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Protected Nutrients Technology and the Impact of Feeding Protected Nutrients to Dairy Animals: A Review

[1,2]S.K. Shelke, [1]S.S. Thakur and [1]S.M. Shete
[1]Dairy Cattle Nutrition Division, National Dairy Research Institute, Karnal, Haryana 132 001, India
[2]Animal Nutrition Group, National Dairy Development Board, Anand, Gujarat 388 001, India

Corresponding Author: S.K. Shelke, Dairy Cattle Nutrition Division, National Dairy Research Institute, Karnal, Haryana 132 001, India Tel: 0091-184-2259062 Fax: 0091-184-2250042

ABSTRACT

In high producing dairy animals, especially during early lactation, the amount of energy and protein required for maintenance of body tissues and milk production often exceeds the amount of energy available from diet which results in a negative energy balance. Traditionally, cereal grains have been used to increase the energy density of diet in the ration of high producing dairy cattle, which adversely affect the dry matter intake, depresses fiber digestion and results in milk fat depression syndrome. Another viable option is to supplement protected fat in the diet of lactating cows and buffaloes which positively affect efficiency of these animals through a combination of caloric and non-caloric effects. Caloric effects are attributable to greater energy content and energetic efficiency of lipids as compared to that of carbohydrates or proteins with the overall benefit being increased milk production. The non-caloric effects include improved reproductive performance and altered fatty acid profile of milk. Whereas, the supplementation of protected protein in the diets of lactating animals increases the milk yield due to proportionate increase in the supply of amino acids to the host postruminally. Feeding protected protein in diets containing supplemental fat may alleviate the decrease in milk protein percentage associated with fat supplementation. Therefore, there is need to avoid negative energy balance during early lactation and to enhance the milk productivity with desirable composition, which will have far reaching benefits on their reproductive performance by supplementation of protected nutrients in the ration of medium and high yielding lactating animals.

Key words: Protected fat, protected protein, milk yield, reproductive performance, cattle, buffaloes

INTRODUCTION

Livestock production contributes significantly to rural economy and could be cash crops in many small holder mixed farming systems. In tropical countries, there is horizontal growth in terms of animal numbers and now needs to achieve vertical growth in terms of improving productivity, so that future demand of milk would be meet. This can be achieved, if the early lactating high yielding and genetically improved cows and buffaloes were fed according to the nutrient requirement with high energy diet. In tropical countries, the majority of livestock subsist on poor quality native grasses, crop residues and agro-industrial byproducts. Therefore, high yielding and genetically improved dairy animals has big challenge to provide the essential nutrients for meeting metabolic requirements and sustaining milk production. Milk yield and optimum reproductive performance are the most important factors in determining profitability of dairy animals and high milk production is always more important for high profitability than the low feeding cost. In early lactating cows and buffaloes, the energy intake through ration doesn't meet the requirement for higher milk production, resulting in a Negative Energy Balance (NEB), which is closely related to reproductive performance (Shelke *et al.*, 2011). Therefore, minimizing the extent and duration of NEB could be beneficial for reproduction besides getting the best productive performance from cows (Tyagi *et al.*, 2010).

Deleterious effect of NEB on productive performance of early lactating animals would be reduced by supplementation of protected fat in the ration through enhancing energy intake (Remppis *et al.*, 2011). Earlier, supplementation of protected fat was considered only as energy source during the transition period leading to improvement in reproductive performance but later it was demonstrated that the effect was also due to Fatty Acids (FA) which act as a precursor of progesterone synthesis via cholesterol and prostaglandins pathway (Staples *et al.*, 1998). Reported improvement from added fat includes improved conception rate (Salem and Bouraoui, 2008; Tyagi *et al.*, 2010), increased pregnancy rate and reduced service period (Juchem *et al.*, 2008; Silvestre *et al.*, 2011). Supplemental protected fat also improved proportion of Unsaturated Fatty Acids (USFA) and Long Chain Fatty Acids (LCFA) in milk fat (Tyagi *et al.*, 2009a; Thakur and Shelke, 2010; Shelke *et al.*, 2011).

Protected protein feeding to lactating animals leads to proportionate increase in the supply of amino acids to the host ruminant for productive/ reproductive purpose, with an overall increase in the efficiency of protein and energy utilization. A series of trials have been conducted on cattle and buffaloes on feeding of protected protein, to see its effect on growth and milk production. The average growth rate and milk production was increased by 15-25 and 10-15%, respectively (Sahoo *et al.*, 2006; Guru *et al.*, 2006; Ghorbani *et al.*, 2007; Foda *et al.*, 2009; Shelke *et al.*, 2011). Various studies showed that, formaldehyde treatment was efficient and cheaper method to protect the protein source from highly degradable cakes in the rumen (Walli, 2005; Shelke *et al.*, 2012) and its feeding significantly increased daily milk yield and protein, fat, SNF, total solids content of milk (Chatterjee and Walli, 2003; Shelke *et al.*, 2011). The technology of feeding formaldehyde treated cakes has been adopted in India by some milk producers and protected protein feed is now being manufactured exclusively by some commercial feed factories, including National Dairy Development Board, Anand, Gujarat (India).

Protected nutrient technology: Protected nutrient technology is one such approach, involving feed management through passive rumen manipulation, by which the dietary nutrients (fat and protein) are protected from hydrolysis, allowing these nutrients to bypass rumen and get digested and then absorbed from the lower tract. The protected nutrients mainly include protected fat and protein and it is also called as bypass nutrients. The other protected nutrients are protected starch, chelated minerals and vitamins. Here, we can discuss only protected fat and protein and its impact of feeding on the performance of cows and buffaloes.

Protected protein: Highly degradable proteinous oil cakes when ingested by ruminants, result in large scale ammonia production, much of it gets wasted as urea excreted through urine. Even the animal has to spent energy to convert ammonia into urea in liver. In order to increase the efficiency of protein utilization from the highly degradable cakes, these proteins need to be protected from excessive ruminal degradation and can be used as protected protein, so that the amino acids from these protein feeds are absorbed intact from the intestines of the animal for tissue protein synthesis as well as for the process of gluconeogenesis in liver (Walli, 2005).

Appropriate technological methods such as physical, chemical or combinations of both, for the proteinous feeds and their by-products can be employed before their inclusion in the rations of livestock for improving productivity. Among the various processing methods, dry roasting and extrusion cooking technologies can be used to improve the digestibility and utilization of proteinous and other feeds by ruminants. Chemical treatments have also been used for the protection of proteins and for this formaldehyde treatment has been the most effective and feasible technology for manufacture of bypass protein.

Sources of protected protein: The protein degradability data (in rumen) obtained by several groups of workers on large number of feed stuffs in India and other countries has revealed that only a few feeds are good sources of naturally occurring protected protein (having lower protein degradability), viz., maize gluten meal, cottonseed cake, fish meal, coconut cake and maize grain. Feeds like linseed cake, deoiled rice bran, soybean meal and *Leucocaenea* leaf meal are of medium protein degradability, while Mustard Cake (MC) and Groundnut Cake (GNC) are highly degradable cakes (Negi *et al.*, 1989; Walli, 2005; Shelke *et al.*, 2011). Negi *et al.* (1989) found that 50 to 70% of total N in tree forages may be present as protected protein. However, these forages contain 16-53% of total N in the form of acid detergent insoluble nitrogen. This is because of the presence of tannins, particularly the condensed tannins which bind the proteins irreversibly and if fed to animals, are capable of corroding the epithelial lining of the gastrointestinal tract. So, tree forages could be used as a source of protected protein only after devising a method for their tannin detoxification, using either some chemical, biological or biotechnological approach. While the proteins of lower protein degradability do not need any protection, highly degradable cakes like MC, GNC and sunflower seed cake need protection against attack of ruminal proteolytic enzymes, for improving their utilization by ruminants.

Methods of protein protection: Among the several methods which allow the escape of dietary protein from ruminal degradation, much of the work was carried out on heat treatment of highly degradable cakes. The problem with ‘heat treatment’ is that it may not be cost effective and moreover, it can also over-protect the protein (Sengar and Mudgal, 1982). Walli (2005) have fine-tuned the heat treatment of GNC and soybean cake and found that heating at 150°C for 2 h as the optimum temperature time combination. Walli and Sirohi (2004) observed that the roasting of soybean at 130°C for 30 min protected its protein from ruminal degradation. Formaldehyde treatment has been used by several workers in India to reduce the protein degradability of high degradable cakes and also to study the impact of its feeding on the productive performance of dairy animals (Guru *et al.*, 2006; Sahoo *et al.*, 2006; Sahoo and Walli, 2007; Shelke *et al.*, 2011). The technology for manufacture of formaldehyde treated mustard cake has been commercialized by the National Dairy Development Board, Anand and the treated cake is available in the market as a commercial product.

Performance of cows and buffaloes fed on protected protein: In India, dairy animals by and large, do not get their required dietary energy through the normal feed which the animals are offered, as the feed is mostly devoid of energy rich grains. Success achieved in terms of increase in milk yield (volume) through the feeding of protected protein in low yielders is essentially due to the supply of more energy to these energy deficient animals, through the same feed, as the extra amino acids supplied through protected protein feeding are converted to glucose in liver. Thus, essentially the feeding of protected protein increases the efficiency of protein and energy utilization within the ruminant system. Numbers of studies have been conducted on feeding of naturally occurring protected protein like cottonseed cake and maize gluten-meal to lactating ruminants, in India with most of these experiments yielding positive results (Walli, 2005; Ramchandran and Sampath, 1995; Chaturvedi and Walli, 2001). Sampath *et al.* (1997) reported significantly higher FCM yield in lactating crossbred cows fed formaldehyde treated GNC (7.8 vs. 9.4 kg day^{-1}). Chatterjee and Walli (2003) fed formaldehyde treated mustard cake to medium producing buffaloes and found a significant increase in milk yield and FCM yield of the animals. Sahoo and Walli (2005) reported that by feeding formaldehyde treated mustard cake to lactating goats increased the milk yield significantly from 1306 g day^{-1} in control group to 1439 g day^{-1} in formaldehyde treated group. Walli and Sirohi (2004) also reported 15% increase in milk yield on feeding of formaldehyde protected mustard cake to crossbred cows. Garg *et al.* (2003) while comparing naturally protected protein (30% UDP) and processed (formaldehyde treated) sunflower seed meal supplement (optimal-bypass with 75% UDP) in crossbred cows, found a significant increase in milk yield, milk fat and milk protein percent. Yadav and Chaudhary (2004) reported significantly increased milk yield and FCM yield in medium producing cows on feeding formaldehyde treated GNC. Similarly, Shelke *et al.* (2011) concluded that supplementation of protected nutrients (protected fat at 2.5% of DM intake and formaldehyde protected cakes) to lactating Murrah buffaloes significantly increased milk yield and milk fat.

Protected fat

Sources of fatty acids: The main sources of Short Chain Fatty Acids (SCFA) are cottonseed oil and palm oil. All the sources of fat contain adequate quantity of Long Chain Fatty Acids (LCFA). The main sources of linolenic acid (C18:3n3) are flaxseed, hemp, canola, soybean, nuts and dark green forages. Ryegrass silage contains as much as 60% of linolenic acid as a percentage of total fatty acids which would encourage high forage systems to increase dietary linolenic acid content. Omega-3 fatty acids are found also in cold water and salt water fish (salmon, trout, mackerel, sardines). The main sources of linoleic acid (C18:2n6) are sunflower seed, safflower, hemp, soybean, nuts, pumpkin seeds, sesame seeds and flaxseed. Gamma-linolenic acid (C18:3n6) is found in evening primrose oil, grape seeds and borage. Dihomo-gamma-linolenic acid (C20:3n6) is found in maternal milk while arachidonic acid (C20:4n6) occurs mainly in meat and animal products. Oleic acid (C18:1) is found in olive, almond, avocado, peanut, pecan, cashew, macadamia nut and butter. Omega 7 in the form of palmitoleic acid (C16:1) is found in tropical oils (coconut, palm). Composition in C18 fatty acids of some edible vegetable oils is presented in Table 1.

Methods of fat protection: The protected fat can be obtained by various methods such as encapsulation technique and calcium salt formation of fatty acids. Calcium salts of fatty acids were produced at NDRI, Karnal by double decomposition method from edible oils and non-edible oils and other products such as acid oil (a byproduct of vegetable oil refining). The calcium salts were prepared by a method described below:

Soybean oil acid oil was heated in a metal container; an aqueous solution of sodium hydroxide was added and again heated to cause saponification, sodium salts so formed were dissolved in excess water. Calcium chloride dissolved in water was then added slowly to the water soluble sodium soaps with stirring causing immediate precipitation of calcium salts. Excess water was removed by squeezing the soaps through cheese cloth. The soap was allowed to air dry and then lumps were

Table 1: Comparison of major fatty acids in some edible oils (w/w % fatty acids)

Oil	C18:0	C18:1	C18:2	C183
Groundnut	2	47	32	0
Canola	2	64	19	8
Safflower	2	12	77	0
Cotton seed	25	21	50	0
Linseed	4	19	14	58
Corn	2	25	60	1
Fish meal	2	25	4	45
Olive	2	76	8	0
Palm	4	39	10	1
Soybean	4	24	53	7
Sunflower	5	20	60	0
Sesame	2	42	45	0
Tallow	15	41	8	1

broken before being mixed with other concentrate ingredients (Mishra *et al.*, 2004). Sugumar and Balakrishnan (2008) also concluded that calcium soaps of sunflower acid oil was selected as the potential protected fat to be used as concentrated energy source in the rations of dairy cows.

Calcium salts are being manufactured commercially from palm fatty acids by single stage fusion technique which is more economically viable and environment friendly. These types of protected fats are commercially available in the market.

Productive performance of cows and buffaloes fed on protected fat: Adding protected fat to dairy rations can positively affect efficiency of dairy cows through a combination of caloric and non-caloric effects. Caloric effects are attributable to higher energy content and energetic efficiency of lipids as compared to carbohydrates or proteins with the overall benefit being increased milk production and the persistency of lactation. The non-caloric effects include improved reproductive performance and altered fatty acid profile of milk. Feeding Ca soaps of fatty acids to high producing lactating cows resulted in higher milk and milk fat production (Sklan *et al.*, 1991). The higher milk and fat production observed in cows fed Ca salts of fatty acids during early lactation may both be due to higher energy intake, more efficient use of fat and by enhanced tissue mobilization before peak production. Wu *et al.* (1993) supplemented tallow, Ca salts of palm oil fatty acids and prilled fat at 2.5% to mid-lactation cows. Added fat increased milk yield over control but source of fat did not affect milk yield.

Fahey *et al.* (2002) observed significant improvement in milk production on dietary supplementation of Calcium salts of fatty acids (Megalac) and Calcium salts of methionine hydroxy analogue (Megalac plus) as compared to that of control. Similarly, Schroeder *et al.* (2002) demonstrated an increase in FCM yield from 23.4 to 26.3 kg day^{-1} on supplementing partially hydrogenated oil to lactating Holsteins cows under grazing conditions. McNamara *et al.* (2003) supplemented Holstein Friesian cows with two protected fat supplements: (1) Megalac Plus (0.4 kg day^{-1}) containing Ca salts of methionine hydroxyl analogue and (2) Megapro gold (1.5 kg day^{-1}) containing Ca salts of palm fatty acids, extracted rapeseed meal and whey permeate and reported higher milk yield in both the groups over that of control. Supplementation of protected fat rich in PUFA to goats not only increased the milk yield but the effects persisted even after the supplement was withdrawn (Sampelayo *et al.*, 2004). In a study on Murrah buffaloes, Thakur and Shelke (2010) reported an improvement of 12.4% in milk yield of buffaloes fed 4% Ca salts of fatty acids. Tyagi *et al.* (2009a) reported that protected fat supplementation at 2.5% of DMI for 90 days postpartum increased the milk production and its persistency up to 120 days after cessation of protected fat feeding.

An increase in the consumption of milk and butter by humans has been widely reported to be associated with a rise in occurrence of cardiovascular diseases. This increase has been linked to the effects of saturated fats, in particular Myristic (C14:0) and Palmitic (C16:0) acids, in elevating the plasma concentration of low density lipoprotein cholesterol, which is recognized as a major risk factor for coronary heart disease. In addition to the impact of dairy foods on human nutrition, there is also the issue of physical and sensory properties of milk fat. Milk fat contains approximately 10 to 12% C14:0, 25-30% C16:0 and 8-11% stearic acid (C18:0); classified as a hard fat and that is why milk fat possesses poor spreadability. Therefore, it may be desirable to increase the C18:0 plus C18:1:C16:0 ratios in milk fat as well as increase the proportion of short chain fatty acids (C4:0 to C10:0). It is now recognized that omega 3 FA are essential for normal growth and important for brain and vision development and immunity in infants; these FA may also play a vital role in prevention and treatment of cardiovascular diseases (Williams, 2000; Ramezani *et al.*, 2008). Ashes *et al.* (1992) observed on feeding protected Canola seed (emulsified and encapsulated in a matrix of aldehyde treated protein), a significantly reduced proportion of saturated fatty acids C16:0, C14:0 and C12:0 in milk fat; there were corresponding increases in the proportions of C18:0, C18:1, C18:2 and C18:3. An increase of 54% in the yield of C18 monounsaturated and polyunsaturated FA was recorded. Jenkins (1998) and Delbecchi *et al.* (2001) reported similar results on supplementing protected Canola oil. Similarly, Fahey *et al.* (2002) reported a low concentration of short chain fatty acids (C4 to C14:1) and higher concentrations of long chain fatty acids (C18:1 and C18:2) than control on feeding Ca salts of FA and methionine hydroxyl analogue. Mishra *et al.* (2004) fed Ca salts of mustard oil FA at 4% of DM to lactating crossbred cows and reported decreased proportion of medium chain FA but increased proportion of $C_{18:1}$, $C_{18:2}$ and $C_{18:3}$ in milk fat. Tyagi *et al.* (2009a) reported that supplementation of protected fat at 2.5% DM significantly increased the proportion of unsaturated FA and LCFA in milk fat of cross bred cows. Shelke and Thakur (2011) observed that total USFA content of milk was increased by 35.73% (41.78 vs. 30.78%) and SFA decreased by 18.70% (52.91 vs. 65.08%) in protected fat and protein supplemented buffaloes.

Reproductive performance of cows and buffaloes fed on protected fat: Good reproductive performance of dairy animals is important as far as the economic returns to the milk producers is concerned and it is related to the good nutrition of the animals. From previous studies, it was observed that there is direct correlation of the dietary energy intake and body condition to the reproductive performance of animals. Dietary energy intake is low and poor body condition can negatively affect reproductive performance of dairy animals, which results in loss of economic returns to the milk producers. Nebel and McGilliard (1993) also concluded that there was correlation between level of milk yield and reproductive traits as well as higher milk yield is associated phenotypically and genetically with reduced reproductive performance in lactating cows. Reproductive disorders are one of the major factors reducing the milk and affecting the production potential of dairy animals (Ali *et al.*, 1999; Taraphder *et al.*, 2007). Therefore, it will be desirable to incorporate the recent innovations i.e., protected protein and fat in the ration of lactating cows and buffaloes in their early lactation period to avoid the NEB and to enhance the milk productivity with desirable composition which may have far reaching positive influence on their reproductive performance.

It has been reported that high milk yield during early lactation retards development of ovarian follicles, prolonging the postpartum interval to first ovulation, therefore, high yield is antagonistic to the expression of estrus and is associated with reduced conception rate (Goff and Horst, 1997). Protected fat supplementation to the dairy animals has been shown to positively affect the reproductive functions at several important tissues, including the hypothalamus, anterior pituitary, ovary and uterus. Types of fatty acids content of the source fat is related to the reproductive performance of animals, as they primarily act on the target tissues. Fat supplementation is a common practice in dairy cattle production, primarily to increase the energy density of the diet. Associated positive and negative effects on reproduction have been reported (Grummer and Carroll, 1991; Ghoreishi *et al*., 2007; Tyagi *et al*., 2009b).

Fat supplementation prepartum: Pregnancy rate was increased by 19% on supplementation of safflower seeds at 0.68 kg day^{-1} (4.7% fat in the diet) with similar energy and protein content to the late-gestation heifers (Lammoglia *et al*., 1996). Bellows *et al*. (2001) also reported supplementation of safflower seeds, soybeans, or sunflower seeds (4.7, 3.8 and 5.1% fat in diet, respectively) for the last 65 day before calving to first-calf heifers increased the pregnancy rates by 94, 90 and 91%, respectively compared to controls (79%) receiving diets with equivalent energy (2.4% fat). However, in another study it was observed that supplementation of sunflower seeds (6.5% fat in diet) during last 68 day before calving did not improve subsequent pregnancy rate compared to control diet (2.2% fat). The contradictory results of both studies were due to mainly forage availability. In second study, 71% more forage availability and greater nutrient quality was reported. The samples of forages were analyzed for protein and fat; protein ranged from 18 to 34% and fat was from 2 to 3.5% depending on forage species. The major fatty acid in the forages was linolenic. This would be the reason for no improvement observed in pregnancy rate of second study, higher quality of forage would meet the nutritional-reproduction response in these heifers and this tended to mask any carryover effect resulting from supplemental fat fed in the gestation diet. Tyagi *et al*. (2009b) reported that protected fat supplementation at 2.5% of DMI 30 days pre partum, increased the calf birth weight and decreased the incidence of retention of foetal membranes.

Fat supplementation postpartum: Freshly calved cows were supplemented with rice bran (5.2% fat in the diet) up to 50 day showed higher pregnancy rate than that of cows receiving a control diet containing 3% fat in the diet (De Fries *et al*., 1998). Wehrman *et al*. (1991) reported that 18% increased in cycling of cows by supplementation with 1.36 kg of whole cottonseed (5.5% fat in diet) 30 day before the breeding season compared to a control diet without added fat. Supplementation of 75% of whole cottonseed to heifers before breeding improved estrus activities and conception rate (Barje *et al*., 2007).

Feeding bypass fat at the rate of 100-150 g day^{-1} to high yielders during the transition period (10 days before and 90 days after calving) could help improving their milk production and reproduction efficiency (Garg *et al*., 2008). National Dairy Development Board of India has standardized the production process of bypass fat supplement on a pilot scale, conducted feeding trials and different methods of manufacturing and economics thereof, for commercial production. Now, bypass fat supplement using palm fatty acid distillate would be produced commercially by setting up a bypass fat plant. The effects of rumen protected fat supplementation on reproductive performance of lactating dairy cows is summarized in Table 2.

Table 2: Effects of supplemental fat on reproductive performance of lactating dairy cows

Reference	Treatment	Treatment period (days)	Production of milk or FCM (kg day^{-1})	Conception rate at first AI	Overall pregnancy rate	Overall conception rate	Days open	AI Per conc.
				------------	(%)	------------		
Schingoethe and Casper (1991)	No oil seeds	28-112	31.40	-	-	-	126	2.15
	Oilseeds		30.90	-	-	-	136	2.38
Lucy *et al*. (1992)	0% Ca, LCFA	15-98	32.50	44.00	45.0	-	-	1.20
	3% Ca, LCFA		34.80**	12.00	10.0**	-	-	0.50
Salfer *et al*. (1995)	0% PHT	1-151	32.00	-	94.0	-	88	1.36
	2% PHT		33.20	-	94.0	-	95	1.25
Son *et al*. (1996)	0% Tallow	15-84	30.70	33.00	44.0	-	-	-
	3% Tallow		31.40	44.00	62.0*	-	-	-
Filley *et al*. (2000)	0% Ca, PFA	50-150	-	-	68.4	-	-	-
	3 % Ca, PFA		-	-	72.2	-	-	-
McNamara	0% fat	10 - 100	28.90	35.00	82.0	-	77	-
	3% Megalac +		30.40*	48.00	84.0	-	77	-
	1.5 kg day^{-1} Megapro Gold		28.90	54.00*	91.0	-	78	-
Tyagi *et al*. (2009b)	0% Ca, SFA	45 days Prepartum -90	17.57	55.56	-	77.78	-	3.71
	2.5 % Ca, SFA	Postpartum	18.65*	50.00	-	80.00	-	2.71

FCM: Fat corrected milk, AI: Artificial insemination, LCFA: Long chain fatty acids, PFA: Polyunsaturated fatty acids, SFA: Saturated fatty acids, PHT: Partially hydrogenated tallow, *$p<0.05$, **$p<0.01$

CONCLUSION

In developing countries like India, supplementation of protected fat and protein is beneficial to medium and high yielding cows and buffaloes but the cost effectiveness of the same needs to be kept in mind. As about the feeding of protected protein, the results of some farm studies and field studies have indicated the usefulness and cost effectiveness of its feeding to cows and buffaloes yielding around 5-8 L of milk.

In addition, milk fat yield and percentage of unsaturated fatty acids in milk fat was increased, resulting improve nutritive value of milk from a human health point of view.

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