# AVAILABILITY OF CALCIUM IN SOME OF THE LEAFY VEGETABLES

By S. C. Devadatta, F.A.Sc. and T. C. Appanna, M.Sc.
(*Christian Medical College, Vellore*)

Received June 26, 1953

In average diets a deficiency of calcium is more likely to occur than a deficiency of any other element. The usual diet of average people in India consists mainly of cereals supplemented with pulses and vegetables. Cereals and pulses although rich in phosphorus, are poor sources of calcium. A known adequate source of calcium would be the vegetables which supplement the cereals and the pulses in the dietary. Because of the richness of calcium in milk, nutritionists have classified this food as nearly indispensable in diet. But even small amounts of milk is beyond the means of the average Indian. The problem of finding some vegetables rich in calcium which would be well utilized in the system is of paramount importance. The present investigation deals with the availability of calcium of some vegetables rich in this element which grow in abundance. Numerous studies have shown that mere measurement of the calcium content of a food is no indication of its value as a dietary source of this element. The true value depends on whether the body can utilize the calcium present in it. The classic example of this is the well-known distinction between the high calcium content of spinach and its complete unavailability to the body because of the presence of oxalic acid as demonstrated by Fincke and Sherman (1935). Rose (1920) studied the utilization of calcium of carrots with healthy young women and concluded that the calcium of carrots was nearly as well utilized as that of milk. Rose and McLeod (1923) made similar study of the utilization of calcium of almonds. When almonds furnished about 73% of the total food calcium, the calcium of the almond was almost as well utilized as that of milk, but when the almonds was made to furnish about 85% of the total food calcium, the efficiency of utilization of calcium of almond was somewhat less.

Fincke and Sherman (*loc. cit.*) made experiments with animals so that the retention of calcium could be determined by the direct analysis of the body at the end of the experiment. Rats were chosen on account of their omnivorous habit and the chemistry of their nutrition is in most respects so closely like those of human beings. The experimental animals were fed on a carefully planned diet through a period during which normal growth

and development involves a relatively large increase in the calcium content of the body. It was found by them that calcium of milk was well utilized, that of kale almost as well, while that of spinach was poorly utilized. Similar investigations were carried by Kao, Conner and Sherman (1938) and M. Speirs (1939).

## Experimental

In our experiments the procedure adopted was essentially that developed by Fincke and Sherman (*loc. cit.*) in their work on the availability of calcium in spinach and kale. It was planned to determine the utilization of calcium of five Indian leafy vegetables commonly consumed by the people, *viz.*, Avati Keerai (*Sesbania grandiflora*), Mola Keerai (*Amaranthus gangeticus*), Chiru Keerai (*Amaranthus spinosus*), Karuvepilai (*Murraya kœnigii*) and Murunga Keerai (*Moringa oleifera*). The vegetables were purchased from the local market, thoroughly washed with distilled water and dried in an oven at a temperature of about 75° C. to avoid charring. The dried vegetables were thoroughly ground, mixed and bottled and stored in refrigerator. Milk was used in the form of skimmed milk powder and the same lot was used throughout the investigation. Butterfat in the form of butter purchased from local market was separately added.

The powdered dry leafy vegetables and the skimmed milk powder were analysed. The results are shown in Table I.

Table I

*Analysis of Skimmed Milk and Leafy Vegetables (Dry Powder)*

| Composition | Skimmed Milk % | Avati Keerai % | Mola Keerai % | Chiru Keerai % | Karuvepilai % | Murunga Keerai % |
|---|---|---|---|---|---|---|
| Calcium .. | 1·460 | 3·99 | 3·18 | 3·56 | 2·40 | 1·56 |
| Phosphorus .. | 0·974 | 0·308 | 0·69 | 0·59 | 1·15 | 0·31 |
| Protein (N×6·25) .. | 39·85 | 29·12 | 33·46 | 31·89 | 17·56 | 25·47 |
| Oxalic Acid .. | .. | 0·021 | 0·047 | 0·053 | 0·058 | 0·047 |

In all cases calcium was determined by the McCrudden method (1911–12) by precipitating calcium as oxalate at pH 4·8 to 5·2 using methyl red as indicator and titrating the oxalic acid with potassium permanganate solution. Phosphorus was determined by the Brigg's (1922) method. Oxalic

acid was determined by precipitation with calcium chloride in presence of acetic acid and subsequent titration with potassium permanganate solution as described by Mazumdar and De (1938).

Healthy young albino rats 28 days old reared on stock diet consisting of $\frac{2}{3}$ whole wheat, $\frac{1}{2}$ whole milk powder, vegetables with bi-weekly addition of vitamins A and D, which is known to support normal growth and calcification, were used as experimental animals. The experimental animals were put in individual iron cages.

The experimental diets were so prepared and planned that they contained almost the same percentage of calcium and phosphorus and protein. The calcium phosphorus ratio was nearly always 1 : 1. In the control diet practically all the calcium was supplied by the skimmed milk powder. In the diets containing greens, half the skimmed milk powder was replaced by enough of the green to supply an equal amount of calcium, the difference in weight being made up with maize starch.

TABLE II

*Composition of Diet and their Ca and P Contents*

| | Control Skimmed *Milk Diet* | Avati Keerai *Diet* 1 | Mola Kerai *Diet* 2 | Chiru Keerai *Diet* 3 | Karuvepilai *Diet* 4 | Murunga Keerai *Diet* 5 |
|---|---|---|---|---|---|---|
| Whole Wheat .. | 65 | 65 | 65 | 65 | 65 | 65 |
| Butterfat .. | 10 | 10 | 10 | 10 | 10 | 10 |
| Skimmed Milk Powder .. | 22 | 11 | 11 | 11 | 11 | 11 |
| Greens Powder .. | .. | 4·0 | 5·0 | 4·5 | 6·5 | 11·0 |
| Salt Mixture .. | 5 | 5 | 5 | 5 | 5 | 5 |
| Maize Starch .. | 3 | 9 | 8 | 8 | 7 | 3 |
| Calcium % .. | 0·341 | 0·327 | 0·331 | 0·338 | 0·345 | 0·319 |
| Phosphorus % .. | 0·420 | 0·330 | 0·351 | 0·314 | 0·309 | 0·381 |

The animals were so chosen that the initial weights of the groups of rats to be compared were approximately the same. Food and water was supplied *ad libitum* and records were kept for the food consumption. The experimental animals were fed the experimental diets from 28 to 60 days of age, a period during which the rate of growth and calcification is very rapid.

TABLE III

*Average Growth of Rats from 28 days to 60 days on Different Diets*

| Diets | No. giving Average rats | Average initial weight of body | Average final body weight net | Average net gain | Average food intake | Average gain per gm. of food |
|---|---|---|---|---|---|---|
| | | gm. | gm. | gm. | gm. | gm. |
| Control | 4 | 41·5 | 112 | 72 | 195 | 0·36 |
| 1 | 6 | 38 | 97 | 59 | 208 | 0·28 |
| 2 | 5 | 34 | 94 | 60 | 190 | 0·31 |
| 3 | 5 | 37 | 93 | 56 | 192 | 0·29 |
| 4 | 6 | 33 | 74 | 41 | 204 | 0·20 |
| 5 | 5 | 38 | 84 | 46 | 194 | 0·23 |

At the end of the period the rats were weighed and killed with chloroform. The gastro-intestinal tract was dissected and the contents removed. The weight of the contents subtracted from the final live weight, gave the net weight which was used for calculation of the percentage of body calcium of 60 days old rats. The alimentary tract was discarded as the calcium content in the walls of intestinal tract was negligible. The rats were ashed in silica basins and finally ashed at dull red heat. The ash was dissolved in 1 : 4 hydrochloric acid, filtered and diluted to 500 c.c. Aliquots of this solution were analysed for calcium by the method already stated.

The results are summarized in Tables IV and V. The bodies of the albino rats on the control diet, diet 1, diet 2, diet 3, diet 4 and diet 5 contained respectively 0·764, 0·820, 0·787, 0·807, 0·906 and 0·810 per cent. of calcium. It will be noticed that the rats on the milk diet contained the lowest and those on diet 4 the highest. This is to be expected since the rats on milk diet grew faster than those on the diet 4. The intermediate values for the percentage of body calcium of rats on diet 1, diet 2, diet 3 and diet 5 were also conforming with their growth rates.

To obtain the storage of calcium it was necessary to know the initial content of calcium of the body of the animal just before the beginning of

TABLE IV

*Calcium Content of 60 days old Rats fed on Diets containing the same amount of Calcium from Milk or Vegetables*

| Diets | No. giving Average rats | Average net body weight (gm.) | Average Calcium intake (gm.) | Average total body calcium (gm.) | Average % body calcium |
|---|---|---|---|---|---|
| Control | 4 | 112 | 0·625 | 0·856 | 0·764 |
| 1 | 6 | 97 | 0·690 | 0·798 | 0·820 |
| 2 | 5 | 94 | 0·650 | 0·741 | 0·787 |
| 3 | 5 | 93 | 0·654 | 0·751 | 0·807 |
| 4 | 6 | 74 | 0·710 | 0·671 | 0·906 |
| 5 | 5 | 84 | 0·620 | 0·681 | 0·810 |

TABLE V

*Calcium Content of 60 days old Rats on Various Diets and thier Utilization Factor*

| Diets | No. giving average rats | Calcium in body at 60 days | | Calcium at 28 days (gm.) | Calcium retained in body (gm.) | Calcium in food eaten (gm.) | Utilization factor |
|---|---|---|---|---|---|---|---|
| | | Total (gm.) | % net wt. | | | | |
| Control | 4 | 0·856 | 0·764 | 0·223 | 0·533 | 0·625 | 0·85 |
| 1 | 6 | 0·798 | 0·820 | 0·283 | 0·515 | 0·690 | 0·74 |
| 2 | 5 | 0·741 | 0·787 | 0·226 | 0·515 | 0·650 | 0·78 |
| 3 | 5 | 0·751 | 0·807 | 0·200 | 0·521 | 0·654 | 0·79 |
| 4 | 6 | 0·671 | 0·906 | 0·286 | 0·305 | 0·710 | 0·54 |
| 5 | 5 | 0·681 | 0·810 | 0·280 | 0·401 | 0·620 | 0·69 |

the experiment. This was obtained by determining the per cent. calcium of the body at 28 days of age.

For an exact basis of comparison a " calcium utilization factor " was calculated for each of the diets. This was obtained by dividing the calcium retained by the calcium intake. The results are given in Table V. The difference of the utilization factor of calcium of diet 4 from that of milk is 0·310. This value is quite significant and therefore the calcium of diet 4 is not quite as well utilized as that of milk. For diet 5 the difference for

utilization factor is 0·160 which is not very far from the figure 0·11 for diet 1. Nevertheless these two vegetables, Karuvepilai and Murunga Keerai serve as a fairly good source of calcium. On the diet 1 the difference of calcium utilization factor from that of milk is 0·11. This figure is not very significant and it appears that the calcium of diet 1 is almost as well utilized as that of milk.

The two species of Amaranthus, Mola Keerai and Chiru Keerai, included in the diets 2 and 3 respectively contain a high amount of calcium, 0·54% and 0·47% respectively. In spite of the fairly high percentage of oxalic acid, these are well utilized and the utilization factor being 0·78 and 0·79 respectively. With a difference of 0·07 and 0·06 from that of the utilization factor of milk at 0·85, they are as well utilized as that of milk.

SUMMARY

Healthy young rats, 28 days old, were placed on six diets in one of which all the calcium was supplied entirely by skimmed milk. In the other diets half of the skimmed milk was replaced by enough ground dried leafy vegetables to provide the same amount of calcium as in the milk diet. At 60 days of age the animals were killed and their bodies analysed for calcium. Comparison of the availability of calcium in these vegetables with that of milk was made by calculating for each an utilization factor which is the ratio of calcium retention to intake. The values for the six diets were: 0·85 for milk diet, 0·74 for diet 1, 0·78 for diet 2, 0·79 for diet 3, 0·54 for diet 4, and 0.69 for diet 5 respectively.

All the five leafy vegetables, *viz.*, Avati Keerai (*Sesbania grandiflora*), Mola Keerai (*Amaranthus gangeticus*), Chiru Keerai (*Amaranthus spinosus*), Curry leaves (*Murraya Kænigii*), Murunga Keerai (*Moringa oleifera*) form good sources of calcium from the point of nutrition, especially Avati Keerai, Mola Keerai and Chiru Keerai, as the calcium in these are used as well as that in milk.

*Names of the Vegetables and Equivalents in other Important Indian Languages*

| | *Botanical Name* | Tamil | Kannada | Hindustani |
|---|---|---|---|---|
| 1. | *Sesbania grandiflora* .. | Avati Keerai | Agase | Agasthi |
| 2. | *Amaranthus gangeticus* .. | Mola Keerai | Yeledantu | Lal Choalai |
| 3. | *Amaranthus spinosus* .. | Chiru Keerai | .. | .. |
| 4. | *Murraya kænigii* .. | Karuvepilai | Karibevu | Gandhela |
| 5. | *Moringa oleifera* .. | Murunga Keerai | Murige | Saijan |

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