

ORIGINAL ARTICLE**The role of macro and micro minerals in calcite and calcium carbonate as livestock feed supplements****Abegaze Beyene^{1,2*} and Ramesh Chopra²**¹College of Agriculture and Veterinary Medicine, Jimma University, P.O. Box: 307, Jimma, Ethiopia²Division of dairy cattle nutrition national dairy research institute (Deemed university) Karnal Haryana, India*Correspondence email: abegazebeyene@yahoo.com**ABSTRACT**

In India, feed industries widely use limestone or calcium carbonate as a cheap source of Ca and other mineral supplements without considering the sources, variability, and limitations of using it as feed. Calcium carbonate and calcite powder are potential mineral concentrates for livestock feeding. However, In contrast to calcium carbonate, calcite powder is not adequately studied as a source of mineral supplements for animal feeding. This being the case, this study was conducted to evaluate calcite powder as an alternate source of Macro minerals, Calcium, Phosphorus, Magnesium and Potassium and trace minerals like Manganese, Copper, Zinc and Iron for dairy cattle feeding. Both calcite powder and calcium carbonate were collected from various parts of India, i.e. Delhi, Gujarat, Maharashtra, Uttarakhand and Uttar Pradesh. The samples collected were thoroughly mixed, processed and subjected to laboratory chemical analysis in triplicate. The results showed that the total ash content of calcite and Calcium carbonate was 96 and 99%, respectively, indicating that limestone (CaCO₃) and calcite powder, collected from different parts of India, were potential mineral supplementary feed sources. The results also showed that calcite powder was superior to calcium carbonate in calcium content 40.03 ± 0.27 and 39.17 ± 0.39 , respectively, showing that the calcium content of calcite powder was not significantly higher than that of calcium carbonate. Calcite powder was also superior to calcium carbonate in Phosphorus content, 0.47 ± 0.18 and 0.14 ± 0.03 , respectively, show that the Phosphorus content of calcite powder was not significantly higher than that of calcium carbonate. Calcite powder was comparable with manganese, zinc and iron contents. Magnesium and Copper contents in Calcite powder are higher than in calcium Carbonate. However, the magnesium level in both calcite powder and calcium carbonate is above the recommended level, especially magnesium in calcite powder is very high, so it should be reduced when feeding animals. The percentage composition of iron was very low in calcium carbonate and calcite powder. The phosphorus level in both sources is below the recommended level; the animals should be supplemented from other sources. The mean Acid insoluble ash (AIA) content in calcite powder and calcium carbonate was 2.84 and 1.42 % of DM composition, respectively. As a high level of AIA lowers the utilization and palatability of nutrients, in our study, calcite powder will lower the utilization and palatability of nutrients more than calcium carbonate. Considering the research outcome, both sources of minerals (Calcium carbonate and Calcite powder) can be used safely to supplement dairy cattle and other domestic animal diets. However, further study into the toxic element content of calcite powder and Calcium Carbonate is required before its large-scale use in feed industries.

Keywords: Lime stone, Trace minerals, Genetic potential, palatability, insoluble ash

INTRODUCTION

The performance of animal production depends on several factors, including the genetic potential of the animals, the environment, nutritional quality and adequacy and managerial practices employed at the farm. Animal nutrition is important in farm economics, as more than 50% of farm expenditure goes towards feeding animals. Dietary nutrients promote the programming and expression of the metabolic pathways that enable the animal to achieve its genetic potential with respect to production and reproduction. Adequate supply of all the nutrients (carbohydrate, proteins, fat, vitamins, minerals, and water) is equally important, as one or more of these can hamper the productivity of animals when requirements are not fulfilled. Minerals are divided into macro and micro minerals. Macro minerals are required by the animals in grams, while micro minerals are required in milligrams or in PPM (parts per million). In the context of nutrition, minerals are inorganic chemical elements required as essential nutrients by organisms to perform functions necessary for life. Minerals constitute a small fraction of the total ration, but these micronutrients are vital in the body.

Mineral elements exist in the animal body's cells and tissues, and their characteristic concentrations vary with the element and tissue. The concentrations of essential elements must usually be maintained within the narrow limits, if the functional and structural integrity of the tissues is to be safeguarded and the optimum growth, health and productivity of the animal are to be maintained. Continued ingestion of diets that are deficient, imbalanced or excessively high in a mineral, induce changes in the form of concentration of that mineral in the body tissues and fluids. Deficiency of minerals leads to the development of biochemical lesions, change or abnormality in physiological functions and development of structural disorders all of which vary with the element concerned and the degree and duration of dietary deficiency (Chesters and Arthur, 1988), level of toxicity and the age, sex and species of animal involved. The homeostatic mechanism in the body can be brought into play, which delay or minimize the onset of such diet-induced changes. Ultimate prevention of the changes requires that the animal be supplied with a palatable and nontoxic diet containing the required minerals, as well as other nutrients, in adequate amounts, proper proportions, and available forms. Many livestock in many parts of the world consume diets that do not meet their requirements (McDowell *et al.*, 1993), necessitating the need for mineral supplementation.

Mineral supplements are nutritional devices required to fortify the common animal feeds and fodders to meet the mineral needs of livestock and poultry at specific levels of animal productivity. The effort to increase animal productivity by manipulating genetic potentials has further accentuated the problem

of mineral nutrition, particularly mineral supplements. In the case of high-producing animals, there is a tremendous daily drainage of calcium through milk and adequate mineral supplements that need to be devised to replenish the daily loss, even though internal regulatory mechanisms in animal can take care of the transient periods of enhanced needs. The specifications of the mineral supplements to be used have been laid down in certain countries. In India, feed industries are widely using limestone or calcium carbonate as a cheap source of calcium and other mineral supplements without considering the sources, variability's, and the limitations of using it as feed. Both calcium carbonate and calcite powder are potential mineral concentrates for livestock feeding. However, In contrast to calcium carbonate, calcite powder is not adequately studied as source of mineral supplements for animal feeding. This being the case, the objective of this study was to evaluate calcite powder and calcium carbonate as alternate sources of major minerals calcium, phosphorus, magnesium, and micro minerals manganese, copper, zinc, and iron for dairy cattle feeding.

MATERIALS AND METHODS

Collection of calcite powder and calcium carbonate

Adequate amount of calcite powder and calcium carbonate were collected from different parts of India i.e Delhi, Gujarat, Maharashtra, Uttarakh and UttarPrade. Some samples of calcite powder and calcium carbonate (CaCO₃) were procured from different sources/ locations and compared with pure CaCO₃ (Table 1). Efforts were made to procure as many batch samples as possible

Table 1. Sources of calcite powder and calcium carbonate

Name of Sample	Place of Collection
Calcite powder	Itola(Gujarat)
	Itola(Gujarat)
	Itola(Gujarat)
	Delhi
	Delhi
	Dehradoon (Utrakhand)
	Sikenderabad (Uttarpradesh)
	Sikenderabad (Uttarpradesh)
	Anand (Gujarat)
Anand (Gujarat)	
Calcium carbonate	Mumbai (maharashtra)
	Mumbai (maharashtra)
	Delhi

Sample and Standard Solution Preparation

Samples of 1-2 gm of dried mineral supplement was taken in silica basin and charred to remove smoke and ashed at 550°C in a muffle furnace for two hrs. Acid extract was prepared by quantitative transfer of ash to a

dried clean glass beaker to which 20 ml of 5 N HCl was added this was boiled for 5 minutes and filtered through what man filter paper No. 42 into 250 ml volumetric flask. The filter paper was washed with hot distilled water until free of acid; the volume was made

to the mark with distilled water. This extract was used for analysis of Mg, Ca, Zn, Cu, Mn and Fe. All the required standard solutions were prepared as seen in Table 2.

Table 2. Preparation of standard solutions for various elements

Element	Salt	Quantity in mg	Yield	Standard range
Calcium	CaCl ₂ .2H ₂ O	40.76	100 ppm	1-20 ppm
Manganese	MnSO ₄ .H ₂ O	31.39	100 ppm	0.5-5 ppm
Preparation of standard solutions for various elements				
Zinc	ZnSO ₄ .7H ₂ O	44.235	100 ppm	0.4-2 ppm
Copper	CuSO ₄ .5H ₂ O	39.89	100 ppm	0.8-8 ppm
Magnesium	MgSO ₄ .7H ₂ O	102.43	100 ppm	0.06-0.6 ppm
Iron	FeSO ₄ .7H ₂ O	50.80	100 ppm	0.8-8 ppm

After adjusting the instrument, the standards and unknown were monitored through the samples spraying device to get the constant reading in the digital display

Laboratory Analysis

The laboratory analysis was done in triplicate for dry matter (DM), total ash, acid insoluble ash (AIA), Mn, Zn, Cu, Mg and Fe as trace minerals. In addition, Total ash and AIA were analyzed by the method of AOAC (2002) Macro & Micro minerals like Ca, Mg, Mn, Zn, Cu, and Fe were analyzed using AAS (Hitachi Z-500), polarized zeeman AAS) using acetylene as fuel and air as oxidant. Specific hollow cathode lamps were used for the determination of each element

Strontium as strontium chloride was added for Ca and Mg estimation in a separate aliquot as releasing (or) damasking agent in the process of dilution of acid extract so that a concentration of 0.2 percent strontium (for Ca) and 0.1 per cent strontium (for Mg) were achieved into the test samples as well as in the respective standards as recommended.

Estimation of total phosphorus

Total phosphorus in the mineral supplement samples was estimated by spectrophotometric method (BIS, 2000) as described below. Molybdo vandate reagent in hot distilled water in a beaker 50 gm of ammonium molybdate was dissolved. A 5 ml ammonia was then added and the volume was made to 500 ml in a volumetric flask with distilled water. In another beaker 1.175 gm of ammonium mono vandate was dissolved in about 200 ml of hot distilled water and to it was added slowly 3.5 ml nitric acid with constant stirring and volume was made in a volumetric flask with distilled water to 500 ml. In a 1L volumetric flask, 200 ml ammonium molybdated solution, 200 ml ammonium mono vandate solution and 135 ml nitric acid was diluted with distilled water to the mark and in soluble particles if present were filtered.

Preparation of standard curve

Phosphorus standard solution (1 mg p/ ml) was prepared in distilled water by dissolving 4.39 g previously dried potassium di-hydrogen phosphate

(KH₂PO₄) in 1L volumetric flask and making the volume to the mark using this standard solution. Working solutions containing 5, 10, 20, 30 and 40 µg P/L respectively were prepared. 10 ml each working solution containing 0, 5, 10, 20, 30 and 40 µg P/ml were pipette to a series of test tubes and 10 ml of molybdovandate reagent was added to each tube. The contents were well mixed and allowed to stand for at least 10 minutes at 20°C. A portion of the content from each tube was transferred to a measuring cell of spectrophotometer and absorbance measured at 430 nm against a reference solution.

Reference solution

It was prepared by diluting 10 ml of molybdovandate reagent with 10 ml distilled water. A standard curve was drawn (Fig. 1) by plotting the absorbance against the corresponding phosphorus concentration. Suitable aliquot of the filtrate obtained was diluted with distilled water to obtain a phosphorus content not exceeding 40 µg/ ml. 10 ml of this solution was transferred to a test tube and 10 ml of molybdovandate reagent was added to it. These contents of the tube were mixed well and allowed to stand for at least 10 minutes at 20°C for proper development of color due to the formation of phosphor molybdovandate complex. There after a portion of this solution was transferred to a measuring cell and absorbance was transferred to a measuring cell and absorbance was measured at 430 nm against a reference solution.

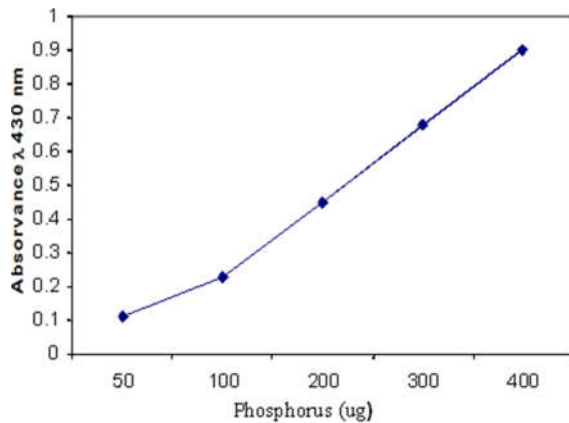


Figure 1: Standard curve for total phosphorus
P content of the samples was calculated from the standard curve as follows: From the standard curve (Fig.1.)

$$0.40 \text{ OD} = 175 \mu\text{g P}$$

$$\text{If OD of the sample is } X = \frac{175}{0.40} \times \mu\text{g P} = 70 \mu\text{g P}$$

If the aliquot taken is a ml out of 250 ml of the HCl extract prepared by ashing 1 gm of the samples then,

$$\text{P percentage in the samples} = \frac{175}{0.40} \times \frac{250}{a} \times 100 \cdot \frac{1}{10^6} \\ = 10.9375x/a = 10.94\%$$

RESULT AND DISCUSSION

Comparative mineral content of calcite powder and calcium carbonate

The results of the relative calcium composition of calcite powder and calcium carbonate are shown in Table 3. In India mineral supplements for livestock feeding are marketed under various trade names. The quality standards of such products are regulated according to specification of ISI/ BIS (1962, 1968, 1982, 1992 and 2002). In most cases due to cost considerations calcium sources in mineral mixtures are included in the forms of

ores, rocks and other locally available natural sources. Apart from compositional variability's, in such sources there could be one or more toxic mineral elements (Ammerman *et al.*, 1977) which may limit the use of such ingredients. The ISI standards (1982) recommended the use of bone meal, chalk powder and DCP as calcium supplements in mineral mixtures. Recently in 2002 in view of growing concern against the use of bovine origin ingredients as mineral supplements to ruminants, in its fourth revision. The BIS withdrew the use of mineral supplement of animal origin and has proposed the use of calcite powder in mineral mixture (BIS, 2002). Indian cattle feed industry is using calcite powder on a wide scale on account of its cheaper and easy availability, but the literature is scanty on the efficiency utilization of calcite from calcite powder in livestock, although it has been used as buffer in high milk producing cows (Keyser *et al.*, 1985). In the present experiment, compositional quality of calcite powder was compared with calcium carbonate which is widely used in the Indian; feed industries as of calcium and other mineral supplements. As shown in Table 3. The content of AIA in calcite powder varied from 0.74 to 4.07 with an average of 2.84 ± 0.39 . The AIA content of calcium carbonate was found to be 1.34 to 1.48 with an average of 1.42 ± 0.04 for AIA.

The calcium content in calcite powder varied from 39.0 to 41.3 percent which was quite high and similar to that of CaCO_3 (Table 3) Lall (1987) reported that carbonates of calcium were rich in calcium as compared to sulphate forms of calcium. The sulphate forms of calcium such as gypsum and phosphogypsum were found to contain 12 - 35.6% calcium. Acid insoluble ash (AIA) content in the calcite powder varied from 0.74 to 4.07% with an average value of $2.41 \pm 1.67\%$. Since calcite powder contained high amount of calcium and low amount of AIA, it could be considered a suitable source of calcium supplement for livestock.

Table 3. AIA, Ca, P, Mg and Fe contents (on percent DM basis) in calcite powder and pure calcium carbonate

Place of collection	Total Ash	AIA	Mg	Ca	P	Fe
Itola (Gujarat)	98.86 ± 0.0	3.47 ± 0.1	5.43 ± 0.0	39.7 ± 0.0	0.02 ± 0.0	0.20 ± 0.0
Itola (Gujarat)	99.08 ± 0.0	4.07 ± 0.0	4.76 ± 0.1	39.0 ± 0.0	0.01 ± 0.0	0.20 ± 0.0
Itola (Gujarat)	96.39 ± 0.0	3.44 ± 0.0	6.00 ± 0.1	39.5 ± 0.0	0.02 ± 0.0	0.20 ± 0.0
Delhi	99.52 ± 0.0	2.42 ± 0.0	6.49 ± 0.1	39.1 ± 0.0	0.04 ± 0.0	0.08 ± 0.0
Delhi	99.52 ± 0.0	3.22 ± 0.0	7.01 ± 0.1	39.9 ± 0.0	0.08 ± 0.0	0.09 ± 0.0
Dehradun (Utrakhand)	99.44 ± 0.0	2.18 ± 0.0	6.76 ± 0.1	39.4 ± 0.0	0.17 ± 0.0	0.09 ± 0.0
Sikandarabad (Uttar Pradesh)	98.04 ± 0.0	1.20 ± 0.0	2.38 ± 0.1	41.3 ± 0.0	0.86 ± 0.0	0.04 ± 0.0
Sikandarabad (Uttar Pradesh)	97.46 ± 0.0	1.15 ± 0.0	2.11 ± 0.0	40.5 ± 0.0	0.93 ± 0.0	0.04 ± 0.0
Anand (Gujarat)	97.63 ± 0.0	0.95 ± 0.0	1.94 ± 0.1	41.3 ± 0.0	1.20 ± 0.0	0.04 ± 0.0
Anand (Gujarat)	98.33 ± 0.0	0.74 ± 0.0	2.22 ± 0.1	40.6 ± 0.0	1.40 ± 0.0	0.04 ± 0.0
	98.42 ± 0.3	2.84 ± 0.4	4.61 ± 0.7	40.03 ± 0.3	0.47 ± 0.2	40.03 ± 0.3

The BIS (2002) restricted AIA content to 2.5 to 3.0% in the final mineral mixtures as high levels of AIA lowers

the utilization and palatability of nutrients. Ammerman *et al* (1984) reported that high levels of AIA

in the ration of livestock depressed the utilization of P and certain other micronutrients. Gohl (1981) suggested that lime stone contained 36.4% of calcium and could safely be fed free choice mixed with salt to livestock. The results presented in Table 3 & 4 further showed that calcite powder contained moderately high levels of iron and manganese that could be added few in to livestock feed. Lall (1987) reported that marble powder

which is also a carbonate form of calcium contained 45-68 ppm iron the value of which was much lower than that found in calcite powder. Maynard and Loosli (1969) reported that calcium content of rock phosphate varied from 20 to 36% and 12 to 18% respectively and that calcium content of rock phosphate are biologically available.

Table 4. Mn, Cu and Zn content (ppm) in calcite powder and calcium carbonate

Material	Place of collection	Mn	Cu	Zn	
Calcite powder	Itola(Gujarat)	162 ± 2.2	19 ± 0.00	34 ± 0.20	
		161 ± 2.2	20 ± 0.10	26 ± 0.20	
	Delhi	174 ± 2.2	19 ± 0.04	29 ± 0.23	
		202 ± 2.2	16 ± 0.10	48 ± 0.20	
		203 ± 2.2	16 ± 0.10	47 ± 0.20	
		Dehradoon (Utrakhand)	189 ± 2.2	16 ± 0.10	45 ± 0.20
		Sikandarabad (Uttar Pradesh)	314 ± 2.2	17 ± 0.10	33 ± 0.20
		Sikandarabad (Uttar Pradesh)	315 ± 2.2	17 ± 0.00	34 ± 0.20
		Anand (Gujaqrat)	312 ± 2.2	17 ± 0.10	40 ± 0.20
			312 ± 2.2	17 ± 0.10	39 ± 0.20
			234.50 ± 22.0	17.40 ± 0.5	37.50 ± 2.40
Calcium carbonate	Mumbai (Maharashtra)	236 ± 0.80	8 ± 0.00	22 ± 0.40	
		229 ± 0.80	8 ± 0.00	19 ± 0.40	
	Delhi	236 ± 0.80	8 ± 0.00	23 ± 0.40	
		233.67 ± 2.30	8 ± 0.00	21.33 ± 1.20	

That Mg content in the calcite powder obtained from Delhi was highest followed by samples procured from Itola and Dehradoon. The presence of Mg in calcite powder moderate quantity was an added advantage. Gohl (1981) suggested that lime stone that contained 36.4% Ca can safely be fed free choice mixed with salt to livestock, however due to high Mg CO₃ content (about 5% in dolomite limestone) it should not be used in feeding of poultry. Maynard and Loosli (1969) reported that P content in rock phosphate varied from 12 to 18% percent and P content of rock phosphate are observable.

In summary, it could be concluded that calcite powder could be used as a possible alternate source of calcium because of it is low than recommended by BIS (2002) restricted AIA content to 2.5 to 3.0%, However, actual use needs to be confirmed on the basis of solubility studies in the rumen and availability of calcium to the animals.

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