutrophil

factor, glial cell-derived neutrophil factor, insulin-like, vascular endothelial, erythropoietin, and adiponectin. However, their functions and effects are not yet well known enough. Although not yet known, many studies on growth factors were known to affect the intestinal, vascular, nervous, and endocrine systems (20). An epidermal growth factor is higher in colostrum than in mature milk and promotes intestinal maturation and repair (41). Brain-derived neutrophil factor and glia-derived neutrophil factor are important for intestinal development in infants by acting on the enteric nervous system (42). IGF-1 and IGF-2 are plentiful in colostrum and are not substantially differ from preterm and term delivery (43). IGF1 protects enterocytes from oxidative stress, thereby stimulating erythropoiesis and hematocrit (44).

Microbiome

Breast milk plays an important role in maintaining a healthy microbiome in neonates and infants as it contains many bacteria and prebiotics. A healthy microbiome is developed in the first 3 yrs. of life and plays an important role throughout life. Depending on different sources of bacteria, different factors influence the microbiota such as the mother's gut bacteria, mode of delivery, mode of breastfeeding, etc. (45). In a 2003 study, Martin et al. demonstrated breast milk as an essential inception of lactobacilli for the gut of infant and found that DNA profile of lactobacilli in breast milk differed from that of the mother's skin (46). In another study, Heikkila and Saris conducted a culture-based study and concluded that an infant ingests 8×104-106 commensal bacteria from 800 mL of her breast milk. A breastfed infant is estimated to ingest 104–106 bacteria per day (if consuming milk of 800 mL per day), and most species belong to the genus *Staphylococcus*, *Streptococcus spp*. It belongs to the genera *Cocci, Lactobacillus,* and *Bifidobacterium* (47). Breast milk feeding practice also affects the infant's gut. There are approximately 10 % of bacteria instigated in the infant when breastfeeding is done directly from the breast, are on the skin encircling the mother's areola. It plays a critical role in decreasing the risk of diseases such as inflammatory bowel diseases, respiratory infections, wheezing, asthma, allergies, and inflammatory conditions. It also regulates cognition, anxiety, mood, etc. by the brain-gut axis. Bacteria including the genus *Streptococcus*. and *Veillonella dispar* are found in breast milk and infant feces, and co-occurrence is reduced when infants are fed expressed breast milk (48). It is also observed that there are some gender-specific interrelations (e.g. when the child was female, there is maternal BMI relation to milk microbiota), whereas factors like strain-specific effects (e.g. maternal atopy related to actinomycete abundance) (49)

***Stanya Pravritti* (stages of Lactation)**

The initiation of lactation developed in 2 stages. In the first 32-40 hrs. after delivery, there is a limited secretion of milk after lactogenesis starts. Late onset of lactogenesis is the onset of breast milk more than 72 hrs. after delivery and is seen more in preterm delivery and maternal obesity (50). Breastfeeding is not an essential component for the commencement of lactation but plays a crucial role in carrying on full lactation (51). Conversion in the mammary gland in the non-lactating woman during her first 3 days postpartum was close to those in the lactating woman, but overturned in the next 6 days, stipulating the commencement of breast recession. Colostrum is abundant in developmental factors such as insulin-like growth factor, epidermal growth factor as well as immune constituents including antibodies IgA, leucocytes, etc. (52). After colostrum, breast milk from 2-4 days after birth to 2 weeks after birth is called transitional milk and is rich in nutrients. At the end of transitional milk, or two weeks after birth, the milk is mature (table 4). Although the composition of breast milk is relatively similar, there are slight changes in composition during lactation. In Ayurveda, the *stanya Pravritti* explains starting from the nourishment of the fetus in which the food intake by the mother is divided into three i.e. nourishment to the fetus, to the mother, and breast. The essence of the food intake reaches the breast through *Stanyavaha Srotas* (fig 2).

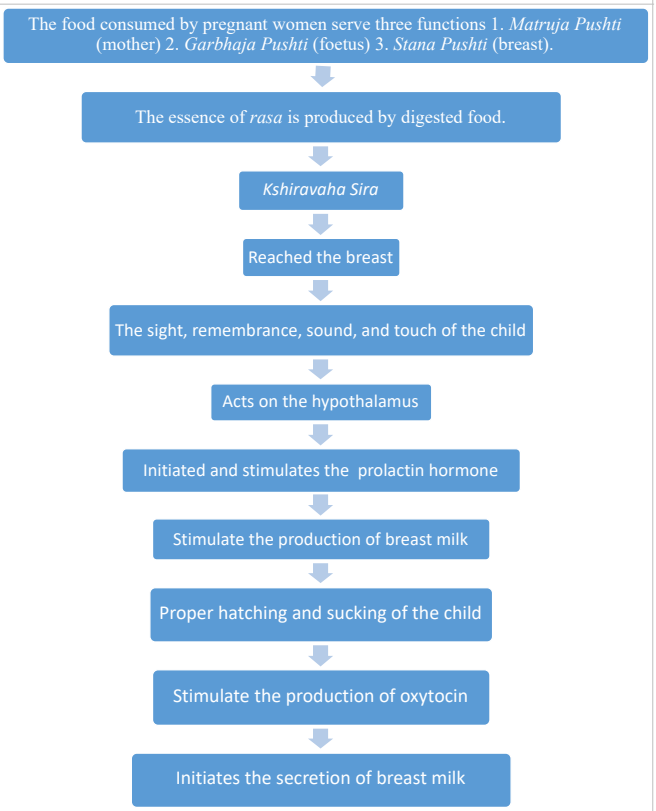
Table 4: Type of breast milk about stages of breastfeeding

Type of breastmilk	Time of secretion	Content
Colostrum	First few days after birth	Antibodies, minerals, vitamins, protein, white blood cells, epidermal growth factor. Electrolyte concentration is more in sodium, chloride, magnesium, etc.
Transitional	2 to 4 days after delivery to 14 days	Antibodies, good bacteria, and other bioactive components
Mature	From four weeks onwards	Sugar, protein, vitamins, minerals, and bioactive components
Foremilk	Beginning of feeding	Mostly water combined with nutrients
Hind milk	Just after foremilk	More in fat

Breast milk examination

In Ayurveda, the examination of breast milk was described in detail by Shusruta Acharya to assess the quality of breast milk (table 7). Organoleptic studies like color, smell, taste, and *Jala* (water) *pariksha* like *Avasadita* (sinking), *tantumat* (thread formation), *and utplavana* (floating). The quality of breast milk is differentiated by the humor known as *tridosha* i.e *Vata,*

Fig 2: Flow Diagram of Lactation (*Stanya Pravritti*) as per Sushruta Samhita.



Pitta, and *Kapha.* The color of pure breast milk was described as *Shankhavabhasham* i.e. white which is similar to the conch shell and in another color of whitish, yellowish, or greyish then the breastmilk is vitiated by the *Kapha, pitta, Vata dosha* respectively. *Jala pariksha* is a method that is performed by dropping a drop of breast milk in water and appreciating the dispersion of breast milk. The milk which is *Shuddha* (pure) will disperse immediately and that is vitiated by *Kapha* will have a white streak on dispersion and will float on the water on *Vata* vitiated. By using the sense of smell, we can differentiate the smell of breast milk having a characteristic smell that denotes *shuddha* and having other smells like that of ghee, oil, and a dead body that of afflicted *dosha*. By the sense of taste, we can get the taste that lingers on our mouth after tasting like sweetness, salty, bitter, pungent, or astringent we can decide its respective *Dosha* involved (53).

The examination of breast milk reflects the mother's pathologies and aid in diagnosing diseases in infants. It is the main cause of diseases in an infant as it is their sole nutrition. The collection of breast milk for the examination should be after three weeks i.e. mature milk. The constituent of milk in the first three weeks varies rapidly and becomes stable after that which is term as mature milk (54). The breast milk examination can also be done on vaccination visits of the child to a community center or pediatrician to get the periodic assessment result. **Color of breast milk**: In the case of *Vata Dushti Stanya*, the breast milk is *Syava* (greyish). A case of the dark brown color of breast milk in which the breast milk is dark brown on 2nd day and becomes normal in color within 72 hours. The brown-colored

Table 5: Breast milk examination in Ayurveda (53)

Parameter	Test	Observation	Result
Color	Color chart	Conch shell	*Shuddha*
		Whitish	*Kapha*
		Yellowish	*Pitta*
		Greyish	*Vata*
Dispersion in water	Dispersion method (as told in Ayurveda text)	Immediate dispersion	*Shuddha*
		White streak/ sedimentation	*Kapha*
		Float on water	*Vata*
Smell	Sense of smell	Characteristic smell	*Shuddha*
		Ghee, oil	*Kapha*
		Dead body	*Pitta*
		No characteristic smell	*Vata*
Anurasa (after taste)	Sense of taste	*Madhura* (sweet), *Lavana* (salty)	*Kapha*
		Tikta, amla, Katu	*Pitta*
		Kashaya (astringent),	*Vata*

breastmilk mostly developed from hemorrhage of the areola or nipples. There is a condition termed "rusty-pipe syndrome." It begins at birth or in early lactation, is painless, and remains anonymous except if the milk is expressed. It is thought to be due to the trauma of delicate capillaries resulting in blood leaks into the secretions (55). In *Pitta Dushta Stanya*, the breast milk is yellowish. In the first few days after delivery, the breast milk mostly is yellow due to the presence of colostrum which diminished rapidly after. The yellow color of colostrum is largely due to carotenoids lutein, all-trans β-carotene, and cis-13 β-carotene. The color index variation in colostrum accounted for 65% of β-carotene (56). There is a case in which the mother intake pills of blue-green algae before and after delivery and was developed with green breast milk indicating a probable adverse drug reaction. On intake cessation, it becomes normal in color and laboratory analysis shows no other abnormalities (57). In other cases, a drug or food metabolite, a chemical reaction, or microbial growth can lead to the coloration of breast milk (58). **Dispersion in water:** The breast milk on dispersion in water mostly is homogeneous and when it shows sedimentation signifies *kapha dushita* stanya. When the breast milk does not mix with the water i.e. float on water thus, signifies *vata dushita*. Scientific evidence for this is lacking. The factor for dissolving depends on the surface area, temperature, and amount of stirring. In *Kapha Dushta Stanya* the sedimentation may be due to *snehata* (fat content) of milk. **The smell of breast milk**: The characteristic smell and taste of breast milk show significant effects in decreasing pain in infants during blood drawing in a randomized controlled study (59). By doing a breast milk examination, the prevention of general diseases in an infant can be achieved by managing the breast milk with *Stanya Shodhana, Stanya Janana ahara, vihara,* and *aushadha* (60).

Physicochemical properties of breast milk

The different physicochemical properties of breastmilk are given in (table 6). pH is a scale of measurement for the concentration of free H+ ions resulting from the separation of acids in milk. The pH of breast milk is alkaline and can be imputed to low levels of phosphate, protein, and casein (61). In a study by Morris et al. (1986), it was found that the pH ranged from 7.0 to 7.1 by 3 months which subsequently reach 7.4 by 10 months and age [61]. The refraction of light by a solution is determined by the presence of specific molecular species and their concentrations (62). The refractive index of milk is the sum of the solvent and solute refractive indices. As a result, this parameter may be used to estimate the total solids content of milk or the quantity of water supplied to milk (63). The viscosity of milk or dairy products is defined by the flow properties. Milk and dairy products are largely influenced by composition, component concentration, pH value, temperature, and heat history. Milk viscosity is primarily determined by casein and fat (64). The presence of different electrolytes (soluble salts), non-electrolytes, and lipids contributes to milk's electrical. Sodium, potassium, and chloride ions are the primary conductors in milk and dairy products (65).

Table 6: Physicochemical properties of breastmilk (61,64,65,66)

Physicochemical	Value
Specific gravity	1.030
Osmolarity	286 mOsm/L
pH	6.35-7.35, 7.74
Water activity	0.995
Refractive index	1.3465 ± 0.0005
Freezing point	-0.523
Viscosity	1.3522
Electrical conductivity	1.46-1.89 mS/cm and decrease with a period of lactation
Surface tension	46.99=/- 1.21 × 10^{-3} N/m

Various subsequent studies have examined breast milk core mycetoma with mixed results (67,68). These differences arise due to differences in the method used in these studies, but common bacteria have been reported, including staphylococci, streptococci, lactobacilli, and propionibacterium. Milk shapes the gut microbiome and immune system in young children, with potential implications for the prevention of asthma and other diseases (69). Facultative anaerobes such as staphylococci, streptococci, enterococci, and Enterobacteriaceae are the first bacteria to colonize the infant's gut and hence act as probiotics and prebiotics (70).

Conclusion

Breast milk is the optimum nutritional supply to newborns and infants having all of the required macronutrients and micronutrients for the child. There is no perfect replacement for breast milk, alternatives are available in the form of infant formula but the full benefits of breast milk cannot be achieved by infant formula, etc. HMOs act as prebiotics for bacteria like *Bacteroides* and *Bifidobacterium* spp., thus enriching the beneficial bacteria in an infant. Breast milk examination is done by using the sense organ to get the organoleptic properties and *Jala Pariksha* as explained in Ayurveda. Physico-chemical properties are essential parameters when investigating the quality of human milk, as the physical and rheological properties are highly dependent on the chemical composition.

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