

**Climate Resilient Livestock Feeding Systems for Global Food Security**

Invited Papers



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## Recent Concepts in Mineral Nutrition

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In India, dairy animals mainly thrive on crop residues, supplemented with limited quantity of green fodder, and cattle feed and/or concentrate feed ingredients. Crop residues based basal diet is poor in essential minerals. It also contains several anti-nutritional factors like silicates, oxalates, gossypol and phytates, which further inhibit their utilization. Under such feeding regime, it has been found that a large number of dairy cattle and buffaloes suffer from reproductive disorders, which are primarily due to micro-nutrient deficiencies. Mineral deficiencies may result in delayed onset of estrus, repeat breeding and/or infertility (Underwood and Suttle, 2010). Impaired reproductive performance results in an increased inter-calving period and reduces the productive life of animals, causing great economic loss to the milk producers, which is often unrealized. As animals do not synthesize minerals, their supplementation through mineral mixture is of paramount importance.

In this paper, an attempt has been made to cover some important aspects of mineral nutrition of dairy animals such as functions of minerals, optimum mineral requirements, mineral requirements during heat stress, mineral interrelationships, mineral status of feeds and fodder, manufacturing of area specific mineral mixtures (ASMM), role of chelated minerals in improving reproduction efficiency, immune status and reducing the incidences of sub-clinical mastitis. Total mineral

mixture production in the country against the tentative requirement, advantages and impact of feeding ASMM to dairy animals, is also covered in the manuscript.

### Minerals availability and requirement for dairy animals

Problems of mineral deficiencies and metabolic disorders in all categories of dairy animals have been reported by many scientists due to lower content and low bio-availability of some essential macro and micro-minerals from different feedstuffs (McDowell, 1992; Sharma et al., 2002; Udar et al., 2003; Garg et al., 2011; Gowda et al., 2005, Gowda et al., 2011). It includes rickets, osteo-malacia, pica, bone, teeth and hoof abnormalities, sway back (neonatal ataxia), de-pigmentation of skin, parakeratosis, anemia, still birth, goitre, poor growth rate, reduced milk yield and fertility. More than 90% of minerals and vitamins deficiency exists at sub-clinical level in livestock (Underwood and Suttle, 2010). Even small imbalances or deficiencies can develop into reproductive, health and milk production related problems (Prasad and Gowda, 2005). Supplementation of minerals helps in efficient utilization of absorbed nutrients and many other ways, for improving growth, milk production and reproduction efficiency. Mineral requirements suggested by National Research Council (NRC, 2001) for lactating dairy cows are given in Table 1.

The concentrations of mineral elements in plants are dependent on the interaction of a number of factors including soil pH, plant species, stage of maturity, yield and climate (Reid and Horvath, 1980). The shedding of the seeds is normally responsible for losses of many minerals so the material remaining e.g. the straw is a poor source. Sometimes, there is a difference of minerals content in feeds and

fodders between hilly and plain region (Gowda et al., 2001; Ramana et al., 2001).

As mineral deficiencies in the ration of animals vary with agro-climatic conditions, mapping of such deficiencies is essential across different zones in various states to develop ASMM for supplementing the ration of animals in effective and economical manner.

Table 1: Mineral requirements for buffaloes and cows

Requirements	Buffaloes		Cows	
	Calcium	Phosphorus	Calcium	Phosphorus
Maintenance (g)	18	13	16	11
Milk yield (g/kg)	4.65	2.98	3.21	1.98
Mg and S : 0.20% of DMI		Copper : 10 ppm		Manganese : 40 ppm
Na : 0.18% of DM Intake		Iron : 50 ppm		Cobalt : 0.50 ppm
K : 0.90% of DM Intake		Zinc : 80 ppm		Selenium : 0.30 ppm
Cl : 0.25% of DM intake		Iodine : 0.60 ppm		Chromium: 0.50ppm

### Mineral deficiency affecting animal productivity

There has not been significant increase in the productivity of indigenous cows, crossbreds and buffaloes over the years. The average daily milk production data of various categories of animals at 6.97kg for crossbreds, 2.27kg for indigenous cattle and 4.71kg for buffaloes (BAHS, 2012), suggests that the productivity of animals is far below than their genetic potential. Even the % in-milk animals of the total breedable animals, is also very low. As per the BAHS data (2012), about 45% animals of the total breedable animals are dry. Amongst others, this could be attributed to deficiency of critical minerals in the ration (Garg and Bhandari, 2011). Low productivity of animals with higher genetic potential can mainly be attributed to imbalanced feeding. Data generated (n=40,802) from different states through ration balancing programme (RBP) using Information Network for Animal Productivity and Health (INAPH) indicate that while energy and protein are in excess in the ration, minerals are deficient in the ration of

more than 85% of milch animals. Thus, even with the available feed resources, there is ample scope to improve productivity by way of providing deficient minerals in the ration of dairy animals.

### Role of macro-minerals for milk production

Calcium (Ca), phosphorus (P) and magnesium (Mg) are often associated with bone development and growth, but these minerals also serve other vital functions. These include growth, energy utilization, membrane structure, muscle contraction and hormone secretion. The Ca and P are two macro-minerals required by the dairy animals and its requirement increases with increased milk production (McDowell, 1992). Milk contains 0.12% Ca and 0.10%P per kg and colostrum still rich in Ca (0.14%). Considering its bio-availability, dairy cows required minimum 3.21 g Ca and 1.98 g P per kg milk yield (NRC, 2001). The requirements of Ca and P are 4.65 g and 2.88 g per kg milk yield, respectively for buffaloes (Kearl, 1982). The requirement of Ca per kg milk yield in buffaloes is higher than

cows. Buffalo milk contain high fat, therefore, high fat diet is required for buffalo which results in fecal Ca losses through formation of insoluble Ca soaps and thus, increases dietary requirements. Milk production is reduced in prolonged Ca and P deficiency because animals are not able to produce milk low in Ca and P contents.

For proper utilization of Ca and P in animal system, vitamin D<sub>3</sub> supplementation and ratio of Ca to P in total diet is also important. While animal can tolerate ratios of between 1:1 and 7:1, excessive Ca may decrease the absorption of other minerals. Therefore, it is recommended to maintain a ratio of Ca to P between 1.5:1 and 3:1. Deficiencies in Ca and P or an imbalance in the Ca to P ratio can result in decreased fertility, milk production, growth and feed efficiency; as well as an increased incidence of metabolic disorders such as milk fever.

Potassium (K), sodium (Na) and chlorine (Cl) are important in water and acid-base balance, muscle contraction, nerve signal transmission and enzymatic reactions. A deficiency of these minerals can result in decreased feed intake, weight gain and milk production. Sulphur (S) is required by ruminants for the synthesis of the S-containing amino acids, cysteine, methionine and the B-complex vitamins, thiamine (B<sub>1</sub>) and biotin. Sulphur is also used in the detoxification of poisonous compounds like those potentially found in most sorghum forages. A deficiency of S can result in reduced feed intake, weight gain and digestibility, and animals may be more susceptible to acidosis.

#### **Calcium status and its impact on milk fever**

A dynamic system involving Ca, P and vitamin D<sub>3</sub> exists to maintain a relatively stable concentration of Ca in the blood. Ca and P are stored in bones and mobilized into the

circulatory system when dietary intake of the two minerals is adequate. Blood Ca level is not a good indicator of a dietary Ca deficiency because blood Ca is reflective of both Ca intake and Ca mobilization from the bones. A common method of minimizing the risk of milk fever is to reduce Ca intake by animals for two weeks before calving. This ensures that the Ca mobilization system is functioning properly before lactation. After calving, dietary Ca is increased to meet the requirement of the lactating animals. Milk fever and sub-clinical hypocalcaemia are the most important macro-mineral disorders that affect transition dairy animals. For prevention, animals need to be supplemented adequate amount of Ca, P and vitamin D<sub>3</sub>. It has been reported that milk fever results in reduced fertility in dairy animals due to its effect on uterine muscle function, slower uterine involution (Borsberry and Dobson, 1989) and reduced blood flow to the ovaries (Jonsson and Daniel, 1997).

Curtis et al. (1983) reported that animals that had suffered clinical milk fever were eight times more likely to develop mastitis than normal animals. The reasons for this phenomenon are a reduction in smooth muscle function at teat sphincter and hence an easy route for infection after milking and an exacerbated suppression of immunity in milk fever animals as compared to normal animals (Goff, 2003). Animals suffering from milk fever and mastitis, their life time production affected at chronic level, reducing overall profitability. One of the best strategies often proposed for milk fever prevention is the restriction of Ca intake prior to calving. This has the effect of making sure that parathyroid hormone and active form of vitamin D<sub>3</sub> are in higher concentrations in circulation on the day of parturition when Ca secretion in colostrum (0.14% Ca) increases suddenly. Milk production and reproduction efficiency of animals can be improved by prevention of milk fever.

### Role of macro-minerals in dietary cation-anion balance (DCAB)

Another method of preventing and controlling milk fever is balancing dry animal rations for anions and cations. Na and K are the cations and chloride (Cl) and S are the anions of interest in formulating anionic diets. The DCAB equation most often used to determine milliequivalents per 100 g of dry matter is:  $mEq/100g = mEq (Na + K) - mEq (Cl + S)$ . Based on current research, the range that achieves the lowest incidence of milk fever is a DCAB of -10 to -15 mEq/100g dry matter (DM). Achieving a DCAB of -10 to -15 mEq/100g requires adjustments in the major mineral levels that are quite different than what is normally supplemented for regular dry animal rations. Table 2 lists recommended

mineral levels for both regular and anionic rations.

Balancing rations for anions affects the animal's acid-base status, raising the amount of Ca available in the blood. Urine acidity is affected by these changes in the animal's acid-base status (Table 3). Checking urine pH can help milk producers and veterinarians to monitor the effectiveness of an anionic ration.

Feeding a combination of different anionic salts is necessary for achieving the desired DCAB. The most commonly fed anionic salts are given in Table 4. Special attention should be given to the degree of hydration of specific salts in formulating rations as well as their costs and availability.

Table 2. Mineral composition for advanced pregnant dry animals

Mineral	Regular (%)	Anionic (%)
Calcium	0.45-0.55	1.40-1.60
Phosphorus	0.30-0.35	0.35-0.40
Magnesium	0.22-0.24	0.28-0.32
Potassium	0.80-1.00	0.80-1.00
Sulphur	0.17-0.19	0.35-0.40
Chlorine	0.20-0.24	0.70-0.80
Sodium	0.10-0.12	0.10-0.12

Table 3: Effect of DCAB on urinary pH of animals

Ration DCAB	Before calving (2 weeks)		At calving
	Urine pH	Acid-base status	Ca status
Positive (>0 mEq/100g)	8.0 to 7.0	Alkalosis	Low blood Ca
Negative (<0 mEq/100g)	6.5 to 5.5	Mild metabolic acidosis	Normal blood Ca

Table 4. Chemical composition of commonly available anionic salts

Mineral salt	Chemical formula	N	Ca	Mg	S	Cl	DM %
Ammonium sulphate	$(NH_4)_2SO_4$	21.2	-	-	24.3	-	100
Calcium sulphate	$CaSO_4 \cdot 2H_2O$	-	23.3	-	18.6	-	79.1
Magnesium sulphate	$MgSO_4 \cdot 7H_2O$	-	-	9.9	13.0	-	48.8
Ammonium chloride	$NH_4Cl$	26.2	-	-	-	63.3	100
Calcium chloride	$CaCl_2 \cdot H_2O$	-	27.3	-	-	48.2	75.5
Magnesium chloride	$MgCl_2 \cdot 6H_2O$	-	-	12.0	-	34.9	46.8

Before incorporating DCAB into a dry animal's ration, there are several factors to be considered for better results. Some of the anionic salts are very unpalatable which can depress intakes significantly in conventional feeding systems. Reduced dry matter intakes as a result of feeding anionic salts can lead to the development of other metabolic disorders. Animals should receive the anionic diet at least 3-4 weeks prior to the expected calving (Beede, 2005). Buffers must not be used in anionic salt rations because they will counter the effect of DCAB.

### Role of micro-minerals

Trace minerals are needed for optimal growth and performance, and play an important role in immune function. The trace minerals that are commonly supplemented to dairy animals include copper (Cu), zinc (Zn), manganese (Mn), iodine (I), cobalt (Co) and chromium (Underwood and Suttle, 2010). Trace minerals are required at very low concentrations, making deficiencies difficult to recognize. Deficiencies of the trace minerals can result in decreased feed intake and weight gain, reduced fertility and libido, retained placentas, abortions and stillbirths, low birth

weights and poor calf performance.

### Immunity and antioxidant functions of trace minerals

Minerals also play important roles as antioxidants in cows and buffaloes. The free radicals are produced in normal metabolism process of the body. Free radicals are chemicals that have one or more unpaired electrons in their outer orbit. There are different kinds of free radicals that comprise oxygen radicals and cellular radicals. The oxygen radicals are the most important and consist of superoxide, hydroxyl, hydrogen peroxide and nitric oxide radicals. It has been estimated that as much as 2% of the oxygen consumed by mitochondria is used to form superoxide radicals.

In normal conditions, the generated free radicals are extinguished by the antioxidant protective system, which includes protective compounds and enzymatic systems. Tissue defense mechanisms against free radical damage include glutathione peroxidase (Se), catalase (Fe) and superoxide dismutase (Cu, Zn and Mn) as the major mineral antioxidant sources (Table 5).

Table 5. Minerals element and metallo-enzymes

Mineral	Metallo-enzymes
Cu	Tyrosinase, lysyl oxidase, Cu-superoxide dismutase, dopamine- hydroxylase
Zn	Carbonic anhydrase, alcohol dehydrogenase, Zn-superoxide dismutase, tibial collagenase
Mn	Pyruvate carboxylase, arginase, Mn-superoxide dismutase
Mo	Xanthine oxidase, liver aldehyde oxidase
Fe	Catalase, peroxidase, cytochrome-A, B & C, lactoperoxidase, verdoperoxidase, succinate dehydrogenase
Se	Glutathione peroxidase

### Role of trace minerals in improving reproduction efficiency

Trace minerals such as Zn, Cu and Mn have been shown to improve reproductive function when supplemented appropriately.

Zn is essential to all animals and plays significant roles in the metabolic activity of dairy animals. It is largely involved in nucleic acid metabolism, protein synthesis and carbohydrate metabolism (Chew, 2000). Zn

and vitamin A is important in maintenance of health and integrity of epithelial tissue, such as skin (teats) and mammary tissue, due to its role in cell division and protein synthesis. An additional mode of action for Zn reducing somatic cell counts (SCC) is related to Zn's role in keratin formation. Zn is required for the incorporation of cysteine into keratin. The keratin lining of the teat canal entraps bacteria and prevents their upward movement into the mammary gland (Singh and Pachauri, 2001). Kellogg (1990) reported that chelated Zn decreased SCC by 22-50%, depending on the dose of Zn used, and increased milk production. Supplementation of Zn in chelate form decreased the rate of intra-mammary infections in dairy animals (Spears, 1996). Supplementation of optimum protein and Zn increases ovulation and has a role in spermatogenesis in male animals (Campbell et al., 1999).

Likewise, adequate Mn and Cu supply in the ration is also necessary to keep the animal in normal cyclicity. Cu is required for optimum performance of cellular structures known as mitochondria for the metabolism and transfer of energy in the cells and tissues. Mn also involved in a large number of enzymatic processes in the body. It is believed that Mn is involved in heat expression. Mn deficiency in animals result in abnormal growth, increased fat deposition, reproductive problems and reduced milk production. Iodine is required for normal foetal development and its deficiency may result into still-birth, abortion, re-absorption of foetus or birth of weak and hairless young ones. Male fertility is also affected and may be resulted into decline in the libido and deterioration of semen quality. Each of the following systems in the body utilizes minerals. Research shows that imbalances or deficiencies in mineral nutrition can affect these systems: Immune system: Cu, Zn, chromium

(Cr), iron (Fe) and selenium (Se), Energy production: Mg, P and Mn, Hormone system: Fe, Mn, Zn, Cu, Mg and K, Vitamin production: Co and S, Blood production: Cu and Fe, Enzyme systems: Zn, Cu, K, Mn, Mg, Fe, Ca and molybdenum (Mo) & Skeletal system: Ca, Mg, Zn, Mn) and P.

#### Mineral requirements during heat stress

Heat stress reduces voluntary feed intake which leads to corresponding decline in heat generated by ruminal fermentation and body metabolism, and thus, help in the maintenance of heat balance. Because of reduced feed intake during heat stress, intake of essential minerals is also reduced. Decrease in intake of most minerals is followed by decrease in retention of these minerals.

Sweating and panting provide evaporative cooling, one of the most effective ways to lower the animal's body temperature. The cooling mechanisms change the animal's mineral requirement, so we must change the mineral content of the diet to compensate for the effects of heat stress. K, Na and Mg are the major minerals, which need to be supplemented at higher level during the period of heat stress. Animal's large surface area actually allows for significant sweating during heat stress and K loss via sweat can be substantial. K is also the primary mineral secreted in milk, which contains 0.15% K. Together, these K losses greatly increase the K requirement of heat stressed cows (from 0.9% to 1.5%), especially high producing cows. Elevated dietary K interferes with the absorption of Mg from the animal's digestive system. When increasing K during heat stress, it is important to increase Mg to overcome this negative interaction.

Panting during heat stress causes significant saliva loss. Saliva is rich in sodium bicarbonate. Feeding sodium bicarbonate replaces both the Na and bicarbonate lost in this

drol. Sodium bicarbonate buffers help maintain proper rumen pH for optimum fermentation. Sodium requirement almost doubles during periods of heat stress. Table 2

contains NRC (2001) mineral recommendations for lactating animals and suggested mineral levels in feed during heat stress/hot humid climate.

Table 6. Recommendations for selected minerals (% DM)

Minerals	As per NRC (2001)	During heat stress
K	0.90	1.20 to 1.50
Na	0.18	0.40 to 0.60
Mg	0.20	0.30 to 0.35

**Mineral interactions in animal system**

Inorganic trace minerals chemically react among themselves. High levels of one inorganic trace mineral can decrease the availability and use of another. Known mineral interactions include Cu-Mo, S-Se, Ca-P, Ca-Zn, Ca-Mn, Fe-Mn and K-Mg (Fig. 1). Trace mineral absorption rates in older animals typically are lower than in younger animals.

the Mo content. Furthermore, in areas with high soil Mo, Cu levels five times higher than normal may be required to overcome Cu deficiency. In contrast, toxic levels of S and Mo in the diet can be counteracted by the addition of Cu.

Organic and inorganic trace minerals have a fundamental difference. The inorganic trace mineral, a finely ground salt, is complexed within an organic matrix – either by nature or man – to produce an organic trace mineral. The organic trace mineral can have two times or greater bio-availability as compared to the inorganic trace mineral. Commercially produced organic trace minerals are more bio-available and more highly absorbed in the small intestine. If the trace minerals are not absorbed here, their nutritional benefits are lost. For the most part, unused, unabsorbed trace minerals are excreted.

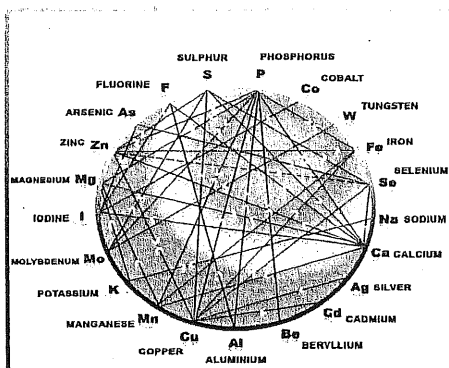


Figure 1: Interrelationships of minerals in animal system

An important example of maintaining adequate balance between minerals is the relationship between Cu, Mo, Fe and S. These minerals form complexes with one another in the body, reducing the amount of Cu absorbed by the animal. Addition of Mo, Fe or S to diets that are already deficient in Cu is of particular concern because these minerals will further exacerbate Cu deficiency. It is recommended that the level of Cu be at least twice as high as

Commercially produced organic trace minerals are protected from interactions with antagonists and vitamins. Antagonists interact chemically with inorganic trace minerals. The interactions bind the inorganic trace minerals, leaving them unavailable to the animal. Antagonists include sulphates, oxides, fibre and many of the inorganic trace minerals themselves. They include the high S content of dried distiller's grain, iron contamination from dirt in corn silage and other fermented feeds, and the minerals in hard water, notably S and Ca.



Minerals in forage/grasses consist of three fractions: i) highly soluble and rapidly released; ii) slow release as cell walls and protein components are degraded and, iii) no release. The major portion of Cu in forages/grasses appears to be contained in the rapid release fraction; however, Zn has been shown to have the lowest percentage release compared to Ca, Mg, K, P and Cu. Increasing the NDF content of the diet has also been shown to decrease apparent absorption of Mn, Zn, Fe and Cu.

### Mineral status of commonly used feedstuffs in different regions in India

Livestock feeding in India is traditional in nature and mostly depends upon the locally available feed resources. These feed ingredients are mostly agro-industrial by-products; vary widely in their minerals and vitamins composition. Availability of green fodder is seasonal and depends upon the rainfall and irrigation facility. Hence, dry roughages like straws of rice, wheat, sorghum, *bajra*, maize, groundnut, soybean and local grasses etc., form bulk of the ruminant animals' diet. Straws and stovers harvested after maturity contain certain minerals and vitamins at very low levels, but contain excess of silicates, phytates, oxalates, gossypol and tannins, which may interfere in the utilization of

minerals and other nutrients (Garg and Bhanderi, 2005). Supplementation of ASMM is not practiced in most parts of the country. Hence, animals depend for their minerals requirement on feeds and fodder offered to them (Table 7). Most of the feed ingredients available for feeding livestock are deficient in one or other minerals, as shown in Table 8. Dairy animals in different agro-climatic regions need to be supplemented with minerals after considering levels of macro and micro-minerals in the feeds and fodders ingested by the animals (Samanta and Samanta, 2002; Garg et al., 2005; Gowda et al., 2004).

### Status of macro-minerals in feeds and fodder

The Ca content is low in straws and stovers (0.10-0.30%), except groundnut straw (1.20-1.67%). However, leguminous fodders such as lucerne green, cowpea green, moth green and guar green are good sources of Ca (1.5-2.50%). Ca content in non-leguminous green fodders such as maize, jowar, bajra, oats and hybrid napier ranges from 0.20 to 0.55%. Carrot leaves, chikodi green, sweet potato creepers are subsidiary green fodder fed to milch animals seasonally and are good source of Ca (1.5-2.0%) and other minerals. Tree leaves also contain substantial amount of Ca (1.90-3.10%), but availability is low due to high tannin content.

Table 7: Traditional feeding practices for dairy animals in India\*

Feedstuffs	Quantity (kg)
Green fodder <sup>a</sup> (Sorghum, maize, oats, green grasses, lucerne, berseem, cow pea etc.)	5-10
Dry fodder <sup>b</sup> (Straws of wheat, rice, maize, sorghum, bajra, soybean, local dry grasses etc.)	6-8
Concentrate ingredients <sup>c</sup> (CS cake, mustard cake, grains of wheat, rice, bajra, barley, maize, sorghum etc.)	1-4
Compound cattle feed <sup>d</sup>	2-3

\*Garg et al., 2000, 2002, 2003<sup>ab</sup>, 2004, 2005<sup>ab</sup>, Bhanderi et al., 2012, 2013.

<sup>a</sup>Green fodder availability is seasonally and depends on irrigation facilities.

<sup>b</sup>Dry fodder forms bulk of ruminant diet, mainly straws of rice and wheat.

<sup>c</sup>Concentrate ingredients may change place to place, mainly locally available.

<sup>d</sup>Cattle feed is fed depending upon the level of milk yield, may or may not be fortified with minerals and vitamins.

Average Ca deficiency in the ration of milch animals is found to be from 4 to 52%, in different states.

P content in straws range from 0.09 to 0.20% but grains are high in P (0.25-0.35%) than Ca. P content in green fodders range from 0.15 to 0.45%. Cakes (>0.50%) and brans (>1.0%) contain substantial amount of P but in brans, most of P is in phytic form, which is low bio-available for animals. Average P deficiency in the ration of milch animals is recorded from 4 to 68%, in different agro-climatic zones (Das et al., 2002).

The Mg level in samples of feed and fodder is adequate (average level > 0.35%). Na level is low in concentrate ingredient (0.04-0.10%) and straws (0.10-0.20%) but high in green fodders (0.20-1.20%). Average Na deficiency in the ration of milch animals is recorded to be 20 to 55%. The K content in feeds and fodder seem to be quite rich (>1.0%), because all samples of feedstuffs are extra ordinarily rich and are far exceeding the K requirement (0.90%). The S content in most of the feedstuffs is 0.10-0.15%, giving wider N:S ratio.

Average S deficiency in the ration of milch animals is reported to be from 14 to 48%, in different states. S supplementation is needed for efficient utilization of nitrogen by rumen microbes and to maintain N:S ratio of 10:1.

#### Status of micro-minerals in feeds and fodder

Copper is one mineral element, which is acutely deficient in different parts of the country. The Cu content is consistently low in straws (1.5-7 ppm) and green fodders (4-9 ppm), whereas, concentrate ingredients are better source of Cu (12-25 ppm), except grains (4-6 ppm). Average Cu deficiency in the ration of milch animals is found to be from 12 to 66%, in different agro-climatic zones of India. Hence, Cu supplementation is necessary in the ration of milch animals.

Likewise, zinc (Zn) is acutely deficient in different agro-climatic zones (average level < 30 ppm) and need to be supplemented at a level of 80 ppm in the total ration for optimum metabolic functions (Arora, 1981). Cakes (45-60 ppm) and brans (60-98 ppm) are good sources of Cu and Zn. Average Zn deficiency in the ration of milch animals is found in the range of 33 to 80%, in different states (Table 8).

Table 8: Mineral profiles of some feedstuffs offered to dairy animals\*

Feedstuffs	(%)				(ppm)			
	Ca	P	Na	S	Cu	Zn	Mn	Fe
Dry fodder (straws of rice, wheat, sorghum, maize, bajra, sorghum etc.)	0.10-0.30	0.09-0.20	0.10-0.20	0.10-0.15	1.50-7.0	5.0-38	15-109	154-691
Green fodder (sorghum, maize, oat, lucerne, berseem, green grasses etc.)	0.20-2.50	0.15-0.45	0.20-1.20	0.06-0.20	4.0-9.0	14-37	27-170	237-1500
Concentrate ingredients (grains of wheat, maize, bajra, sorghum, barley, CS cake, groundnut cake, sesame cake, rice bran, wheat bran & pulse chunies)	0.01-0.27	0.26-0.62	0.04-0.10	0.02-0.34	4.0-25.0	30.0-98.0	7.0-74.0	42.0-701
<b>Mean value**</b>	0.29	0.24	0.074	0.13	6.85	26.0	48	306
<b>Requirements***</b>	0.42	0.34	0.18	0.20	10	80	40	50

\*Garg et al., 2000, 2002, 2003<sup>ab</sup>, 2004, 2005<sup>ab</sup>, Bhandari et al., 2012, 2013; \*\*based on analysis of more than 2000 samples; \*\*\*as per standard requirements given in Table 1.

Cobalt is found to be marginally deficient in feedstuffs (0.09-0.6 ppm), in different agro-climatic zones. Co deficiency in the ration of animals ranges from 0-30%, in different states. Mn (45-80 ppm) and Fe (150-1500 ppm) levels in most of the feeds and fodder are found to be adequate. Mo content in feeds is within the safe limit (average level < 1.5 ppm) and giving Cu:Mo ratio wider than 5.0. Selenium content in most feedstuffs is adequate (average level > 1.0 ppm).

A survey work conducted by the National Dairy Development Board (NDDB), Anand and the ICAR institutes/state agricultural universities in Gujarat, Rajasthan, Kerala, Punjab, Maharashtra, Madhya Pradesh, Uttar Pradesh and Andhra Pradesh, Haryana, Karnataka and other states indicate that Zn, Cu, Co, Mn and I are deficient in the ration of animals (Baruah et al., 2000; Garg et al., 2000,

2002, 2003<sup>ab</sup>, 2004, 2005<sup>ab</sup>; Gowda et al., 2004; Bhandari et al., 2012, 2013, Yadav et al., 2002). In several situations, there is deficiency of Ca, P and S, whereas, Mg, K, Fe and Se are adequate in the ration of dairy animals (Fig. 2). Based on the mineral mapping work, ASMM formulations have been developed for the above states having appropriate levels of Cu, Zn, Co, Ca, S and other minerals, which were found to be deficient in the ration of animals.

Overall, it is apparent from mineral studies conducted by NDDB and other ICAR institutes (Kumar et al., 2002; Datt and Chhabra, 2005; Gowda et al., 2011) that the ration of dairy animals is deficient in Ca, P, Mg, S, Cu, Zn, I, Cr and Co, and are required to be supplemented for improving growth, milk production, and reproduction efficiency.

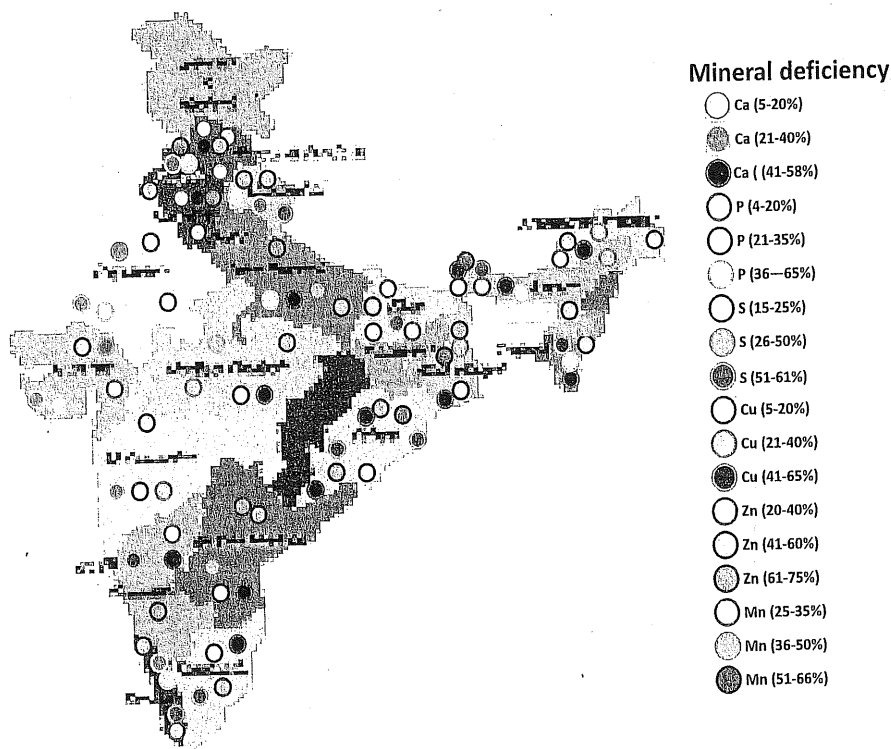


Fig. 2: Mineral deficiency in the ration of animals

## Manufacturing of area specific mineral mixtures (ASMM)

Mineral elements exist in many chemical forms such as sulphates, carbonates, chlorides, oxides, etc. There is considerable difference in the bio-availability of mineral elements provided from different sources. The form of minerals chosen for use should depend on the biological value, cost and availability in the area, stability and type of diet used. Dicalcium phosphate (DCP) is first choice of mineral ingredients used for blending mineral mixture for supplementing Ca and P to dairy animals. However, it is necessary that the levels of fluorine, lead and arsenic are kept as low as possible to prevent its being toxic to livestock. DCP should be of genuine and good quality and should not be of animal origin.

### Availability of DCP in India

For production of mineral mixture, feed grade DCP is used @ 65-75% along with other salts. Feed grade DCP is manufactured either from rock phosphate or from the bones of fallen animals. Due to spread of Mad Cow Disease in ruminant animals, also referred as Bovine Spongiform Encephalopathy (BSE), feeding of bone based DCP is confined only to poultry, whereas, rock phosphate based feed grade DCP is used for feeding dairy animals. For this reason, Bureau of Indian Standards (BIS) revised the specifications of feed grade DCP, ensuring that bone based DCP is not used for animal feeding. Even the Department of Animal Husbandry, Dairying and Fisheries also issued a notification in this regard to all stake-holders.

Bone and rock phosphate based feed grade DCP is manufactured in India at about 80,000 and 40,000 tonnes annually, respectively. For supplementing the ration of dairy animals and poultry, about 2 lakh tonnes of mineral mixture is produced annually, which contains about 1.20 lakh tonnes feed grade DCP of bone and rock phosphate origin.

## Mineral salts required for production of mineral mixture

In manufacturing mineral mixture for proper utilization in the body tissue, it is necessary to consider their bio-availability (defined as the percentage of total nutrients in feedstuffs, which are actually utilized by the animals for various production purposes), compatibility, solubility, particle size, density and chemical stability. Mineral mixture should be formulated using DCP ( $\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$ ) and sulphate form of mineral salts, except magnesium oxide. The different mineral salts, which could be used for mineral mixtures are DCP (Ca & P), calcite powder/limestone powder (Ca), magnesium oxide (Mg), ferrous sulphate (Fe), zinc sulphate & zinc methionate, zinc glycinate (Zn), potassium iodide and potassium iodate (I), sodium thio-sulphate (S), copper sulphate, copper methionate, copper glycinate (Cu), cobalt sulphate and cobalt chloride (Co) & manganese sulphate, manganese methionate, manganese glycinate (Mn) and chromium chelate (trivalent form). Active element content in these compounds is given in Table 9.

Concerned agencies in the country need to manufacture ASMM, using highly bio-available salts. Supplementing crucial trace minerals in the form of glycine and methionine-based chelates is better option for curing these deficiency problems in animals. NDDB, Anand has been providing technical assistance and technology to the dairy cooperatives, for establishment of mineral mixture plant for manufacturing ASMM.

### Improvement in palatability of mineral mixture

Palatability of mineral mixture is poor and the animals may not readily consume it. To make it palatable to animals, a sweetening agent "Sucram" @ 150 g and a flavouring agent "Lacto-vanilla coconut" @ 150 g per tonne of mineral mixture may be added.

Table 9. Commonly used mineral salts along with active element, commercial grade for animal feeding

Mineral Element	Name of the compound	Chemical formula	Active element (%)
Calcium (40.08)	Di-calcium phosphate, di-hydrate	$\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$	Ca: 23
	Mono-calcium phosphate, monohydrate	$\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O}$	Ca: 16
	Calcium carbonate/ Limestone powder	$\text{CaCO}_3$	Ca: 37
Phosphorus (30.97)	Di-calcium phosphate, di-hydrate	$\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$	P: 18
	Mono-calcium phosphate, monohydrate	$\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O}$	P: 24
Sodium (22.99)	Sodium chloride	$\text{NaCl}$	Na: 39
Iodine (126.9)	Potassium iodide	$\text{KI}$	I: 75
	Potassium iodate	$\text{KIO}_3$	I: 58
Copper (63.55)	Copper sulphate, anhydrous	$\text{CuSO}_4$	Cu: 35
	Copper sulphate, penta-hydrate	$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	Cu: 25
	Copper Chloride	$\text{CuCl}_2$	Cu: 45
	Copper methionate	$\text{CuC}_5\text{H}_{10}\text{NO}_2\text{S}$	Cu: 12
	Copper glycinate	$\text{CuC}_4\text{H}_8\text{N}_2\text{O}_4$	Cu: 16
Zinc (65.39)	Zinc sulphate, anhydrous	$\text{ZnSO}_4$	Zn: 33
	Zinc sulphate, monohydrate	$\text{ZnSO}_4 \cdot \text{H}_2\text{O}$	Zn: 30
	Zinc sulphate, heptahydrate	$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	Zn: 22
	Zinc methionate	$\text{ZnC}_5\text{H}_{10}\text{NO}_2\text{S}$	Zn: 12
	Zinc glycinate	$\text{ZnC}_4\text{H}_8\text{N}_2\text{O}_4$	Zn: 18
Manganese (54.94)	Manganese sulphate, anhydrous	$\text{MnSO}_4$	Mn: 31
	Manganese sulphate, monohydrate	$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	Mn: 29
	Manganese methionate	$\text{MnC}_5\text{H}_{10}\text{NO}_2\text{S}$	Mn: 12
	Manganese glycinate	$\text{MnC}_4\text{H}_8\text{N}_2\text{O}_4$	Mn: 15
Cobalt (58.93)	Cobalt sulphate	$\text{CoSO}_4$	Co: 20
	Cobalt chloride	$\text{CoCl}_2$	Co: 25
Iron (55.84)	Ferrous sulphate, anhydrous	$\text{FeSO}_4$	Fe: 30
	Ferrous sulphate, monohydrate	$\text{FeSO}_4 \cdot \text{H}_2\text{O}$	Fe: 28
	Ferrous sulphate, hepta-hydrate	$\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$	Fe: 20
Magnesium (24.3)	Magnesium sulphate, hepta-hydrate	$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	Mg: 9
	Magnesium oxide	$\text{MgO}$	Mg: 52
Sulphur (32.06)	Sodium thiosulphate, anhydrous	$\text{Na}_2\text{S}_2\text{O}_3$	S: 39
	Sodium sulphate	$\text{Na}_2\text{SO}_4$	S: 22
Selenium (78.96)	Sodium selenite	$\text{Na}_2\text{SeO}_3$	Se: 44
	Sodium selenate	$\text{Na}_2\text{SeO}_4$	Se: 40
Chromium (51.99)	Chromium glycinate (trivalent)	$\text{C}_6\text{H}_{12}\text{CrN}_3\text{O}_6$	Cr: 7
	Chromium (III) picolinate	$\text{Cr}(\text{C}_6\text{H}_4\text{NO}_2)_3$	Cr: 12

Cr has been shown to be an active component of the glucose tolerance factor (GTF), which helps enhancing carbohydrate metabolism. Cr also helps in enhancing metabolism of proteins and fats and improving immune status of animals. Bio-availability of Cr from feeds,

fodders and inorganic salts is negligible. Bio-availability of trivalent Cr if fed in the form of organic chelate, is very high (>80%). In view of this, many mineral mixture manufacturing units have started incorporating Cr chelate @ one kg per tonne of mineral mixture.

### Advantages of feeding area specific mineral mixtures

- Improves milk production.
- Improves reproduction efficiency in male and female animals.
- Reduces inter-calving period, more productive life of animals.
- Improves growth rate of calves, hence early puberty.
- Improves efficiency of feed utilization.
- Better immune response; hence better resistance against infectious diseases.
- Calves born are healthy.
- Improves general health of animals.
- Mineral mixture is more effective, if crucial trace minerals are incorporated in the form of chelates.

### Promoting use of ASMM through ration balancing advisory services

Due to high cost and lack of awareness, very few farmers feed mineral mixture to their animals. Mineral mixture presently produced in the country is not sufficient to cover even 2% of milch animals, due to which milk producers suffer heavy loss on account of low productivity and poor conception rate.

Various initiatives are being taken by different agencies to improve the productivity of milch animals, for meeting the future demand of milk and milk products. Amongst others, milk producers are now being advised to feed a balanced ration to their animals which helps in not only improving productivity but also helps in reducing the cost of feeding (Garg and Bhandari, 2011). This assumes greater significance in the wake of constant increase in the price of concentrate feed ingredients. For these initiatives, use of ASMM by the milk producers is of vital importance as mineral mixture not only improves milk production but

its feeding also helps in improving the conception rate in milch animals. In the ration balancing implemented areas, there was increase in sale of mineral mixture. As a result, there was improvement in general health of animals, improvement in conception rate and significant reduction in veterinarian's visit for animal treatment.

### Bio-availability of minerals

Bio-availability is the measure of the amount of an ingested nutrient that is absorbed and made available to the body for metabolic use. Bio-availability is important because nutritional intake must be available to various body systems for growth, maintenance of body tissues, reproduction and other performance factors. No matter how high the nutrient levels are or how well formulated the product is, if the nutrient is not bio-available for use by body tissue, then money and effort have been wasted. The most common source of minerals are natural feeds and fodder which the animal consumes. However, amount of the various minerals in plants and feed ingredients seldom matches the need of the animal and that the availability in these forages and feeds also vary.

The actual amount of mineral content is variable depending on plant maturity, presence of antagonistic compounds and when the forage or grain is produced on land that may have been under production for years and in which many of the minerals have been extracted through plant growth and harvest. As the genetic potential of the animal improves, mineral supplementation strategies become more vital and are influenced by a variety of factors, including feed and fodder mineral bio-availability, mineral interactions, stage of production, and even breed.

Adequate intake and balance are required for proper functioning of all the metabolic processes including immune

response and reproduction (McDowell, 2000). Depending on the factors mentioned above, bio-availability can vary widely and must be considered when diet is formulated. In view of this, bio-available minerals are required to be supplemented in the ration of animals.

### Mineral mixture production and requirement in India

NDDB standardised the process and plant design for the production of mineral mixtures (Figure 3), several agencies are now producing and marketing mineral mixtures for growing and lactating animals in different states. So far, with NDDB's assistance 28 such plants, each of 12 tonnes per day capacity, are producing about 40,000 tonnes of ASMM annually under dairy cooperatives. It is estimated that about 40,000 tonnes of mineral mixture is produced by the private sector. However, this currently addresses the need of only 2% of India's milch animals, if mineral mixture supplemented in the ration of crossbred cows and buffaloes @ 100 g per day and @ 75g per day to indigenous cows (Table 10).

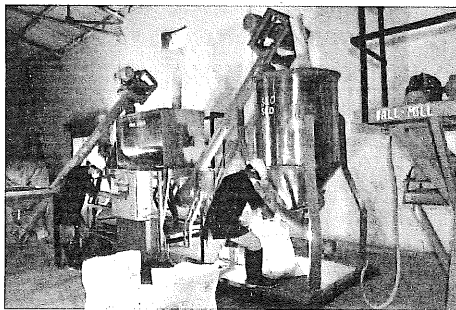


Figure 3: Mineral mixture manufacturing unit designed by NDDB

### Implications of mineral deficiency in the ration on milk production, its quality and reproduction efficiency

Ca and P are in the greatest amounts. 90% of total body Ca and 80% of total body P are stored in the bones. Skeletal stores of Ca and P are used to meet short-term deficiencies. These minerals must be replaced because long-term deficiencies of either mineral can cause bones to weaken and even break. A decrease in either or both can cause a decrease in efficiency of gain and lead to lower body condition scores. During lactation, low amounts of either Ca or P will reduce milk production. Deficiency of minerals in the ration of animals also reduces fat and SNF% in milk. P deficiency can also delay mature animals from returning to heat following parturition and can delay puberty in developing heifers.

Mn, Zn & Cu play important roles in removing superoxide radicals (free radicals) from the body. Superoxide radicals disrupt cellular membranes and cause cellular damage leaving the mammary gland more susceptible to infection, fibrosis and lost milk production. Animal needs iodine for the formation of two hormones ( $T_3$  &  $T_4$ ) that are produced by the thyroid gland. The thyroid hormones are responsible for controlling the metabolic rate of the body. Lack of iodine indirectly influences growth rate, milk production and reproduction. Iodine deficient animals may have delayed puberty and frequently do not show signs of heat. Goiter develops when iodine deficiency is severe.

Table 10: Animal population and mineral mixture requirement

Type	No. of milch animals*	Requirement of mineral mixture (kg/year)	Total mineral mixture (MT/year) requirement
Crossbred cows	1,44,06,980	36.5 (@100g/d)	5,25,854/-
Indigenous cows	4,80,41,966	27.37 (@75g/d)	13,14,908/-
Buffaloes	4,86,41,479	36.5 (@100g/d)	17,75,414/-
Total	11,10,90,425		36,16,176/-

\*Source: Livestock Census, 2007

Cu deficiency can result in delayed puberty, delayed estrus, decreased conception rates, early embryonic death and necrosis of placenta. Bulls may have poor semen quality and reduced libido. Bulls may become sterile in severe deficiency because of damage to the testicular tissue. Rough hair coats and depigmentation of hair are also signs of Cu deficiency.

The amount of Mn required for reproduction is at least 30% higher than the requirement for growth. Mn plays an important role in the process of energy metabolism and enzyme activation. A deficiency seriously affects reproductive performance of dairy animals. Animals with a Mn deficiency do not show heat, have decreased conception rates & higher abortion rates.

Zn is essential for epithelial tissue integrity, cell division and repair. Adequate Zn levels are vital for repair of the uterine lining following calving, return to normal estrus and maintenance of the uterine lining necessary for implantation and nourishment of embryos. Zn deficiency delays sexual maturity & abnormal estrus. Zn also affects the testicular development of bulls, reduces spermatogenesis and delays maturation of sperm in bull. Deficiency of minerals also increases age at first calving, inter-calving intervals, early embryonic death and reduces conception rate. Microbial protein synthesized in rumen facilitates creation of amino acids. Microbial protein provides ruminants 2/3 of their requirements of amino acids. Rumen microbes need trace amount of minerals and vitamins for their optimum growth. Insufficient level of S and P in the ration reduces the level of synthesis of microbial protein in rumen. In addition, certain trace minerals (Cu, Zn, Co, Mn, I) are also required for microbial multiplication in the rumen.

### **Use of mineral chelates in the ration of dairy animals**

Mineral amino acid chelate is the product resulting from the reaction of a metal ion from a soluble metal salt with amino acid (s) with a mole ratio of one mole of metal to one to three (preferably two) moles of amino acids to form coordinate covalent bonds. The average molecular weight of the hydrolyzed amino acids must be about 150 atomic mass units (AMU) and the resulting chelate must not exceed 800 AMU.

Traditionally, minerals are supplied to the livestock through ASMM in which minerals are present in the inorganic form. One of the major disadvantages of using such supplements is that the minerals from such sources are not fully absorbed due to antagonism and anti-nutritional factors present in the diet. Even, retention is comparatively low after the absorption. Supplementing Cu, Zn, Mn and Cr in the form of chelates would be a better option to alleviate mineral deficiency in dairy animals. In view of this, dairy animals need to be supplemented minerals with high bio-availability.

### **Supplementing deficient trace minerals in the form of chelates**

For increased bio-availability of minerals, certain crucial trace minerals need to be added in the form of chelates, to a minimum level of 50% in the ASMM formulation, which would have marked effect on the reproduction efficiency of animals. Amino acids are ideal chelators or ligands from both chemical and nutritional points of view. Amino acids meet all the chemical requirements to form heterocyclic rings of atoms which are the defining feature of the chelate. Amino acids are also ideal from a nutritional aspect. The body is very efficient at absorbing individual amino acids and di-peptides (2-amino acids linked



together through a peptide bond). Metal amino acid chelates resemble these compounds, which allow the minerals to be carried in with the amino acids during absorption. Finally, the amino acids, once released from the metal, can be used to build proteins or provide energy.

Glycine is the smallest amino acid and has low molecular weight with a high bonding capacity to the minerals, as compared to methionine, cystine and other amino acids. Glycine chelates form a homogenous dispersion in the intestinal gut inducing optimal absorption. In view of this, glycinate is preferred source of chelates for supplementing certain crucial trace minerals. Trace mineral antagonisms often result in metabolic deficiencies in dairy animals. However, it is possible to avoid the problem of antagonisms by replacing all or part of the

inorganic trace minerals in mineral mixture with organic trace minerals (OTMs). The spectrum of Zn-glycinate obtained by Fourier Transform Infra-Red (FTIR) Spectroscopy is shown in Figure 4.

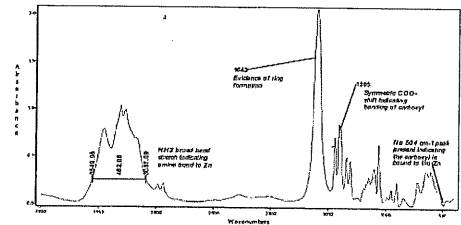


Figure 4: Spectra of zinc glycinate generated by FTIR analysis

These OTMs consist of trace minerals chemically bound to an amino acid, or carbohydrate moiety, called a ligand, thus forming a chelate, which is more bio-available (80% higher than inorganic minerals) and prevents the problems associated with mineral

Table 11. Differences between inorganic and chelated minerals

Inorganic minerals	Mineral chelates/organic minerals
These are simple mineral compounds in the form of sulphates, carbonates, oxides, chlorides etc.	These are complex compounds such as metal amino acid chelates.
Inorganic mineral compounds cannot be utilized in their natural state.	Organic mineral compounds can be utilized in their natural state. e.g. Zn-glycinate, Cu-methionate
Only a fraction of minerals are absorbed as ions, whereas, most of them get absorbed by passive absorption. Absorption in the animal body is generally poor, unable to fulfill the requirement of minerals for optimum animal performance.	Organic minerals are absorbed as a tail of amino acids; hence, there is no competition at the site of absorption with the other minerals and antagonists substances, resulting into high absorption in the animal body.
Ions are unstable molecules that need to bind with other organic molecules. For passive absorption, a carrier in the form of organic molecule is always required, which may or may not be available.	Mineral chelates are complete stable molecules and are easily absorbed passively through the intestinal wall.
Common inorganic forms of trace minerals may interact with other feed ingredients such as lipids, proteins, fiber or other minerals, anti-metabolites, inhibiting their absorption. Hence, bio-availability of inorganic minerals ranges from 5 to 50% only.	There is no inhibition by any anti-nutritional substances. Trace mineral chelates have high bio-availability (>90%) and do not react as readily as single ions.
Increasing supplementation beyond a level could affect absorption of other minerals due to mineral inter-actions.	Mineral chelates do not affect absorption of other minerals.

deficiencies (Spears, 1996). About 90% of chelated minerals are retained after absorption. Growth of ovarian follicles is faster and pregnancy rates are higher in animals fed chelated minerals. Differences between inorganic and organic minerals are given in Table 11.

### How chelated minerals work?

Chelated minerals are carried along with its bound amino acid into the intestinal cell during absorption. The chelate having glycine/methionine as a ligand is not digested prior to absorption and does not get dissociated in the stomach (Fig. 5). It remains the same molecule as the one ingested and it is easily absorbed, due to its small molecular size (less than 300 daltons), through the membranes of the cell walls of the micro-villi. Chelates are neutral molecules (with no electrical charge) that improve the intestinal absorption of trace elements reducing the interference from metabolites that form insoluble complexes with ionic trace elements.

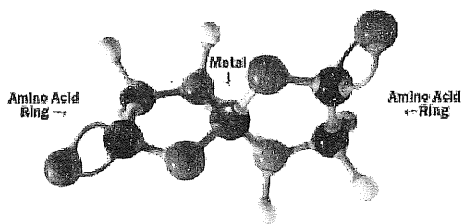


Figure 5: Single amino acid chelate

### Effect of supplementing chelated trace minerals on the reproduction efficiency of dairy animals

Infertility is a major cause of concern to the milk producers, which is primarily due to deficiency of various micro-nutrients in the ration of animals. Loss of one heat costs approx. Rs. 2,000-3,000 to the milk producer. Trace minerals crucial for reproduction are

usually deficient in the diet of animals. In view of this, a supplement was formulated containing Cu, Zn and Mn in the form of organic chelates and iodine along with coated vitamins A, D<sub>3</sub> and E, required for reproductive functions, and was tested under field conditions.

Thirty five animals with normal reproductive organs, but suffering from anoestrus were selected. Nine anoestrus animals served as control, and remaining twenty-six were given daily 5g supplement per animal, containing chelated Cu, Zn and Mn and fat-soluble vitamins A, D<sub>3</sub> and E. Minerals (Cu, Zn and Mn) and progesterone were analyzed in the blood serum before feeding of bolus and at bi-weekly intervals after feeding the supplement. Copper (0.920.01 ppm) and Zn (1.020.03 ppm) levels increased significantly ( $p < 0.05$ ) in the blood serum of animals under experimental group, as compared to control Cu (0.500.02 ppm) and Zn (0.600.03 ppm). However, level of Mn in experimental (0.060.01 ppm) and control (0.040.01 ppm) groups was similar (Table 12).

Twenty-four animals in experimental group exhibited estrus, after on an average 27 days of feeding the supplement; however, only two animals exhibited estrus in control group during this period. In control group, progesterone level did not rise and remained low (0.25-3.81ng/ml), but in experimental group the serum progesterone varied from 0.30-20ng/ml. Animals that showed estrus were subsequently inseminated either through natural service or by artificial insemination (Garg et al., 2008). The study showed that the supplementing Cu, Zn and Mn in the form of chelates, along with vitamins A, D<sub>3</sub> and E can help in curing the problems of anoestrus/repeat breeding in dairy animals.

Table 12: Average Cu, Zn, Mn and progesterone levels in blood serum of animals on feeding a minerals &amp; vitamins based supplement

Week	Cu (ppm)		Zn (ppm)		Mn (ppm)		Progesterone (ng/ml)	
	Control	Exp.	Control	Exp.	Control	Exp.	Control	Exp.
0	0.46	0.50	0.59	0.64	0.04	0.04	2.75	4.32
	0.02	0.02	0.04	0.03	0.01	0.00	(0.32-3.54)	(0.15-17.1)
2	0.50	0.76	0.60	0.76	0.04	0.04	2.13	2.05
	0.04	0.02	0.03	0.03	0.00	0.00	(0.36-5.61)	(0.0-9.85)
4	0.53	0.78	0.61	0.88	0.04	0.05	0.43	2.71
	0.03	0.02	0.03	0.02	0.01	0.00	(0.22-0.87)	(0.26-13.88)
6	0.52	0.86	0.59	0.95	0.04	0.05	1.54	3.04
	0.02	0.01	0.03	0.02	0.01	0.00	(0.12-2.10)	(0.16-15.52)
8	0.50	0.92*	0.60	1.02*	0.04	0.06	1.36	5.27
	0.02	0.01	0.03	0.03	0.04	0.01	(0.25-3.81)	(0.30-20.21)

n= 9 in control, n=26 in experimental. \*P<0.05; Figures in the parentheses indicate range.

### Effect of supplementing chelated trace minerals and vitamins on the incidences of sub-clinical mastitis

Mastitis is an infectious disease causing great economic loss, due to reduction in milk yield. Sub-clinical mastitis is 30-40 times more prevalent than the clinical mastitis. It is reported that supplementation of certain vitamins and minerals during the advanced stage of pregnancy can help in reducing the incidences of sub-clinical mastitis. In view of this, a feed supplement containing chelated minerals (Cu, Zn, Mn & Cr) and coated vitamins (vitamins A & E) was formulated and fed daily @ 10 g per head to 93 high yielding crossbred cows, with a history of clinical and sub-clinical mastitis in the previous lactation. The supplement showed encouraging results when fed four weeks prior to calving. The incidence of sub-clinical mastitis in these cows post calving reduced by 80%, as confirmed by the *Mastect* and *California Mastitis Test* (Bhanderi and Garg, 2012).

### Bio-availability of minerals from inorganic and organic sources

Minerals are important for improving production and reproduction efficiency in dairy

animals. Amongst others, Cu, Zn and Mn are widely deficient in the ration of animals, impairing livestock productivity. In a study, a group of calves was given a mineral mixture with inorganic minerals, while another group was given a mineral mixture containing Cu, Zn and Mn in the form of chelates. There was significant improvement in average daily body weight gain, total immunoglobulins, superoxide dismutase (SOD) and ferric reducing antioxidant power (FRAP) activities in the group supplemented with chelated minerals. Serum Cu, Zn and Mn levels were significantly higher (P<0.05) in calves supplemented with organic minerals. The same treatment was also applied for lactating crossbred HF cows and results of study under field conditions are given in Table 13.

It can be seen from Table 13 that there was significant improvement in immunoglobulins IgG, IgA and IgM and ferric reducing antioxidant power (FRAP) activities in the experimental group receiving chelated trace minerals. There was also significant (P<0.05) reduction in days to first service and days to conception and number of AI per conception in experimental group as compared to the control.

Table 13: Effect of supplementing chelated minerals on different parameters

Particulars	Control group (n=35)	Experimental group (n=35)
<b>Reproduction parameters</b>		
Days to first service	81±5.6	69*±4.5
Services per conception	3.20±0.31	1.9*0±0.25
Days to conception	158±15.60	108*±14.50
<b>Blood serum parameters</b>		
Serum IgG (mg/ml)	0 day	23.16±3.45
	60 days	24.56±3.06
Serum IgA (mg/ml)	0 day	0.76±0.05
	60 days	0.78±0.05
Serum IgM (mg/ml)	0 day	2.31±0.21
	60 days	2.39±0.28
Serum FRAP (µM/l)	0 day	456.76±45.67
	60 days	506.23±60.78
Serum NEFA (mEq/l)	0 day	0.42±0.06
	60 days	0.93±0.06

\*Means with different superscript in a row differ significantly (P<0.05)

#### Impact of feeding ASMM on reproduction efficiency under field conditions

The study was conducted in the Kolar union of Karnataka which is predominantly cross-bred cow area and Jaipur union of Rajasthan which is buffalo dominated area. ASMM is being regularly sold in these districts for quite some time. Concerned milk unions provided data on average sale of mineral mixture for top 20 dairy cooperative societies (DCSs) for past 4-5 years. These top 20 DCSs were arranged in descending order according to sale of mineral mixture per member or per village and thereafter, top 10 villages per union were selected for further investigation. In addition, another set of 5 villages were identified in the same geographical area with the help of milk unions, wherein, there was no sale of mineral mixture to serve as control sample. Accordingly, 30 sample villages were selected in Jaipur and Kolar districts of India.

Total enumeration of the all milch animals owning households in each of the 30 villages was undertaken to collect information

on animal owning households, animal-wise mineral mixture feeding, milk production, reproductive characteristics etc. Information was also gathered regarding packaging of mineral mixture, storage at farmer level, feeding practices, means of feeding measurement, usage of spoon provided in the mineral mixture packet etc. This was done through both personal observation and through questionnaire. The study revealed that age at first calving was 3-6 months lower in the animals supplemented with area specific mineral mixture. Number of artificial insemination (AI) per conception was lower in mineral mixture supplemented animals. On an average 63% milch animals conceived after 13 months of calving on area specific mineral mixture supplementation, whereas, 58% milch animals conceived after 15 months of calving in non-supplemented group. Increase in daily milk production was found to be in the range of 6-7% in mineral mixture supplemented animals. Lactation length, lactation yield and general health of animals were also improved on the feeding mineral mixture.

### Impact of feeding ASMM in a balanced ration on the immune status

The impact of feeding ASMM in a balanced ration on the immune status, urinary allantoin level and eggs per gram (EPG) in feces of dairy animals was studied. To demonstrate this, a study was undertaken in lactating *Gir* cows. Eighteen cows yielding daily 7 to 10 kg milk per animal were divided into two equal groups of nine each, based on milk yield, fat% and stage of lactation. Animals in control group were fed on a traditional ration comprising 10 to 13 kg green jowar fodder, 3 to 5 kg wheat straw, 1.5 kg cottonseed oil cake and 3.0 kg crushed maize. The ration of animals in experimental group was balanced for crude protein, metabolizable energy and minerals with the help of ration balancing software, using the same ingredients and incorporating required quantity of ASMM.

On feeding a balanced ration containing ASMM in experimental group for sixty days, the level of serum immunoglobulins like IgG, IgM and IgA (mg/ml) increased by 9.22, 0.41 and 0.24, respectively, as compared to the control. The urinary allantoin level increased from 10.85 to 17.97 mmol/litre and reduction in EPG of faecal samples from 184 to 77, in the experimental group. The cost of milk production was considerably lower in the experimental group fed a balanced ration compared to the control group. The study revealed that balancing ration using ASMM not only helped in improving the level of milk production and reducing the feeding cost, but also resulted in improving the immune status of dairy animals.

### Conclusion

Minerals are inorganic nutrients usually required in small amounts, for improved growth, milk production and reproduction efficiency. Minerals are present

in all body tissues and fluids and their presence is necessary for the maintenance of certain physicochemical processes which are essential to life. Balance among the nutrients, protein, energy, minerals and vitamins, is a key component in striving towards optimum animal production. Balance among the minerals themselves is also an important consideration and often poses a large challenge due to antagonist interactions that can occur between minerals. Adequate mineral intake and absorption is required for a variety of metabolic functions including immune response to pathogenic challenge, growth and reproduction. As mineral status of animal declines from adequate to marginal, immunity and enzyme functions are compromised followed by the loss of performance and reproduction. Animals in sub-clinical or marginal deficiency status are often difficult to identify, however, supplementation of minerals can result in improved production.

There is urgent need to produce and popularize ASMM for different regions, to combat deficiency of minerals in the ration of dairy animals. Immunity, growth and reproduction are influenced by trace minerals. Therefore, crucial trace minerals (Cu, Zn, Mn and Cr) could be supplemented in the form of glycine-based chelates for better bio-availability. Formulation strategies should account for mineral forms, levels and for possible antagonist combinations such as Ca: Zn and Cu: Mo ratios. Regular supplementation of ASMM containing bio-available mineral salts/trace mineral chelates would help in increasing productive life and productivity of dairy animals.

### References

- Arora, S.P., 1981. Zinc and vitamin A relationship in metabolism. In: Gawthorne, J.M., (ed.). TEM-4. Perth, Australia, Springer-Verlag, Berlin, New York. pp.572
- BAHS, 2012. Basic Animal Husbandry Statistics. Ministry of Agriculture, Department of Animal Husbandry, Dairying and Fisheries, Government of India, New Delhi.

- Baruah, K.K., Newar, S., Baruah, A.Bhuyan, D., 2000. Status of macro and micro-mineral levels in post parturanoestrus in swamp buffaloes in relation to soil and plants. *Indian J. Dairy Sci.* 53(6), 424-427.
- Beede, D.K., 2005. Formulation of rations with optimal cations and anions (DCAD) for lactation. *Proc. Tri-State Dairy Nutrition Conference-2005*. 93-112.
- Bhanderi, B.M., Garg, M.R., 2012. A study on reducing the incidences of sub-clinical and clinical mastitis in dairy cows by feeding a vitamins and minerals based strategic feed supplement. *Indian J. Dairy Sci.* 65, 388-392.
- Bhanderi, B.M., Garg, M.R., Goswami, A., 2013. Study on availability of various macro and micro-minerals in lactating buffaloes under field conditions of Sabarkantha district of Gujarat. *J. Buffalo Sci.* 2, 12-17.
- Bhanderi, B.M., Garg, M.R., Sherasia, P.L., 2013. Macro and micro-minerals content in feeds and fodders and their requirement for dairy animals in Bikaner district of Rajasthan. *Indian J. Dairy Sci.* 66, 39-46.
- Borsberry, S., Dobson, H., 1989. Peri-parturient diseases and their effect on reproductive performance in five dairy herds. *Vet. Rec.* 124, 217-219.
- Campbell, M.H., Miller, J.K., Schrick, F.N., 1999. Effect of additional cobalt, copper, manganese and zinc on reproduction and milk yield of lactating dairy cows receiving bovine somatotropin. *J. Dairy Sci.* 82, 1019.
- Chew, B.P., 2000. Micro-nutrients play role in stress, production in dairy animals, *Feedstuffs* 72, 11-12.
- Curtis, C.R., Erb, H.N., Sniffen, C.J., Smith, R.D., Powers, P.A., Smith, M.C., White, M.E., Hillman, R.B., Pearson, E.J., 1983. Association of parturient hypo-calcemia with eight peri-parturient disorders in Holstein Cows. *J. Am. Vet. Med. Assoc.* 183, 559-561.
- Das, P., Biswas, S., Ghosh, T.K., Haldar, S., 2002. Micronutrient status of dairy cattle maintained by farmers in the new alluvial zone of West Bengal. *Anim. Nutr. Feed Technol.* 2, 19-26.
- Datt, C., Chhabra, A., 2005. A review: Mineral status of Indian feeds and fodders. *Indian J. Dairy Sci.* 58, 305-320.
- Garg, M.R., Arora, S.P., Bhanderi, B.M., Sherasia, P.L., Singh, D.K., 2000. Mineral status of feeds and fodders in Kaira district of Gujarat. *Indian J. Dairy Sci.* 53, 291-297.
- Garg, M.R., Bhanderi, B.M., Sherasia, P.L., 2002. Trace minerals status of feeds and fodders in Junagadh district of Gujarat. *Indian J. Dairy Sci.* 55, 154-158.
- Garg, M.R., Bhanderi, B.M., Sherasia, P.L., 2003a. Macro-mineral status of feeds and fodders in Kutch district of Gujarat. *Anim. Nutr. Feed Technol.* 3, 179-188.
- Garg, M.R., Bhanderi, B.M., Sherasia, P.L., 2003b. Trace mineral status of feeds and fodder in Dahod and Panchmahal districts of Gujarat. *Anim. Nutr. Feed Technol.* 3, 27-36.
- Garg, M.R., Bhanderi, B.M., Sherasia, P.L., Gulati, S.K., Scott, T.W., 2004. Feeding strategies to reduce cost of milk production. *Proc.: "Nutritional Technologies for Commercialization of Animal Production Systems"*, XI Animal Nutrition Conference held at College of Veterinary Sciences, JNKVV, Jabalpur, M.P. (India) during January 5-7<sup>th</sup>, 2004.
- Garg, M.R., Bhanderi, B.M., Sherasia, P.L., 2005a. Assessment of adequacy of macro and micro mineral content of feedstuffs for dairy animals in semi-arid zone of Rajasthan. *Anim. Nutr. Feed Technol.* 5, 9-20.
- Garg, M.R., Bhanderi, B.M., Sherasia, P.L., 2005b. Mineral status of feeds and fodders in Dahod and Panchmahal districts of Gujarat. *Indian J. Dairy Sci.* 58, 1-6.
- Garg, M.R., Bhanderi, B.M., Gupta, S.K., 2008. Effect of supplementing certain chelated minerals and vitamins to overcome infertility in field animals. *Indian J. Dairy Sci.* 61, 181-184.
- Garg, M.R., Bhanderi, B.M., Gupta, S.K., 2011. A study on availability of various mineral elements in milch buffaloes. *Anim. Nutr. Feed Technol.* 11, 91-102.
- Garg, M.R., Bhanderi, B.M., 2011. Enhancing livestock productivity through balanced feeding. *Proc. 14<sup>th</sup> Biennial National Conference of Animal Nutrition Society of India on "Livestock Productivity Enhancement with Available Feed Resources" from 3<sup>rd</sup> to 5<sup>th</sup> November, 2011 held at G.B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand.* pp. 11-21.
- Goff, J., 2003. Managing hypo-calcemia and milk fever. *Cattle Practice: British Cattle Veterinary Association, Frampton-on-Severn, U.K.* 11(2), 75-79.
- Gowda, N.K.S., Prasad, C.S., Ramana, J.V., Ramachandra, K.S., 2001. Micronutrient content of soil, feeds, fodders and blood samples of animals in southern dry and eastern dry zones of Karnataka. *Indian J. Anim. Sci.* 71(2), 150-154.
- Gowda, N.K.S., Ramana, J.V., Prasad, C.S., Singh, K., 2004. Micronutrient content of certain tropical conventional and unconventional feed resources of southern India. *Trop. Anim. Health Prod.* 36, 77-94.
- Gowda, N.K.S., Prasad, C.S., 2005. Macro and micro nutrient utilization and milk production in crossbred dairy cows fed finger millet and rice straw as dry roughage source. *Asian Australas. J. Anim. Sci.* 18, 48-53.
- Gowda, N.K.S., Pal, D.T., Verma, S., Garg, M.R., Prasad, C.S., 2011. Minerals in livestock production under different agro-eco systems of India and measures to ameliorate imbalances. *Proc. 14<sup>th</sup> Biennial Animal Nutrition Conference on "Livestock Productivity Enhancement with Available Feed Resources" held at GBPUA&T, Pantnagar, Uttarakhand, 3-5 November, 2011.* pp. 81-96.
- Jonsson, N.N., Daniel, R.C.W., 1997. Effects of hypocalcaemia on blood flow to the ovaries of the sheep. *J. Vet. Med. A.* 44, 281-287.
- Kearl, L.C., 1982. Nutrient Requirements of Ruminants in Developing Countries. *International Feedstuffs Institute, Utah State University, UMC 46, Logan, Utah*

- 84322 USA.
- Kellogg, D.W., 1990. Zinc methionine affects performance of lactating cows. *Feedstuffs*, 62, 15.
- Kumar, B., Pachauri, S.P. Kumar, N., 2002. Evaluation of trace elements status of dairy cattle, feed, fodder and soil at medium elevation in hills to establish soil-plant-animal-relationship. *Indian J. Anim. Sci.* 72(4), 332-336.
- McDowell, L.R., 1992. *Minerals in Animal and Human Nutrition*. Academic Press, San Diego, CA. pp. 49-51.
- McDowell, L.R., 2000. Recent advances in minerals and vitamins on nutrition of lactating cows. University of Florida, Gainesville, FL. pp. 1-14.
- N.R.C., 2001. *Nutrient Requirements of Dairy Cattle*, 7<sup>th</sup> Revised edition. National Academy of Sciences, Washington, DC.
- Prasad, C.S., Gowda, N.K.S., 2005. Importance of trace minerals and relevance of their supplementation in tropical animal feeding system: A review. *Indian J. Anim. Sci.* 75(1), 92-100.
- Ramana, J.V., Prasad, C.S., Gowda, N.K.S., Ramachandra, K.S., 2001. Mineral status of soil, feed, fodders and blood plasma of animals in northern dry and northern transition zone of Karnataka. *Indian J. Dairy Sci.* 54(1), 40-46.
- Reid, R.L., Horvath, D.J., 1980. Soil chemistry and mineral problems in farm livestock: A review. *Anim. Feed Sci. Tech.* 5, 95-167.
- Samanta, A., Samanta, G., 2002. Mineral profile of different feed and fodders and their effect on plasma profile in ruminants of West Bengal. *Indian J. Anim. Sci.* 19, 278-281.
- Sharma, M.C., Joshi, C., Sarkar, T.K., 2002. Therapeutic efficacy of minerals supplement in macro-minerals deficient buffaloes and its effect on haemato-biochemical profile and production. *Asian Austral. J. Anim. Sci.* 15, 1278-1287.
- Singh, S.V., Pachauri, S.P., 2001. Zinc, immunity and mastitis, *Pashudhan*, 16:1.
- Spears, J.W., 1996. Organic trace minerals in ruminant nutrition. *Anim. Feed Sci. Tech.* 58, 151-163.
- Udar, S.A., Chopde, S., Dhore, R.N., 2003. Mineral profile of soil, feeds and fodder and buffaloes in western agro-climatic zone of Vidarbha. *Anim. Nutr. Feed Technol.* 3, 165-172.
- Underwood, E.J., Suttle, N.F., 2010. *The Mineral Nutrition of Livestock*. 4<sup>th</sup> ed. CAB International Publishing Co., London, U.K.
- Yadav, P.S., Mandal, A.B., Dahiya, D.V., 2002. Feeding pattern and mineral status of buffaloes in Panipat district of Haryana state. *Anim. Nutr. Feed Technol.* 2, 127-138.