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# Influence of Caecectomy on the Bioavailability of Minerals from Vegetable Protein Supplements in Adult Roosters

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**ABSTRACT :** The present study was designed to assess the influence of caeca on the availability of calcium, phosphorus, magnesium, manganese and copper from soybean, sunflower, rapeseed, sesame, fish and meat cum bone meal in adult roosters. The excretion of endogenous origin minerals viz., copper, magnesium, manganese and calcium was significantly ($p<0.001$) higher in caecectomized than in normal roosters. The difference in the endogenous excretion was 50; 60.45; 40.35 and 29.63 per cent for copper, magnesium, manganese and calcium, respectively, in caecectomized roosters. The caeca played a pivotal role in the reabsorption of endogenous origin calcium, magnesium, manganese and copper. The mechanism of phosphorus absorption by the caecal epithelium was negligible. The caecectomized roosters underestimated the bioavailability of copper in sunflower meal and manganese in almost all the test feedstuffs. The present investigation revealed that the caeca played a critical role in the absorption of minerals from vegetable protein feedstuffs which escape digestion and absorption in the small and large intestinal segments. (**Key Words :** Mineral Availability, Feed Ingredient, Caecectomized Rooster)

## INTRODUCTION

Mineral requirements of poultry have received little attention over the last 40 years. A review of scientific reports over the last 15 years indicated that only little effort (<1.5%) have been expended on minerals research (Leeson and Summers, 1997). Undoubtedly the major reason for this lackluster is their relative economic importance. They represent less than 0.5% of total diet cost which is overwhelmed by the costs of energy and amino acids. Animal Nutritionists generally rely on the recommendations of NRC (1994) as a starting point in poultry diet formulation. A concern often raised about these values is that the NRC values are necessarily based on somewhat historic data based solely on information published in referenced scientific journals prior to 1980. With changes in genetics of different poultry species, and dramatic changes in performance, the validity of such historic data is often questioned. The resurgence of interest in trace mineral nutrition has been brought about by concerns for the environment and particularly the level of all nutrients in manure.

The major concern in estimating mineral requirements is the knowledge of bioavailability of minerals from an array of poultry feedstuffs. There is very little information available on this topic. O'Dell et al. (1972) and Nwokolo and Bragg (1980) reported 40-70% availability of Mg, Mn, Cu and Zn in canola meal and 50-78% availability in soybean meal. There is virtually no information available to account for such variability, and so predicting the bioavailability of minerals in common ingredients becomes a tenuous exercise. Several workers have investigated the balance of nitrogen, amino acids and phosphorus in poultry nutrition, and there are ongoing attempts at limiting their concentration in manure. Recently data on the bioavailability of amino acids and nitrogen are generated by ileal digestibility method for a wide range of poultry feedstuffs (Payne et al., 1971; Nordheim and Coon, 1984; Sibbald, 1986; Green et al., 1987; Ravindran et al., 1999; Short et al., 1999; Van Leeuwen et al., 2000). But to the authors knowledge there are no reports on the bioavailability of minerals for various feedstuffs. Hence the present study was designed to evaluate various vegetable protein supplements for its mineral bioavailability and to assess the impact of caeca on mineral absorption.

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**Table 1.** Composition of mineral free diet

| Ingredient | mg/kg |
|---|---|
| Glucose | 312.00 |
| Sucrose | 500.00 |
| Cellulose | 85.65 |
| Refined soya oil | 100.00 |
| Vitamin mixture* | 2.35 |

* Vitamin mixture: Vit. premix[a] 0.2 g; B complex-0.15 g ; Choline chloride (50%) 2.0 g.

[a] Vitamin premix (mg/kg): Vitamin A 82,500 IU, $B_2$ 50 mg, $D_3$ 62,000 IU, K 10 mg.

## MATERIALS AND METHODS

### Experimental procedure

Adult White Leghorn Cockerels (n = 60), 25 weeks of age, selected from a single flock of same hatch were used for the present study. About 30 cockerels were caecectomized as per the methodology described by Maiti et al. (2006).

### Caecectomy

The Birds were fasted for 24 h before surgery. Initially the birds were sedated by intravenous administration of diazepam (at 2.5 mg per kg body weight) into wing vein. ketamine was administered intramuscularly on breast muscle (at 75 mg per kg body weight) after two minutes to induce anaesthesia. Feathers from the abdominal area were removed carefully and the area was disinfected with povidone iodine solution. A surgical drape was placed over the site of operation. A laparotomy was performed through a horizontal incision of approximately 4-5 cm at a position nearly midway between the pelvis and the distal end of the breastbone. The caeca, mesentery and the adjacent intestinal segments were exposed out. Each caecum was carefully detached from the mesentery by hand from the distal end towards the ileo-caecal junction. During this process haemostypticum was sprayed as coagulant to arrest bleeding if any. After isolating both the caeca, each was cut as near as possible to the junctions of the caeca and the intestine.

After the removal of caeca, the cut ends were sutured by using catgut (size 1/0) and an antibiotic (oxytetracycline) solution was applied over the blind ends. The exposed intestine and mesentery was returned to the peritoneal cavity, 1.5 ml of oxytetracycline was sprayed into the peritoneal cavity to prevent secondary infections and adhesions. The peritoneum and the muscular layers were sutured by catgut (size 1/0). The skin layer was closed with silk thread. The birds were allocated to test four weeks after surgery.

The caecectomized cockerels (30n) and the intact birds (30n) were divided into five replicates of six birds each. Each bird was confined to an individual metabolic cage. All the birds were starved for 48 h. Each replicate of caecectomized and intact birds was fed with one test ingredient. The first replicate of birds of both the treatments was precision fed with 50 g of mineral free diet to determine the endogenous mineral excretions. Soybean meal was fed to second group of birds as per Farrell's (1978) rapid ME method. The other three replicates of both the treatments were precision fed (Sibbald, 1976) with screw pressed sesame meal (50 g), solvent extracted sunflower meal (35 g) and screw pressed rapeseed meal (35 g) respectively. Drinking water was provided *ad libitum*. The composition of mineral free diet was designed as per Nwokolo et al. (1976) with slight alterations (Table 1).

The excreta were collected for the following 48 h at 8 h intervals (three times daily). The excreta of each bird were oven dried individually at 80°C for 24 h, finely ground and stored separately in a sealed airtight polythene bag at -24°C (deep freezer) until further analysis. The dry matter, total ash and mineral aliquots of feedstuffs as well as excreta samples were estimated as per the AOAC (1995) procedure.

### Mineral analysis

Calcium was estimated as per the methodology of Talapatra et al. (1940). Total Phosphorus was analyzed as per the AOAC (1995) procedure. Copper, magnesium and manganese were determined using Atomic absorption spectrophotometer (Spectra A220 model, VARIAN, Australia), by absorbance method. The true mineral availability was calculated using the following equation

$$\text{TMA}(\%) = \frac{M_i - (M_e - EM_{MF})}{DMI}$$

Where, DMI is the quantity of feed dry matter intake (g), $M_i$ is the quantity of mineral intake (mg), $M_e$ is the quantity of mineral excreted (mg) and $EM_{MF}$ is the quantity of endogenous mineral voided by mineral free diet fed cockerel (mg).

All the experimental procedures were approved by the Animal Ethics Committee of the Indian Veterinary Research Institute University and Indian Council of Agricultural Research. The data obtained from the above experiments were subjected to statistical analysis (independent 't' test, and ANOVA) as per standard procedure (Snedecor and Cochran, 1980).

## RESULTS

The mineral composition of the vegetable and animal protein supplements are presented in Table 2. A wide range in total ash content was observed among vegetable protein sources (7.59-16.98%). The ash content was highest in sesame meal and least in soybean meal. Sesame meal was the richest source of magnesium and copper among the

**Table 2.** Nutrient composition of vegetable and animal protein sources (DM basis)

| Attributes | Soybean meal | Rapeseed meal | Sesame meal | Sunflower meal |
|---|---|---|---|---|
| Dry matter (%) | 91.43 | 92.81 | 89.48 | 91.57 |
| Total ash (%) | 7.59 | 9.23 | 16.98 | 12.21 |
| Ca (%) | 0.37 | 1.01 | 3.34 | 1.18 |
| Total P (%) | 0.75 | 1.23 | 1.36 | 1.28 |
| Mg (%) | 0.13 | 0.16 | 0.17 | 0.15 |
| Mn (mg/kg) | 69.16 | 62.13 | 52.64 | 59.83 |
| Cu (mg/kg) | 15.30 | 11.89 | 27.63 | 26.48 |

**Table 3.** Endogenous mineral output ($bird^{-d}$) by intact and caecectomized cockerels fed mineral free diet

| Attributes | Caecectomized | Intact | Probability |
|---|---|---|---|
| Copper (mg) | $0.08^a$±0.008 | $0.04^b$±0.003 | *** |
| Magnesium (mg) | $8.37^a$±0.79 | $3.31^b$±0.20 | *** |
| Manganese (mg) | $0.57^a$±0.05 | $0.34^b$±0.02 | *** |
| Calcium (g) | $108^a$±0.005 | $76^b$±0.002 | *** |
| Phosphorus (g) | 30±0.013 | 31±0.002 | NS |

The values are means of six cockerels.
[a, b] Means in a row bearing different superscripts are significantly different.
*** p<0.001.

**Table 4.** True mineral availability (%) from vegetable and animal protein feedstuffs in caecectomized and intact cockerels

| Attributes | Caecectomized | Intact | Probability |
|---|---|---|---|
| Soybean meal | | | |
| Calcium | 36.24±1.07 | 39.92±2.09 | NS |
| Copper | $21.61$±$1.69^a$ | $13.94$±$1.95^b$ | * |
| Magnesium | 22.65±0.61 | 25.14±2.65 | NS |
| Manganese | 47.87±1.07 | 50.09±1.69 | NS |
| Phosphorus | 19.32±0.73 | 19.22±1.25 | NS |
| Rape seed meal | | | |
| Calcium | 31.43±0.97 | 33.33±1.57 | NS |
| Copper | 70.44±1.80 | 71.09±0.88 | NS |
| Magnesium | 23.70±1.05 | 24.03±1.10 | NS |
| Manganese | $36.23$±$1.14^b$ | $41.34$±$0.96^a$ | ** |
| Phosphorus | 15.11±1.38 | 15.57±1.93 | NS |
| Sesame meal | | | |
| Calcium | 32.21±1.35 | 32.58±1.20 | NS |
| Copper | 36.25±1.42 | 34.93±1.25 | NS |
| Magnesium | 26.63±1.93 | 26.69±2.29 | NS |
| Manganese | $37.36$±$0.91^b$ | $44.19$±$1.11^a$ | *** |
| Phosphorus | $30.19$±$1.72^a$ | $18.87$±$1.14^b$ | *** |
| Sunflower seed meal | | | |
| Calcium | 44.63±0.92 | 43.80±1.47 | NS |
| Copper | $28.98$±$1.72^b$ | $36.44$±$1.21^a$ | ** |
| Magnesium | -16.36±2.16 | -17.16±1.94 | NS |
| Manganese | $47.54$±$2.54^b$ | $64.27$±$1.39^a$ | *** |
| Phosphorus | 7.77±1.25 | 8.50±0.77 | NS |

The values are means of six cockerels.
[a, b] Means in a row bearing different superscripts are significantly different.
* p<0.05; ** p<0.01; *** p<0.001.

vegetable protein sources.

The excretion of metabolic origin minerals viz., copper, magnesium, manganese, calcium and phosphorus are presented in Table 3. The endogenous outputs of these minerals with the exception of phosphorus were significantly (p<0.001) higher in caecectomized cockerels than in normal roosters.

The bioavailabilities of minerals from various protein supplements are presented in Table 4. The average availability of calcium ranged from 31.43% to 44.63%. The mean copper availability from soybean was significantly (p<0.001) higher in caecectomized as compared to intact cockerels. On the contrary the utilization of copper from undecorticated sunflower meal was markedly (p<0.01) higher in intact birds than their caecectomized counterparts. No significant difference was observed between caecectomized and intact cockerels for magnesium availability from various protein supplements studied. The availability of manganese from almost all the protein supplements with the exception of soybean meal was significantly (p<0.01 to 0.001) higher in intact birds as compared to caecectomized ones. The average availability of phosphorus ranged between 7.77% and 30.19%. The utilization of phosphorus from sesame meal was significantly (p<0.01 to 0.001) higher in caecectomized as compared to intact cockerels.

## DISCUSSION

The endogenous output of copper, magnesium, manganese and calcium were significantly higher in caecectomized cockerels. It appeared that caeca might markedly reduce the excretion of minerals through absorption as observed in intact cockerels. Evidences existed on caecal absorption of calcium, magnesium and phosphorus in rats (Ohta et al., 1994 and 1997). The difference in the endogenous excretion was to the tune of 50; 60.45; 40.35 and 29.63 per cent for copper, magnesium, manganese and calcium respectively in caecectomized cockerels. These observations revealed that the caeca played a significant role in the absorption of minerals. However there was no significant difference in the excretion of metabolic origin phosphorus between caecectomized and intact cockerels. This was contradictory to the earlier reports (Ohta et al., 1994 and 1997) in rats. The present observation reveals that the caecal epithelium of avian digestive tract does not contribute significantly to the absorption of phosphorus.

The availability of calcium and magnesium in all the four vegetable protein meals was similar between caecectomized and intact cockerels. Several reports have revealed that caeca of rats play a significant role in calcium and magnesium absorption (Rayssiguier and Remesy, 1977; Younes et al., 1996; Ohta et al., 1997). Milinkovic et al. (2000 and 2001) have investigated *in vitro* on bags made of live epithelia of swine caeca and proximal colon and observed that the caecum and proximal colon represented important sites of magnesium absorption. However in the present study there was no difference in the utilization of calcium from vegetable protein meals. It may be attributed to the higher digestibility value of these oil meals. Better the solubility of minerals higher will be the bioavailability. The copper of soybean meal was utilized well by caecectomized birds, while in sunflower meal the copper availability was significantly higher in intact birds as compared to their respective counterparts. The higher availability in intact cockerels might be due to the copper absorbing capacity of caecal epithelia. On the contrary, Turner et al. (1987) reported in sheep that copper absorption took place through the abomasum, small intestine and colon, but not from the rumeno-reticulum or caecum. The higher crude fibre content of sunflower meal (25%) would have increased the passage rate, thereby reducing its retention time in the small intestine.

The availability of manganese from rapeseed, sesame and sunflower meal was significantly higher in intact as compared to caecectomized cockerels. Lack of reports in this aspect opens the avenue of further research. However, it can be inferred from the present finding that caeca played a pivotal role in manganese absorption. Though there was no difference in the phosphorus bioavailability from soybean, rapeseed and sunflower meal but the phosphorus of sesame meal was better utilized by caecectomized as compared to intact cockerels. It reflected that the solubility of phosphorus might be higher in sesame meal as compared to other vegetable protein meals.

Overall the caecectomized cockerels underestimated the bioavailability of copper in sunflower meal and manganese in almost all the test feedstuffs and overestimated the availability of copper in soybean and phosphorus in sesame meal. The present findings suggest that more studies on the role of caeca in mineral absorption, phosphorus absorption in particular have to be carried out. In a nutshell, it can be concluded that the caeca plays a pivotal role in the absorption of minerals *viz.*, calcium, phosphorus, magnesium, manganese and copper, which escapes digestion and absorption in the small and large intestinal segments.

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