EVALUATION OF CALCIUM AND IRON SUPPLEMENTS TO PRESCHOOL CHILDREN

By

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A Thesis Submitted to the University of Madras in Partial Fulfilment of the Requirements for the Degree of Master of Science,
March, 1975.

ACKNOWLEDGEMENT

The author wishes to express her deepfelt gratitude and thanks to Dr.(Mrs.) Rajammal P. Devadas, M.A., M.Sc., Ph.D. (Ohio State), Principal, Sri Avinashilingam Home Science College for Women, Coimbatore, for her inspiring guidance during the entire course of this study. The author records her sincere thanks to Tmt. Nirmala Krishnamurthy, M.A., M.S.(Iowa), Reader in Biochemistry, Sri Avinashilingam Home Science College for Women, Coimbatore, for her valuable advice and help.

Grateful acknowledgement are due to the children and their mothers for their full co-operation and participation in this study.

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I. INTRODUCTION

The nutritional needs of children are critically important during the period 0-6 years. The preschoolers are more vulnerable and comparatively less attended than other groups of children (Devadas 1971).

A review of the status and problems of the preschool child in India may well begin with a statement of the quantitative dimensions of the problem. There are 110 million children below the age of six in India, that is about 20 per cent of the total population in the country (India 1973). The net addition to the population of pre school children is of the order of 20 million per year.

The first problem of the pre school child in India is to survive. 40% of all deaths in the country still take place among children below the age of five, and the death rate among pre school children is 21-35 per thousand (Mina Swaminathan, 1973). It is estimated that 75% of the child population can be classified as 'not healthy', due to major and minor illness (Gopalan, 1975).

The major causes of morbidity and mortality are gastrointestinal complaints and respiratory infections and

Nutritional disorders of various types. In order to promote normal growth it becomes necessary to identify dietary deficiencies that currently impose constraints on normal growth and development in our children.

Gopalan and Narasinga Rao (1971) have collected data in India on the dietaries of Pre school children. Results revealed that in the current dietaries of our pre-school children the major bottleneck iscalories and not proteins. If the children were to get greater quantity of the same food in amounts that would meet their calorie requirement, their protein needs would be met. It must, however, be emphasised that the existing diets of children were deficient in several minerals and vitamins also. The diets were low in riboflavin,Vitamin C, iron, calcium and vitamin A.

Besides the main problems of shortage of calories and proteins, anemia due to iron deficiency, growth retardation due to Calcium deficiency and blindness due to vitamin A deficiency are serious problems among Indian pre-school children (Gopalan, 1975).

According to Dhar et al (1969) iron deficiency Anemia is the most prevalent type of anemia in the world, particularly in a developing country like India. Various factors combine to determine the high incidence, such as maternal illhealth, undernutrition, faulty feeding, knowledge with inadequate feeding supervision and prolonged breast feeding for economic reasons or superstitions. Surveys in Indian population is around 50%, of whom 90% have iron deficiency. Sood (1967) conducted a study on children who were admitted to the paediatric service of AIIMS hospital, 90% of these children between 6-36 months, showed evidence of iron deficiency.

Radiological appearance suggesting generalised osteoporosis is frequently seen in the bones of children suffering from protein calorie malnutrition. This has generally been attributed to protein deficiency perse. It is however, possible that prolonged calcium deficiency as a result of inadequate dietary intake of Calcium by these children may be a contributory factor (ICMR, 1968). Rajalakshmi et al (1973) suggest that Calcium deficiency is a crucial factor in the aetiology of skeletal retardation in young children.

Studies carried out by NIN showed that the existing diets of pre school children are deficient in several minerals and vitamins. The average intake of iron by the pre school children was 5-9 mg/day against 12-20, and calcium was 193 mg/day against 400-500 recommended allowances ICMR, 1968).

Thus along with an increase in the calories and protein content of the diets through the addition of cereals, and lgumes qualitative improvements such as addition of vitamins, minerals are also needed in the diet of pre school children.

Children who were growing and forming new bones and teeth need more calcium than adult. If their diet does not provide enough the result is reduced growth. It is the consensus of many nutritionists today that enough calcium must be provided to young children to ensure a well clacified skeleton when adulthood is attained; (FAO/WHO, 1962). Thus besides calories and protein, vitamin A, calcium and iron may be the other biggest constraints in the diets of our pre-school children. The present study is an evaluation of the supplement of two of these nutrients namely calcium and iron along with additional food sources of calories and protein to a group of pre school children.

An earlier study was carried out in this laboratory (Devadas, 1974) to evaluate the growth promoting ability of opaque-2 maize, ordinary maize and skimmed milk with selected groups of the pre-school children. The supplementary diets used in this study were isocaloric (450) and isoprotein diets (10 g). The results showed that the growth rate of target children fed with opaque-2 maize diet ranked next to skimmed milk diet. An examination of the nutrient content of the diets revealed that the skimmed milk diet provided an excess amount of calcium than the opaque 2 maize diet while all the other nutrient contents were similar. It was felt that the growth promoting ability of skimmed milk diet may be due to its better quality protein or perhaps due to higher calcium content since all the other nutrients were equal and appeared to be adequate in the two experimental diets.

The present study "Evaluation of Calcium and iron supplements to pre school children" was carried out to answer the above specific question whether the better growth promoting ability of skimmed milk diet is due to better quality protein or due to higher calcium content when compared with the opaque 2 maize diet. Thus the main objective was to find out the effect of calcium supplementation with opaque 2 maize diet in comparison with skimmed milk diet, when fed to pre-school children for a period of six months.

Apart from growth retardation, iron deficiency anaemia was prevalent among the target children. (Devadas et al. 1974). The target children consumed a total of 10 mg of iron/day from their home diets and from the experimental diets which is incorporated with leafy vegetables. But inspite of this apparently satisfactory intake, iron deficiency anaemia was found to be very severe among these children. To over come this deficiency prophylactic iron (60 mg.) was supplemented to the target children in this study.

The indices used for the evaluation of calories, protein, calcium and iron supplements were growth rate as indicated by the monthly body weights, lengths, measurements of chest, arm and head circumference for a period of six months, calcium balance, skeletal status as evidenced by xray photographs, haemoglobin levels and food and faecal iron analysis in a sub sample.

II. REVIEW OF LITERATURE

Nutritional disorders prevalent among Indian children:

Extensive studies carried out by Gopalan and Narasingharao (1971) and Devadas et al (1972) in India indicated that among the children of poor segments of our population there is considerable degree of growth retardation.

Gopalan (1974) points out that in our country to day the incidence of severe forms of protein calorie Malnutrition like kwashiorkor and marasmus, has been estimated to be around one to two percent of all children between the ages of 1 and 5 years. This figure does not include the mild and moderate forms of malnutrition, nearly 80 per cent of pre-school children are suffering from I and IInd degree mal-nutrition.

Apart from protein-calorie malnutrition, vitamin A deficiency is another major nutritional problem contributing to a considerable amount of preventable blindness. It is believed that in India alone, these may be over one million cases of blindness arising from vitamin A deficiency (Gopalan, 1975). Anaemia due to iron deficiency ranks as the next nutritional deficiency.

Prevalence and causes of calcium deficiency:

Radiological appearance suggesting generalised osteoporosis is frequently seen in the bones of children, suffering from protein calorie malnutrition. This has generally been attributed to protein deficiency perse. It is however, possible that prolonged calcium deficiency as a result of inadequate dietary intake of calcium by these children may be a contributary factor (ICMR, 1968).

Eight children with signs of protein calorie malnutrition, whose ages ranged from 1 to 4 years were studied. (ICMR 1968). Five were marasmic children and the other three were suffering from marasmic Kwashiorkor with minimal oedema. The calcium infusion test was done on the second day of admission in the first five children and on the fourth day in the oedematous children after the oedema had completely disappeared. Two normal children of the same age group were also studied for comparison.

Twenty four hour urine collections were made for the estimation of basal excretion of calcium and on the next day 15 mg/kg of calcium in the form of calcium gluconate in 300 ml of normal saline, was given intravenously over a period of 4 hours. The amount of calcium excreted in the urine during these 4 hours of infusion was determined and the amount of of calcium retained calculated, as a part of the infused calcium. The results of the study showed that basal excretion of calcium average 18.0mg/24 hrs. (Range 10-40). 85% of the infused calcium was retained in the malnourished children.

These results suggest that in children suffering from protein caloric malnutrition, who show radiological evidence of osteoporosis, the capacity of bones to retain infused calcium is not impaired, and that, there alterations in bone matrix may not be responsible for osteoporosis. It may be mentioned here, however, that while the 4 hour infusion test may reveal the extent of calcium retention, this may not necessarily indicate mineralisation of osteoid tissue.

Vitamin D and proteins assists in the absorption of calcium. Phytic acid, fats, and oxalic acid inhibit the absorption of calcium (Davidson and Passmore 1973).

Effect of vitamin D supplements on Calcium absorption in Children:

Children of the poor communities depend almost entirely on sunshine for their vitamin D and get very little of it in their diets. In a study conducted by Perera and Vinodini Reddy (1971), it was observed that on diets similar to those they consume at home the absorption of calcium was only about 20% of the intake. When the diet was supplemented with 200 I.u. of vitamin D daily 5 of the 6 children showed considerably increased absorption. This would suggest that though there was no biochemical evidence of vitamin D deficiency, the absorption of calcium in these children was perhaps being limited by inadequacy of vitamin D. When the level of vitamin D was further increased in these children aged between 2-5 years to 400 I.U., there was no appreciable change in calcium absorption in these children. Thus indicating that maximal absorption was occuring at the level of 200 I.U. of vitamin D. The daily retention of calcium required for normal skeletal growth throughout childhood hasbeen estimated to the about 100 mg. a level that was achieved in the above study with the use of 200 I.U. of vitamin D daily.

<u>Effect of phytate on the absorption of calcium:</u>

According to Dent <u>et al</u> (1973) ultra violet irradiation of a 14 year old boy who developed rickets while consuming a diet high in phytate resulted in a prompt rise in serum calcium and 25 hydroxy cholecalciferol, a fall in serum parathyroid hormone level and improvement in calcium balance, and cure of rickets while he was still consuming the high phytate diet.

Nutritional rickets is reported in a 15 year old Indian boy. His diet before admission was estimated to contain 1200 mg calcium, 1.5 ug. vitamin D and 230 mg. phytic acid per day. He was put on a diet with low phytic acid content but with a comparable amount of vitamin D. On this diet, the plasma calcium returned to normal over four weeks and rickets healed (Wills et al 1972).

Dietary intake of calcium:

Diet surveys carried out in India (Mitra 1953) have shown that the average intake of calcium is in the neighbourhood of 469 mg. percapita. More than 60 per cent of this calcium is derived from cereals and other vegetable foods. Further, availability of calcium in such foods may be limited by associated factors like phytate, oxalic acid and high phosphorus content. Average intake of calcium of the predominantly rice eating population may be lower than the above figure. The only group which consumes a high level of dietary calcium is the one whose stapple cereal is ragi which is known for its high calcium content. Though the average intake of calcium by Indians is not more than 500 mg. per day, there is no evidence of apparent ill effects attributable to calcium deficiency. Analysis of skeletons of Indians by Shenolikar (1966) has shown that their bones had normal calcium concentration.

Human Metabolic Studies with reference to calcium:

A few calcium metabolic studies carried out in Indian adults, also indicated that calcium balance is achieved at relatively low intakes. Basu and his co-workers (Basu et al 1939; Basu and Ghosh 1943; Basu and De, 1948; Basu and Nath, 1946) have carried out calcium metabolism studies in Indian male adults on low calcium intakes roughly corresponding to intakes on their normal diets. They consider from one of their studies that 388 mg. per day may be the maintenance requirement of calcium for Indians. From other studies of theirs, it can be seen that positive calcium balance could be achieved at intakes ranging from 177 to 485 mg/day.

Calcium balance studies carried out at the nutrition Research Laboratories (Shenolikar, 1968), also indicated that calcium balance can be achieved in Indian subjects on low intakes. Calcium balance study was carried out in four female and three male adult subjects on intakes corresponding to their normal intakes. All the four women with calcium intakes in the range of 386-448 mg./day were in positive balance. Of the three men studied, two were in positive balance on calcium intake of 500 mg./day and one was in negative balance on a similar intake. These balance studies, not with standing their limitations, indicate that the Indian adult can be in calcium balance on intake ranging from 250-500 mg. Adult women appear to be better adapted to lower intakes.

Kamalanathan *et al* (1965) conducted a study on the effect of supplementing a poor Indian rice diet with 100 g. of wild green leafy vegetables (providing about 480 mg.) calcium) or with 35 g. skimmed milk powder (providing about 480 mg. calcium) on the metabolism of nitrogen, calcium and phosphorus in six adolescent girls aged 19-21 years.

The intake and retention of calcium on the different diets were as follows: Basal rice diet (period I) 217.8 mg. and 2.6 mg; Experimental diet containing green leafy vegetables (period II) 702.9 mg. and 307.8 mg; Basal rice diet (period III) 217.6 mg. and 7.2 mg. and Experimental diet containing skimmed milk powder (period IV) 704.6 mg. and 396.1 mg. respectively.

Effect of calcium supplementation on the growth of children:

Diet surveys indicate that the average calcium intake of children in India is 300 mg. or less. However, Kurian *et al* (1960) have shown that children (10-11 years) can be in positive balance of 120 mg. per day even on low intakes of 354 mg. per day. The diet survey data would indicate that calcium intakes of Indian children are sub-normal and the stature and growth of these children are also substandard.

Attempts to demonstrate that subnormal growth of Indian children is due to low calcium intake have not yielded conclusive results. Aykroyd and Krishnan (1938, 1939) demonstrated that calcium lactate supplementation (0.5-1.0 gm per day) to a group of children aged 2½ - 7 years, with sub-normal dietary calcium intakes of 310 mg. a day, resulted in an improved growth. However, in a recent study by Bhansal et al (1964) at the Nutrition Research laboratories, supplementation of calcium glycerophosphate (2.5 g. to children below 1 year and 5 g. for those above 1 year) for a period of forty-two weeks to a group of children of 6-30 months in a rural community, with calcium intakes of 178-85 mg. per day, on their usual diets, did not bring about any improvement in growth. These studies thus leave the question of calcium requirement of Indian children still open.

A study was conducted by Venkatachalam (1971) with 38 children age ranging from 6 to 30 months (14 males 24 females). Calcium glycerophosphate in syrup was given. Control group was given syrup only. The study was conducted for 42 weeks. Initial weight, height, chest and head circumference were noted. Radiometry was done for the length and area of radius, ulna and III metacarpal, number and size of ossification centres. The results did not show any significant difference between the two groups. Also, there is increase in the values for the various other measurements taken on 12th, 25th and 42nd weeks did not show any significant difference between the control and the supplementary group suggesting that the calcium supplementation had not provided the children with any benefit with respect to growth. Also, the experimental and control group children did not show any difference with regard to the incidence of common illness.

Since the calorie and protein intake was low, the question arose whether the lack of beneficial effects of calcium supplementation in these children consuming a low calcium diet was due to the inadequacy of the other nutrients in the diet. Therefore the growth studies could become a useful method for the assessment of adequacy of calcium but only when the other dietary factors and non-dietary influences are kept under careful control.

A study was carried out at Baroda by Rajalakshmi et al (1973) on "The effects of supplements varying in carotene and calcium contents on the physical, biochemical and skeletal status of pre-school children". Studies were made of the effects of food supplements varying in carotene and calcium content on the nutritional status of children aged 2-6 years. Three groups of children who given a dietary supplement for six months and a fourth group of controls were investigated.

The supplement was the fermented food dhokla, made of wheat and bengal gram (Group I) or dhokla with added greens (Group 2) or dhokla with added greens and lime powder (Group 3).

Studies were made of effects of the above supplements on height, weight, skeletal status as judged by increments in bone age, and cortical thickness of the 2nd metacarpal bone and femur, and biochemical status as judged by the composition of blood (Haemoglobin) serum (Protein, albumin and vitamin A) and urine (creatinine, nitrogen, thiamine, riboflavin, and vitamin C).

All the groups given the supplements were found to be superior to the controls as judged by these criteria. The size of the experiment generally increased from Group I to Group 3 and the difference were most marked for bone age and cortical thickness.

The results suggested that calcium deficiency is a crucial factor in the aetiology of skeletal retardation in young children and that at the levels of calcium (560 mg) and phosphorus (980 mg) consumed resulted in a significantly higher growth rate.

<u>Prevalence and causes of anaemia in children:</u>

Anaemia due to the deficiency of iron was prevalent in kiryat shmonah, an upper Galillee community. Complete hematological examinations were performed by Levy et al (1970) in 247 children in Kiryatshmoneh, anaemia was found in 52 per cent of 247 healthy children aged 1 day to 6 years. The prevalence of the anaemia and of the iron and folate deficiencies rose from birth to the age of 2-3 years, after which it declined gradually.

The anaemia in children born to anaemic mothers was much more frequent than among those delivered by non-anaemic women. Poor iron and folate stores at birth, malnutrition and frequent gastrointestinal infections may have contributed to the wide spread deficiency state observed in this group of children.

According to Dhar et al (1969) iron deficiency anaemia is the most prevalent type of anaemia in the world, particularly in a developing country like India. Various factors combine to determine the high incidence, such as, maternal ill health, under nutrition, faulty feeding, knowledge with inadequate feeding supervision and prolonged breast feeding for economic reasons or superstitions. Surveys in Indian population is around 50 per cent, of whom 90 per cent have iron deficiency.

It is a common knowledge that anaemia in infancy is extremely prevalent in the developing countries. There are a large number of reports from India Pointing to this high prevalence rate. Sood (1967) conducted a study on a group of 377 children of all ages admitted to the paediatric service of AIIMS hospital. 79 per cent of these children had haemoglobin less than 10 g. per cent. 45 of these anaemic children, between 6-36 months, were studied indetail, and 90 per cent of them showed evidence of iron deficiency.

Besides these factors like increased demands of growth, status of iron stores at birth and maternal nutrition, chronic blood loss plays an important role in the aetiogenesis of iron deficiency anaemia. It is known that a number of children are prone to gastrointestinal bleeding. Hookworm in festation has a major role to play in causation of anaemia in many countries. In India also hookworm infestation is quite prevalent in the rural areas and thus may in part be responsible for the high frequency of iron deficiency in this country.

Intake of Dietary Iron by Indians:

Results of diet surveys in India (Ramalingaswami and Patwardhan, 1949; Mitra, 1953) indicate that the average intake of iron is 30 mg. per day per consumption unit, the range being 6 to 10.9 mg. Diet survey results suggest that 6 per cent of the average families studied had dietary iron intakes less than 15 mg. and 10 per cent less than 20 mg. To this, one should add iron derived from cooking vessels.

Studies at the nutrition research laboratories have shown that a significant amount of iron can be derived from this source. Not withstanding the uncertainties of iron intake figures obtained from diet surveys, it is evident that the average dietary intake of iron by Indians is by no means low. It is surprising, therefore, that iron deficiency anaemia should be so widely prevalent. Indeed the results of duplicate meal analysis of diets of anaemic subjects, carried out at the nutrition Research Laboratories, have shown that nearly half the subjects has marginal or low intakes while the other half the had intakes which were quite high compared to the western recommended allowance. It would, therefore, appear that there are other factors like poor absorption of dietary iron or excessive loss of body iron operating under these conditions.

Iron absorption:

Several iron balance studies in healthy Indian adults, consuming different types of diets, have been carried out employing chemical balance method. Apart from determining iron absorption from these diets, effects of phytate, calcium and ascorbic acid on absorption of iron from the above dietaries have also been determined.

De (1950) reported that the iron retention was 5.86 mg. when iron intake was 24.84 mg. It was observed by Hussain and Patwardhan (1959) that 20 per cent of iorn was absorbed from a well balanced diet based on rice or wheat containing 38.44 mg. iron.

Cereals, which form a major part of the Indian dietaries, contain phytate which interferes with iron absorption. Hussain and Patwardhan (1959 b) have shown that in balanced diets containing 8 and 40 per cent phytate, iron absorption was markedly reduced to 11 and 3 per cent respectively.

In another experiment, Apte and Venkatachalam (1962) showed that an intake of about 20 mg. of dietary iron was necessary to result in any significant absorption of iron when the diet contained cereals with 40 per cent phytate. It is known that dietary calcium combines with phytate.

It was shown by Apte and Venkatachalam (1964) that the iron absorption from diets, containing phytate, could be increased by increasing the dietary calcium. Iron absorption from such a diet, containing 1500 mg. calcium, was found to be maximal. Ascorbic acid also has been shown to improve iron absorption by these workers (Apte and Venkatachalam, 1965). It would appear from these studies that though iron absorption is decreased by phytate in cereals and other foods, liberal amounts of calcium and ascorbic acid in the diet seem to counteract the phytate effect. However, diets of poorer segments of population in India not only are rich in cereals containing phytate but are poor in calcium and ascorbic acid and hence iron absorption may be expected to be poor in these diets. Chemical balance studies, employing such diets, gave values for iron absorption ranging from 7 per cent to 28 per cent with an average of 13 per cent (Apte and Venkatachalam, 1965).

Effect of iron supplementation on the nutritional status of children:

Devadas *et al* (1971) conducted a study with 60 children receiving school lunch. These children were supplied with 25 g. greens per day per child. Another group of 60 children, comparable in age and socio-economic status and participating in the school lunch served as controls.

The criteria used for the evaluation of the nutritional status were height, weight, haemoglobin value and clinical condition of the children. After six months of the experimental period, the nutritional status of children receiving greens had improved significantly over the other groups, particularly with regard to the clinical picture.

Devadas *et al* (1973) conducted another study in which 105 children participated in a school lunch programme. They were divided in to three groups of 35 children each comparable in age, height, weight and haemoglobin levels. Group I control group was a comparable group of 35 children who were not participating in the school lunch, Group II was given the basal school lunch while groups III and IV were given 37.5 g. amaranthus cooked in aluminium and iron utensils respectively along with the school lunch for a period of seven months.

It was observed that the children receiving amaranthus cooked in iron utensil had improved their nutritional status significantly over the other groups, particularly with regard to the haemoglobin levels.

III. EXPERIMENTAL PROCEDURE

The present study namely "Evaluation of calcium and iron supplements to a selected groups of pre-school children" was carried out on young infants of the age 18-30 months of age, at Coimbatore District.

The specific objectives are:

To compare the growth and nutritional status of children consuming calcium and iron supplemented opaque 2 maize diet with the growth and nutritional status of children who were fed diets with ordinary maize, skimmed milk diet and opaque 2 maize diet.

To assess the nutritional status in terms of their monthly weight, height, measurements of chest, arm and head circumference for a period of six months.

The experimental diets namely opaque 2 maize, ordinary maize and skimmed milk provided equal quantities of protein and calories. But the calcium content of the maize diets were only 123 mg. per day per child whereas skimmed milk diet provided 456.6 mg. per day per child which resulted in a difference of 332.2 mg. of calcium per day per child. In-order to make the calcium content of the skimmed milk diet and opaque 2 maize diets similar, 319 mg. of lime powder was supplemented to opaque 2 maize diet which would provide 332 mg. of calcium per day per child.

Nutritional anaemia is one of the major health problems in India and is particularly seen among pre-school children. (Gopalan et al, 1971). To overcome this deficiency, 60 mg. of iron in the form of ferrous sulfate per child per day were also fed. An excess amount of iron was provided due to poor efficiency of absorption.

Thus 25 children (Group A) were given the ordinary maize diet, 25 children in (Group B) were given opaque 2 maize diet, 25 children in (Group C) were given calcium and iron supplemented opaque 2 maize diet, and 25 children in (Group D) received the skimmed milk diet. A smilar group of 25 children (Group E), of the same age, sex and of the same socio-economic background served as controls.

Anthropometric measurements, clinical and biochemical examinations were used as the criteria for the evaluation of calcium and iron supplements.

Selection of the villages:

Four villages, namely Jothipuram, Periannaickenpalayam, Venkatapuram and Goundampalayam in Periannaickenpalayam block in Coimbatore District, were selected for the study because of the good rapport already established in these villages. There were no other feeding or research projects currently in operation in those villages.

Socio-economic Survey:

All the families in the villages were enumerated and a socio-economic survey was done using the questionnaire (Annexure X).

Selection and grouping of children:

One hundred and twenty five children (18-30 months of age) belonging to the poor socio-economic status were selected for this study from Jothipuram, Perianaickenpalaya, Venkatapuram and Goundampalayam villages. This age group was selected because of the rapid growth at this age and need for calorie, protein and mineral supplementation at this age. The target children were divided in to five groups of 25 children each according to age, sex, parity and anthropometric measurements.

Conducting the Bench mark survey:

Base line information on the dietary intake and food practices of the children in the five groups was collected inorder to plan and proceed on the feeding trial.

Food weighment survey:

Since food weighment surveys are the most accurate for fact finding, a three day food weighment was conducted to assess the home food consumption pattern of the children and to assess their nutritional status. Five children from each of the experimental groups (A,B,C,D) and five children from the control group (E) were selected randomly on the basis of the willingness of their parents to co-operate with the investigator. All the raw foods used and their cooked preparations were weighed with the lever balance up to the accuracy of 1g. using the preforma given in (Annexure II).

The individual consumption of the foods by the target children was weighed and corrections were made for the plate wastage and in between consumption of snacks. Thus nutrient intake of the children for the three days was calculated.

Composition and Nutritive value of the supplement:

Table I gives the composition and nutritive value of the diets and supplements used in this study.

TABLE I

COMPOSITION AND NUTRITIVE VALUE OF THE SUPPLEMENT USED (QUANTITY/CHILD/DAY)

Groups	Ingreidients	Quantity g.	Calories	Protein g.	Calcium mg.	Iron mg.	B-carotene mg.	Thiamine mg.	Riboflavin mg.	Niacin mg.	Vitamin C mg.
A	Maize - ordinary	100	342	8.7	10.0	2.0	99.0	0.420	0.100	1.8	-
	Amaranthus	25	11	1.0	99.2	6.4	1389.0	0.008	0.075	0.3	24.8
	Jaggery (cane)	20	77	0.1	16.0	2.3	34.0	0.004	0.008	0.1	-
	Oil (Ground nut)	5	45	-	-	-	-	-	-	-	-
	Total		475	9.8	125.2	10.7	1504.0	0.432	0.183	2.2	24.8
B	Opaque 2 maize	92	315	8.8	9.2	1.8	82.8	0.386	0.092	1.65	-
	Corn starch	8	28	0.1	-	-	-	-	-	-	-
	Amaranthus	25	11	1.0	99.2	6.4	1380.0	0.008	0.075	0.3	24.8
	Jaggery (cane)	20	77	0.1	16.0	2.3	34.0	0.004	0.008	0.1	-
	Oil (Groundnut)	5	45	-	-	-	-	-	-	-	-
	Total		476	10.0	124.4	10.5	1496.8	0.398	0.175	2.05	24.8

Contd.......

Groups	Ingredients	Quantity g.	Calories	Protein g.	Calcium mg.	Iron mg.	B-carotene mg.	Thiamine mg.	Riboflavin mg.	Niacin mg.	Vitamin C mg.
C	Opaque 2 maize	92	315	8.8	9.2	1.8	82.8	0.386	0.092	1.65	-
	Corn starch	8	28	0.1	-	-	-	-	-	-	-
	Amaranthus	25	11	1.0	99.2	6.4	1380.0	0.008	0.075	0.3	24.8
	Jaggery (cane)	20	77	0.1	16.0	2.3	34.0	0.004	0.008	0.1	-
	Oil (Ground nut)	5	45	-	-	-	-	-	-	-	-
	Lime powder (mg)	0.519	-	-	332	-	-	-	-	-	-
	Iron tablet (mg)	60	-	-	-	60.0	-	-	-	-	-
	Total		476	10.0	456.4	70.5	1496.8	0.398	0.175	2.05	24.8
D	Skimmed milk	22	78	8.3	301.4	0.31	-	0.099	0.361	0.22	1.1
	Tapioca	80	126	0.6	40.0	0.72	-	0.040	0.080	0.24	20.0
	Amaranthus	25	11	1.0	99.2	6.4	1380.0	0.008	0.075	0.3	24.8
	Jaggery (cane)	20	77	0.1	16.0	2.3	34.0	0.004	0.008	0.1	-
	Oil (Groundnut)	15	135	-	-	-	-	-	-	-	-
	Total		427	10.0	456.6	9.73	1414.0	0.151	0.524	0.86	45.9
Recommended Allowance ICMR 1971			1200	19.0	400 to 500	15 to 20	1000	0.6	0.7	8.0	30 to 50

Distribution of the quantity of the test food:

It was observed that children could consume at one time 20 g. (raw weight basis) of cereals on an average since the 20 g. became more than 60 g. on cooking.

Calculating the supplementation to meet one third of the daily recommended allowances it was decided to distribute the proposed quantity of maize in to three servings as morning snack, 20 g. of maize flour were used to prepare 'laddu', for the lunch 52 g. of maize were used to prepare rice and 20 g. were used to prepare the evening snack 'laddu'. Table II presents the distribution of the quantity of the test foods per child per day.

TABLE II

DISTRIBUTION OF THE QUANTITY OF THE TEST FOODS PER CHILD PER DAY

	MAIZE DIETS										
	A Ordinary maize			B Opaque 2 maize			C Supplemented opaque 2 maize			D Skimmed milk	
	Mid-morning	Lunch	Mid-afternoon	Mid-morning	Lunch	Mid-afternoon	Mid-morning	Lunch	Mid-afternoon	Mid-morning	Lunch
Maize (g)	20	60	20	20	52	20	20	52	20	Nil	Nil
Cornstarch or Potato (g)	Nil	Nil	Nil	-	8	-	-	8	-	Nil	Nil
Skimmed milk (g)	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	11	11
Tapioca/potato (g)	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	-	80
Amaranthus (g)	-	25	-	-	25	-	-	25	-	-	25
Jaggery (g)	10	-	10	10	-	10	10	-	10	10	10
Oil (groundnut g)	1	3	1	1	3	1	1	3	1	-	15
Lime Powder (mg)	-	-	-	-	-	-	100	319	100	-	-
Iron (Mg)	-	-	-	-	-	-	60	-	-	-	-

Standardisation of the recipes based on maize:

The recipes for the maize variety were standardised and prepared with all the safety measures of hygiene in the following ways:

1. Laddu/payasam
2. Uppuma
3. Uppuma with greens
4. Tomato maize bath
5. Vegetable maize bath
6. Tamarind maize bath.

Acceptability tests were conducted on these recipes on 100 selected children who were not included in the study. There is considerable literature to support the conclusion that taste and flavour of a production contribute to its acceptability and unless a product is acceptable to the mother, it is not likely to be fed to the target child. It is also necessary to ensure that the ingredients used are not identified with negative characteristics, for example 'flatulent', causes indigestion or 'low social status'. All these factors were ensured in the selection of the ingredients for the supplement, and their taste and flavour were found accpptable and not associated with any negative characteristics, by giving different tastes in the preparations instead of using one monotonous preparation. Varieties were tried on children and the best accepted receipes were finally selected.

Feeding the children with the supplement:

The feeding trial was started on August 1, 1974. The duration of the feeding was six months. The food and supplements were served daily for six days per week.

Lime powder was incorporated in the preparation of maize laddu and uppuma which were served during the midmorning, midafternoon and lunch times. 510 mg. of lime powder which provides 332 mg. of calcium per day per child was thus supplemented to the opaque 2 maize diet. 60 mg. of iron in ferrous sulfate tablet form was given after the midmorning snack.

Care was taken to see that the children consumed the diets and supplements served. A record of daily attendance of children participating in the feeding trial was maintained along with the exact quantities of the diet consumed noting the plate wastage, if any.

Evaluation of the effects of the different diets and supplements:

a. **Nutritional assessment of children:**

For the nutritional evaluation of supplements, various parameters were used to assess the nutritional status of children at the beginning and at the final stage. They were:

A. Anthropometry

B. Skeletal status

C. Clinical assessment and D. Biochemical examination.

A. **Anthropometry:**

Growth of children was assessed interms of their body weights, heights, head circumference, arm and chest circumference. Body weights and heights were recorded once a month. All the other anthropometry measurements were recorded once at the beginning and again at the end of the study. The weights is being recorded up to 0.5 g. with a level balance, which was corrected for zero errors with standard weights after every six weighings. The same balance was used throughout the study.

Since recording body heights (standard height) of children 18-30 months old is impossible, crown to heel measurement was taken using a specially constructed

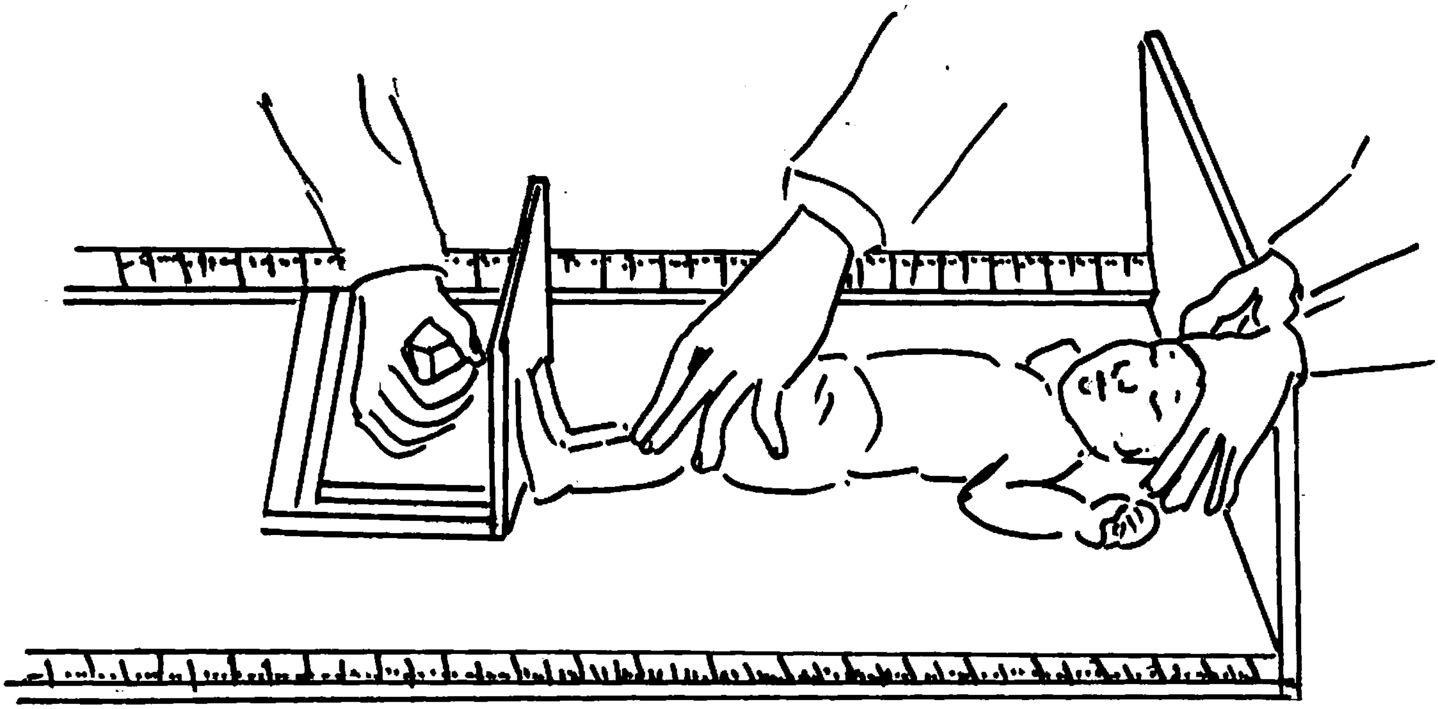


FIGURE I

INFANTO METER.

'Infante meter' shown in figure I. This is a wooden board about 5' long with a fixed head board and a sliding foot piece. The infant was laid on the board with his or her head positioned firmly against the fixed board. The sliding foot piece is then moved to obtain firm contact with the child's heel. Readings were being taken to the nearest 0.1 cm. (Jelliffe, 1966).

Head circumference was measured with a fibre glass tape measuring passing it over the supraorbital ridge and occipital prominense. Readings were taken to the nearest 0.1 cm.

Chest circumference of the child was measured by taking the average of the inspiredand expired chest measurements the nearest 0.1 cm with a fibre glass tape, with the child sitting on his mother's lap or in a stool.

B. <u>Skeletal status</u>:

Skeletal maturity is a measure of how far the bones of an area have progressed towards maturity, not in size, but in shape and their relative positions one to another, as seen in a radiograph. In theory, any or all parts of the skeleton can be used, but in practice, the hand and wrist are the most convenient areas, and the ones about which much is known. The bone maturity assessment was made by comparing the given radiograph with a set of standards and assigning the 'bone age' (Tanner *et al*. 1968).

Anterio-posterior radiographs of hand and pelvis were studied for two children from each experimental group at the beginning and at the end of the study with the help of a radiologist. The 'Bone age' was calculated by comparing with the standard. [FIGURE II]

C. Clinical assessment:

Clinical examinations were carried out with the help of a physician at the beginning and of the study for all the target children using the nutrition assessment schedule Annexure III.

D. Biochemical examination:

1. Haemoglobin:

Haemoglobin estimationsof the finger prick samples of the blood of the 12 target children were carried out using cyanmethohaemoglobin method at the beginning and at the end of the study (Cartwright, 1958) (Annexure IV).

2. Urinary calcium level:

The early morning urine samples of ten randomsly selected children each from the experimental and control

BONE AGE CALCULATION
[FIGURE II]

were analysed at the beginning and at the end of the study for calcium content using clark and collip method (Varley, 1969) (Annexure V).

3. <u>Faecal iron</u>:

The faecal iron of ten children from the experimental groups were estimated at the beginning and at the end of the study using the method given in NIN (1971) (Annexure VI).

<u>Analysis of food</u>:

One tenth of the day's food consumed by each of the five children from the five groups were collected and analysed for calcium and iron content to determine the consumption of calcium and iron from the home diets of the children. This was done at the beginning and at the end of the study.

Phytate and oxalate content of the foods were also calculated. This was done inorder to know the extent of interference of phytate and oxalate in the absorption of calcium.

IV. RESULTS AND DISCUSSION

A study was designed to evaluate the calcium and iron supplements to a selected group of preschool children of the age groups 18-30 months who were receiving maize based diets. The growth and nutritional status of the target children were evaluated in terms of their monthly body weight, crown to heel length, measurements of chest, arm and head circumference for a period of six months. The specific index for calcium supplement was 'Bone age' calculation through x ray photographs. An evaluation of urinary calcium, faecal calcium, phytate and oxalate content of the diets were further used to throw further light on the utilisation of dietary calcium. Faecal iron and dietary iron were used to evaluate the availability of iron supplements in the diet. As for iron supplementation, haemoglobin level was used as the index for evaluation.

An earlier study was carried out in this laboratory (Devadas, 1974) to evaluate the 'growth promoting ability of opaque 2 maize, ordinary maize and skimmed milk' with selected groups of pre school children. The supplementary diets used in this study were isocaloric (450) and isoprotein (10 g.). The results showed that the growth rate of target children fed with opaque 2 maize diet ranked next to skimmed milk diet. An examination of the nutrient content of the diets revealed that the skimmed milk diet provided an excess amount of calcium than the opaque 2 maize diet. It was felt that the growth promoting ability of skimmed milk diet may be due to its better quality protein or perhaps due to higher calcium content since all the other nutrients were equal and appeared to be adequate in the two experimental diets.

The present study was carried out to answer the above specific question whether the better growth promoting ability of skimmed milk diet is due to better quality protein or due to higher calcium content when compared with the opaque 2 maize diet. Thus the main objective was to find out the effect of calcium supplementation with opaque 2 maize diet in comparison with skimmed milk diet, when fed to pre school children for a period of six months.

Apart from growth retardation, iron deficiency anaemia was prevalent among target children. They consumed about 10 mg. of iron/day from their home diets and from the experimental diets. But inspite of this apparently satisfactory intake, iron deficiency anaemia was found among these children. To overcome this deficiency prophylactic iron was supplemented to the target children in this study.

125 children were selected from five different rural areas and all of them belonged to low socio economic group. They were divided into five groups of 25 in each group and were fed with calcium and iron supplemented opaque 2 maize

diets, ordinary maize diet, opaque 2 maize diet, skimmed milk diet and home diet.

The growth and nutritional status of children consuming calcium and iron along with opaque 2 maize diet was compared with the growth and nutritional status of children who were fed with ordinary maize diet, opaque 2 maize diet, skimmed milk diet and home diet.

General background of the target children:

The target beneficiaries belonged to low economic status. The average monthly income of the majority of the families of the target children was up to Rs.200. The (75%) majority of the families of the target children in the experimental villages were daily wage earners or industrial workers. Minority had primary school education. The children for the experiment were selected according to the age and they were free from known diseases. They were not participating in any other feeding programme.

Details of weaning and feeding practices of the target children:

From the data collected it was observed that at the age of twelve months liquid and solid foods were not usually introduced. At the age of twelve months, 50% of the infants were not given any solid foods and of these 25% were not introduced even to liquid supplements. At this age 20% have not received any other food than breastmilk.

At the age of 13 to 24 months, 50% of the children were given liquid and solid supploments along with breast milk. Only 7% received breast milk and 43% of the children received adultfood.

At the age of 2 to 3 years 80% of the children received adultfood. 18% received liquid supploments along with the adult food and only 2% were breast fed.

Table III shows the introduction of weaning foods at different ages.

TABLE III

INTRODUCTION OF LIQUID AND SOLID SUPPLEMENTS AT VARIOUS AGES (Percentage)

Age of wean-ing	Breast feed-ing alone	Breast feeding & liquid sup-plement	Breast feed-ing and li-quid and solid supp-lement	Adult food and liquid supple-ment	Adult food only
1 year	20	25	50	5	-
1 - 2	7	-	50	-	43
2 - 3	2	-	-	18	80
3 and above	-	-	2	-	98

The common weaning foods are rice, rice with rasam, cowsmilk, puffed rice, biscuits, murukku, fruits and coffee.

Table IV gives the dietary pattern and mean daily food consumption of target children.

TABLE IV

THE MEAL PATTERN AND MEAN DAILY FOOD CONSUMPTION OF TARGET CHILDREN

Name of the meal	Menu/Quantity consumed in g.
Breakfast	Cold rice (left over) 257/Iddli with chutney/kali
Lunch	Rice 260/kali/sambar 30/rasam 40.
Tea	Coffee
Dinner	Rice 240/kali/sambar/rasam 30

This table shows that the target children consumed adult food in very small quantities. There were absolutely no special weaning foods fed to these poor children.

The common breakfast items of the target children were rice/kali/iddli. Lunch consistedmainly of a rice/ragi/sorghum preparation with a watery sambar with little or no vegetable or with rasam.

Quantity of raw foods consumed by the target children are presented in Table V.

TABLE V

THE MEAN DAILY FOOD CONSUMPTION OF TARGET CHILDREN

Food stuff	Quantity (g)
Rice-Parboiled/sorghum/ wheat/ragi	170
Redgram dhal	10
Onion-small	5
Buttermilk	20
Ground nut oil	3

From the above table it is clear that the calorie and protein requirements are inadequate in the foods consumed by the target children. This was primarily due to their low economic condition and due to lack of knowledge among the mothers on low cost nutritious foods which may be fed to their young children.

Calorie and protein intake of target children:

Table VI gives the mean calorie and protein intake of target children.

TABLE VI

THE MEAN CALORIE AND PROTEIN INTAKE OF TARGET CHILDREN

Nutrients	Recommended Allowance ICMR 1971	Ordinary maize diet			Opaque-2 maize diet			Calcium and iron supplemented opaque-2 maize diet			Skimmed milk diet			Control
		Home diet	Supplement	Total	Home diet	Supplement	Total	Home diet	Supplement	Total	Home diet	Supplement	Total	
Calories/day	1200	773	475	1248	809	476	1283	796	476	1272	812	427	1239	805
Calories/kg of body weight/day	114			138			143			142			138	89
Protein (g)/day	18.3	16.1	9.8	25.9	18.8	10	28.8	20	10	30	20.3	10	30.3	21.2
Protein (g)/kg of body weight/day	1.25			2.8			3.2			3.3			3.3	2.3

The mean intake of calories in the home diets were found to be below the recommended allowances of ICMR (1971). Supplementation of the diets of the children with ordinary maize diet, opaque 2 maize diet and skimmed milk diet has helped in bridging the gap existing in the intake of calories.

Calcium and iron intake of target children:

Table VII shows the calcium and iron intake of target children.

TABLE VII

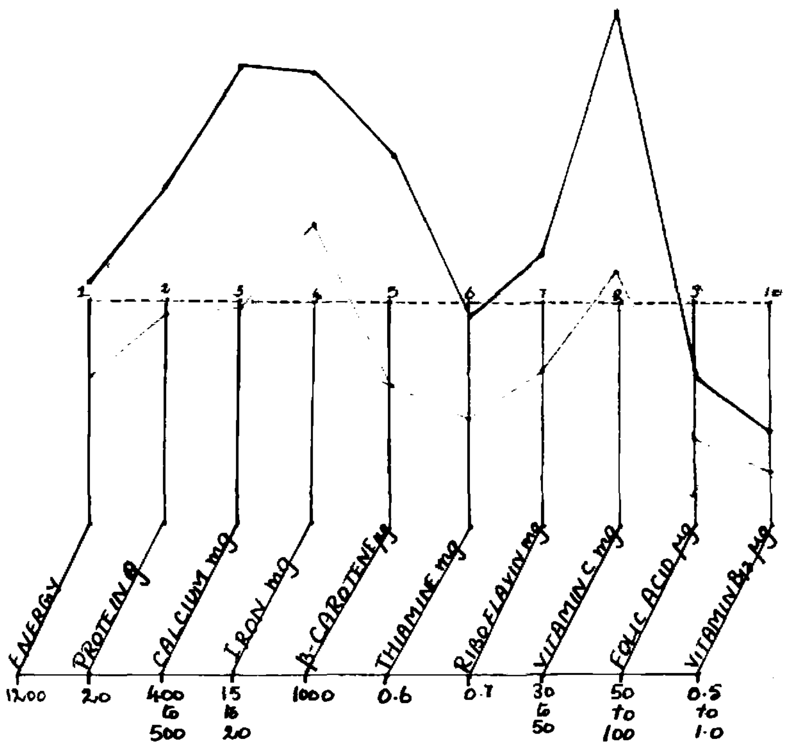
THE MEAN CALCIUM AND IRON INTAKE OF TARGET CHILDREN

Nutrients	Recommended Allowance ICMR 1971	Ordinary maize diet			Opaque-2 maize diet			Calcium and iron supplemented opaque 2 maize diet			Skimmed milk			Control
		Home diet	Supplement	Total	Home diet	Supplement	Total	Home diet	Supplement	Total	Home diet	Supplement	Total	
Calcium (mg)	400-500	130	125	255	163	125	288	63	457*	520	453	457	910	186
Iron (mg)	15-20	12	11	23	10	11	21	10	71**	81	20	10	30	7

* Calcium content of this diet is raised due to the supplementation of <u>519 mg. of lime powder</u> which provided 332 mg. of calcium per child per day.

** Iron content of this diet is raised due to the supplementation of 60 mg. of ferrous sulphate in the form of a tablet.

MEAN NUTRIENT INTAKE OF TARGET CHILDREN IN SKIMMED MILK GROUP



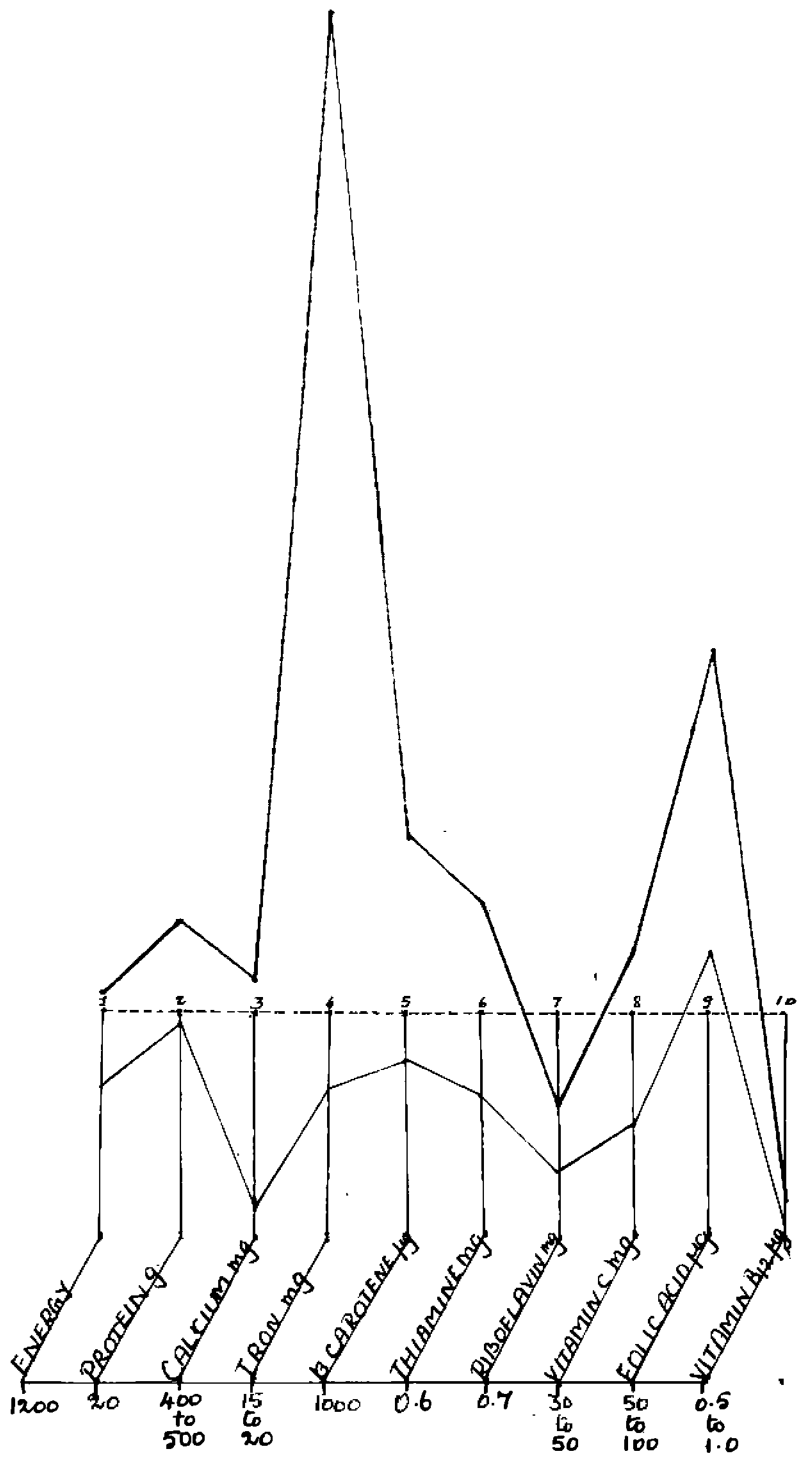
KEY:

HOME DIET + SCHOOL DIET

.... RECOMMENDED ALLOWANCE

—— HOME DIET

MEAN NUTRIENT INTAKE OF TARGET CHILDREN IN SUPPLEMENTED OPAQUE-2 MAIZE DIET.



KEY:

——— HOME DIET + SCHOOL DIET + SUPPLEMENT

- - - - - RECOMMENDED ALLOWANCE

——— HOME DIET

The mean intake of calcium and iron in the diets were found below recommended allowances of ICMR (1971). Supplementation of the diets of children with maize based diets and skimmed milk diet increased the intake of calcium and iron. Calcium and iron content of the calcium and iron supplemented opaque 2 maize diet was greater then compared to the other diets.

Vitamin intake of target children:

Table VIII gives the mean vitamin intakes of target children.

TABLE VIII

THE MEAN VITAMIN INTAKES OF TARGET CHILDREN

Nutrients	Recommended Allowance ICMR 1971	Ordinary maize	Opaque-2 maize	Calcium and iron supplemented opaque 2 maize	Skimmed milk	Control
B-Carotene (μg)	1000	1581	1750.8	1754.8	1616	127
Thiamine (Mg)	0.6	0.9	0.86	0.88	0.55	0.44
Riboflavin (Mg)	0.7	0.48	0.41	0.42	0.85	0.32
Vitamin C (Mg)	30-50	37.2	48.8	49.8	90.9	7.31
Folic acid (μg)	50-100	199	196	196	49	28
Vitamin B_{12} (μg)	0.5-1.0	0.12	0.13	0.11	0.32	0.11

This table shows that folic acid and vitamin B_{12} . necessary for the proper utilisation of iron in the body is not present in adequate amounts in all the five groups. This might be the reason for insignificant increase in Haemoglobin content of the target children even after prophylactic dosage of iron in their diets.

Effect of calcium supplementation:

Table IX shows the initial and final body weights and crown to heel lengths of the target children.

TABLE XI

MEAN INITIAL AND FINAL BODY WEIGHTS AND LENGTHS OF THE TARGET CHILDREN

Diet group	Age in months	Weight in kg.			Height in cm.		
		Initial	Final	Difference	Initial	Final	Difference
Ordinary maize	12-17	9.38	10.09	0.99±0.97	78.7	81.14	2.44±1228
Opaque-2 maize		9.74	10.74	0.99±0.47	82.1	84.6	2.5±1-0
Supplemented opaque-2 maize		9.30	11.04	1.74±0.33	79.2	81.9	2.7±1.15
Skimmed milk		9.88	11.20	1.32±0.36	79.8	83.4	3.6±0.82
Control		9.86	10.40	0.54±0.28	72.8	73.3	1.5±0.85
Ordinary maize	18-24	11.5	12.23	0.73±0.39	89.3	91.7	2.4±0.72
Opaque-2 maize		10.57	11.78	1.21±0.73	86.0	88.8	2.8±0.09
Supplemented opaque-2 maize		10.7	11.96	1.26±0.28	80.8	94.0	3.2±1.23
Skimmed milk		10.98	12.32	1.34±0.48	84.9	87.9	3.0±0.08
Control		10.20	10.58	0.38±0.77	80.7	82.1	1.4±0.41
Ordinary maize	25-30	12.99	13.94	0.95±0.507	87.9	90.3	2.4±0.43
Opaque-2 maize		12.59	13.58	0.99±0.61	88.0	90.8	2.8±0.78
Supplemented opaque-2 maize		12.53	13.92	1.39±0.56	88.7	91.9	3.2±1.24
Skimmed milk		12.74	14.21	1.47±0.53	85.0	88.0	3.0±0.78
Control		12.44	12.93	0.59±0.087	87.9	88.3	1.3±0.944

In each age group, the increments in weights and lengths of the maize based diets and skimmed milk were compared with the control. The increments were found to be significant in all the groups. ('t' values are given in Annexure VII). The increments in skimmed milk diet were found to be significant over opaque-2 maize diet but not significant over supplemented opaque-2 maize diet.

Bansal *et al* (1964) found that in Indian children subsisting on low levels of calcium (300 mg/day) supplementation of calcium to a level of 500-700 mg/day for a period of six months did not bring about any significant improvement in their growth performance. It was observed that there were no significant differences and radiosteometric measurements between the experimental and control groups.

Gopalan (1970) carried out experiments to decide whether the lack of response to calcium was due to other obstacles in growth caused by protein or calorie deficiency. Additional calories and protein were supplemented and even in such situations it was found that the increased calcium intake was not associated with any significant improvement in growth or physical development.

Table X gives the mean weight and length of the target children according to the experimental groups, regardless of age difference.

TABLE X

MEAN WEIGHT AND LENGTH OF TARGET CHILDREN ACCORDING TO EXPERIMENTAL GROUPS

Diet Group	Weight in kg.			Length in cm.		
	Initial	Final	Difference	Initial	Final	Difference
Ordinary maize	11.23	12.11	0.86±0.0105	86.2	88.7	2.42±0.0001
Opaque 2 maize	10.9	12.09	1.19±0.014	87.6	90.5	2.92±0.66
Supplemented opaque 2 maize	10.82	12.25	1.46±0.205	84.97	88.40	3.53±0.07
Skimmed milk	11.1	12.27	1.17±0.145	83.36	86.5	3.14±0.19
Control	10.83	11.59	0.76±0.01	80.2	82.3	2.1±0.08

This table reveals that calcium and iron supplemented opaque 2 maize diet group children were better than the other groups with regard to the increment in height. Skimmed milk diet ranked first with regard to increase in weight. But the difference between the calcium and iron supplemented diet and skimmed milk diet were not statistically significant.

Table XI indicates the growth index of the target children. $\left(\frac{\text{weight}}{\text{Height}} \times 100\right)$ (Swaminathan, 1971).

TABLE XI

GROWTH INDEX OF TARGET CHILDREN

Diet groups	Initial	Final
Ordinary maize	0.15	0.17
Opaque 2 maize	0.14	0.15
Calcium and iron supplemented opaque 2 maize	0.15	0.15
Skimmed milk	0.16	0.17
Control	0.16	0.16

Growth index of the children show that the target children were in normal growth at the beginning and at the end of the study.

Table XII shows the mean head, arm and chest circumference of the target children.

TABLE XII

MEAN HEAD, ARM AND CHEST CIRCUMFERENCE OF THE TARGET CHILDREN

Groups	Age in months	Arm (cm)			Head (cm)			Chest (cm)		
		Initial	Final	Difference	Initial	Final	Difference	Initial	Final	Difference
Ordinary maize	12-17	12.9	13.8	0.9	44.1	45.6	1.5	43.4	44.9	1.5
	18-24	13.0	14.0	1.0	45.6	47.2	1.6	43.9	45.5	1.6
	25-30	14.75	15.6	1.15	46.75	48.5	1.75	48.2	50.0	1.8
Opaque 2 maize	12-17	12.8	13.6	0.8	44.0	45.5	1.5	43.2	44.5	1.3
	18-24	14.9	15.3	0.4	46.4	47.4	1.0	43.4	44.8	1.4
	25-30	15.0	15.5	0.5	47.4	48.5	1.1	45.1	46.3	1.2
Calcium and iron supplemented opaque-2 maize	12-17	12.75	14.6	1.85	43.3	45.0	1.7	45.7	47.9	2.2
	18-24	13.6	15.64	2.04	44.5	46.1	1.6	46.9	49.2	2.3
	25-30	14.0	16.1	2.1	45.3	47.0	1.7	47.3	50.00	2.7
Skimmed milk	12-17	12.8	14.9	1.8	43.5	45.6	2.1	45.9	48.2	2.3
	18-24	13.8	15.84	2.04	44.7	46.9	2.2	46.9	49.5	2.6
	25-30	14.2	16.4	2.2	45.5	47.7	2.2	47.5	50.5	3.0
Control	12-17	12.5	13.0	0.5	43.8	44.9	1.1	42.8	43.9	1.1
	18-24	13.0	14.0	1.0	45.8	47.2	1.4	43.8	45.2	1.4
	25-30	14.7	15.8	1.1	46.5	47.9	1.3	46.4	48.0	1.6

There were significant increments in the measurements of the arm, head and chest circumferences of the children belonged to the calcium and iron supplemented opaque-2 maize diet and skimmed milk diet over ordinary maize diet, opaque-2 maize diet and home diet.

Skeletal status:

Anterio posterior radiographs of hand and pelvis were taken for two children in each group at the beginning and at the end of the study (Figure IX) calcium and iron supplemented group showed increments in bone age but were not significant. This might be due to the inhibitory action of phytate present in the cereal in the absorption of calcium.

This study suggests that the skeletal retardation found in young children may be due to energy protein deficiency. In addition vitamin A and calcium deficiency may be also crucial factors in promoting growth.

Table XXXI shows the bone age of the target children. Bone age was calculated by comparing the radiographs with the standard (Tanner et al. 1968).

X-RAY PHOTOGRAPH

FIGURE - IV

TABLE XIII

BONE AGE OF THE TARGET CHILDREN

Group	Bone age		Bone age x 100 / chronological age	
	Initial	Increment	Initial	Increment
Ordinary maize	1	0.4	67	3
Opaque-2 maize	0.6	0.5	40	15
Calcium andiron supplemented opaque-2 maize	1.5	0.9	75	21
Skimmed milk	0.6	0.8	30	16
Control	0.6	0.4	30	10
Mean		0.6		13.0
		± 0.21		± 6.3

The above table shows that the increment in bone age of the children who received calcium and iron supplemented opaque-2 maize diet ranked first. This increment might be due to the increased amount of calcium supplemented to the diet. But the increments were not significant.

Table XIV shows the skeletal status of target children. Cortical thickness of the femur was measured at the distance of half the maximum pelvic width from the lowest point of contact between the acetabulum and the head of the femur.

TABLE XIV

SKELETAL STATUS OF TARGET CHILDREN

Groups	Area of III metacarpal and its phalanges sq. mm.		Cortical thickness mm. 2nd metacarpal bone		Cortical thickness mm. Femur [illegible]	
	Initial	Increment	Initial	Increment	Initial	Increment
Ordinary maize	46.5	4.0	1.5	0.2	3.8	0.5
Opaque-2 maize	50.0	4.1	1.6	0.3	4.0	0.7
Calcium and iron supplemented opaque-2 maize	52.0	4.8	1.8	0.5	4.1	1.0
Skimmed milk	50.8	4.4	1.7	0.4	3.8	0.9
Control	46.2	2.3	1.5	0.1	3.9	0.3
Mean		3.92 + 0.37		0.3 + 0.15		0.68 + 0.5

Skeletal status of the children showed that the results were not statistically significant.

<u>Calcium intake versus urinary and faecal calcium:</u>

Lime powder 519 mg. was supplemented to opaque-2 maize diet to equalise the calcium content of this diet with that of skimmed milk diet. Lime powder was included in the midmorning, mid-afternoon and lunch preparations and children consumed it quite easily.

The total intake and excretion of calcium by the target children is presented in the table XV.

TABLE XV

TOTAL INTAKE AND EXCRETION OF CALCIUM BY THE TARGET CHILDREN

Groups	Calcium mg.			
	Intake	Excretion		Balance
		Urine	Feaces	
Ordinary maize	255	52	195	+ 88
Opaque-2 maize	288	71	107	+110
Calcium and iron supplemented opaque-2 maize	520	130	220	+170
Skimmed milk	1210	365	685	+160
Control	188	37	75	+ 76
Mean				100.8 + 37.9

The retention of calcium in the calcium and iron supplemented opaque-2 maize was greater than the other diets. Skimmed milk diet ranked next. The retention in the calcium supplemented diet was not significant and this might be due to the interference of oxalate and phytate in the absorption of calcium.

Table XVI shows the phytate and oxalate content of the different diets which might have interfered with the absorption of calcium.

TABLE XVI

PHYTATE AND OXALATE CONTENT OF VARIOUS DIETS

Group	Phytate mg.	Oxalic acid mg.
Ordinary maize	389	199
Opaque-2 maize	386	197
Calcium and iron supplemented opaque-2 maize	386	199
Skimmed milk	310	190
Control	370	185

Effect of iron supplementation:

Table XVII gives the mean haemoglobin content of the target children.

TABLE XVII

MEAN HAEMOGLOBIN CONTENT OF THE TARGET CHILDREN

Group	Mean value (grams per 100 ml)		
	Initial	Final	Increment
Ordinary maize	7.0	8.6	1.6
Opaque-2 maize	6.8	8.9	2.1
Calcium and iron supplemented opaque-2 maize	7.1	8.6	2.5
Skimmed milk	7.1	9.5	2.4
Control	6.6	7.7	1.1
Mean			1.94 ± 0.52

WHO (Average) 10.89/100 ml.

Judged by the WHO standards all the target children in the five groups were anaemic to start with. Greens was supplemented in all the diets except the control one which improved the haemoglobin content of the target children. Supplementation of iron in the form of ferrous sulphate to the opaque-2 maize diet improved the haemoglobin content and it was not significant.

Table XVIII gives the percentage classification of target children according to the severity of anaemia.

TABLE XVIII

PERCENTAGE CLASSIFICATION OF TARGET CHILDREN ACCORDING TO SEVERITY OF ANAEMIA

Groups	Normal		Mild*		Moderate*		Severe*	
	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Ordinary maize	10	16	50	54	20	15	20	15
Opaque-2 maize	15	20	52	56	23	18	10	6
Calcium and iron supplemented opaque-2 maize	15	35	55	60	25	5	5	Nil
Skimmed milk	12	17	54	53	22	20	12	10
Control	15	6	45	49	32	32	8	13

*A deficient of upto 2g/100 ml. of haemoglobin from the normal mean (WHO standard) is considered as mild, 2 to 4g/ml. as moderate, 4g. or more/100 ml. as severe anaemia.

Due to high prevalence of anaemia among the target children prophylactic dosage of iron was supplemented with opaque-2 maize diet. This increased the haemoglobin content of the target children and in turn decreased the severity of anaemia among the target children to a great extent. Gopalan *et al*, 1974, stated that the prevalence of anaemia decreased with increasing age. Moderate and severe degrees of anaemia were more frequently seen in younger children below 3 years of age.

Iron intake and excretion:

Table XIX gives the mean intake and excretion of iron by the target children.

TABLE XIX

MEAN INTAKE AND EXCRETION OF IRON BY THE TARGET CHILDREN

Groups	Intake	Iron mg. Excretion Faeces	Balance
Ordinary maize	23	22.9	+ 0.1
Opaque-2 maize	21	20.5	+ 0.5
Calcium and iron supplemented opaque-2 maize	80.5	79.0	+ 1.5
Skimmed milk	30	29.5	+ 0.5
Control	7.2	7.0	+ 0.2
Mean			0.56 + 0.496

The results of the iron supplementation to the opaque-2 maize diet showed thatthere was little increase in the absorption of iron when compared to all the other diets. Layrisse *et al* (1973) have shown that maize meal to which 60 mg of fortification iron in the form of ferric chloride has been added permits the mean absorption of only 0.3 mg. of iron in an iron depleted population. In the present study also the rate of absorption of iron in the iron supplemented opaque-2 maize diet was higher than the other diets but was not significant.

Apte and Venkatachalam (1964) showed that iron absorption from diets containing phytate can be increased by increasing dietary calcium, the maximal absorption being observed when the diet contained 1500 mg. calcium.

However, the diets of the poor segments of population in India are not only predominantly cereal based, containing high phytate, but they are also low in calcium and ascorbic acid. Iron absorption from such diets may, there, be expected to be poor. Chemical balance studies employing such diets gave values for iron absorption ranging from 7 to 28% with an average of 13% (Apte, 1968).

Gopalan (1970) has suggested that for all practical purposes the average net absorption may be considered not to exceed 10%.

Clinical examination:

The clinical picture of the target children at the beginning and at the end of the study is presented in Table XX.

TABLE XX

CLINICAL PICTURE OF TARGET CHILDREN

Symptoms	Ordinary maize		Opaque-2 maize		Calcium and iron supplemented opaque-2 maize		Skimmed milk		Control	
	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Protein calorie Malnutrition Mild	12	9	10	8	10	6	11	7	12	10
Moderate	3	1	3	-	2	-	2	-	3	1
Severe	-	-	-	-	-	-	-	-	-	-
Vitamin A deficiency	5	3	4	2	3	1	3	1	4	4
Vitamin B-complex deficiency	8	5	7	4	7	3	7	2	8	7
Anaemia	19	16	21	15	20	10	20	12	20	18

The most predominant clinical syndroms were mild forms of protein calorie malnutrition and anaemia. To considerable extent vitamin A deficiency and B complex deficiency symptoms were also present. The number of children manifesting the various symptoms of deficiency diseases had decreased to a great extent at the end of the study especially in the calcium and iron supplemented diet and skimmed milk diet in comparison with ordinary maize diet, un-supplemented opaque-2 maize diet and home diet.

In conclusion, this study demonstrates the feasibility of improving the growth of children by supplementing locally available limepowder which will increase the absorption of iron and also help in the growth of bones. This also suggests that calcium deficiency is a crucial factor in the aetiology of skeletal retardation in young children. Supplementation of iron in the from of ferrous sulphate tablet in excess amount hoops to increase the haemoglobin content and will provent anaemia to a considerable extent. This supplementation must be carried out for a longer time to get good results.

Results show that the growth promoting ability of skimmed milk diet in the provious study was due to its better quality protein and not due to its higher calcium content.

V. SUMMARY AND CONCLUSION

A study was designed to evaluate the calcium and iron supplements in addition to a maize based feeding to a selected groups of preschool children of the age 18-30 months. The growth and nutritional status of the target children were evaluated in terms of their monthly body weight, crown to heal length, measurements of chest, arm and head circumference for a period of six months. The specific index for calcium supplement was 'Boneage' calculation through xray photographs. An evaluation of food calcium, urinary calcium, faecal calcium, phytate and oxalate content of the diets were used to throw further light on the utilisation of dietary calcium. As for iron supplementation haemoglobin on finger prick blood samples was used as the index for evaluation. Faecal iron and dietary iron analysis were carried out to evaluate the availability of iron supplements in the diets.

An earlier study was carried out in this laboratory (Devadas, 1974) to evaluate the 'growth promoting ability of opaque-2 maize, ordinary maize and skimmed milk with selected groups of preschool children. The supplementary diets used in this were isocaloric (450) and isoprotein (10g.). The results showed that the growth rate of target children fed with opaque 2 maize diet ranked next to skimmed milk diet. An examination of the nutrient content of the diets revealed that the skimmed milk diet provided an excess amount of calcium than the opaque-2 maize diet. It was felt that the growth promoting ability of skimmed milk diet may be due to its better quality protein or perhaps due to higher calcium content since all the other nutrients were equal and appeared to be adequate in the two experimental diets.

The present study was carried out to answer the above specific question whether the better growth promoting ability of skimmed milk diet is due to better quality protein or due to higher calcium content when compared with the opaque-2 maize diet. Thus the main objective was to find out the effect of calcium supplementation with opaque-2 maize diet in comparison with skimmed milk diet, when fed to preschool children for a period of six months. Calcium lactate tablets were costly and could supply only less amount of calcium, therefore low cost indigenous source, lime powder was supplemented in the opaque-2 maize diet as a source of calcium. 319 mg. of lime powder which provided 332 mg. of calcium was supplemented per child per day.

Apart from growth retardation, iron deficiency anaemia was prevalent among target children. They consumed about 10 mg. iron/day from their home diets and from the experimental diets. But inspite of this apparently satisfactory intake, iron deficiency anaemia was found among those children. To overcome this deficiency prophylactic iron was supplemented 60 mg/day/child to the target children in this study.

The results of the study revealed that all the target children belonged to low socio economic status. They consumed adult food in very small quantities and there were absolutely no special weaning foods fed to them. The calorie requirement was found to be inadequate in the foods consumed by the target children. This was primarily due to their low economic condition and due to lack of knowledge among the mothers on low cost nutritious foods which may be fed to their young children.

The mean intake of calories in the home diets were found to be below the recommended allowances of ICMR (1971). Supplementation of the diets of the children with ordinary maize diet, opaque-2 maize diet and skimmed milk diet has helped in bridging the gap existing in the intake of calories. The mean intake of calcium and iron in the home diets were found below recommended allowances. Supplementation of home diets with maize based diets and skimmed milk diet improved the calcium and iron content of the diets of target children. Incorporation of lime powder and iron tablet in opaque-2 maize diet increased calcium and iron content.

The increments in body lengths and weights of supplemented groups of children were found to be higher than the control group of children. But the increments among the three maize based and skimmed milk diets were not significant. The increments in head, arm and chest circumference of the supplemented groups of children were not significant.

Anterio-posterior radiographs of hand and pelvis of the calcium and iron supplemented opaque-2 maize diet children showed increments in boneage but the increments were not significant. This might be due to the inhibitory action of phytate present in the cereal in the absorption of calcium or due to too short a study period or other constraints in the growth of the children. Increments in the area of III metacarpal bone and femur were there, but the increments were not significant among the target children.

The retention of calcium in the calcium and iron supplemented opaque-2 maize diet was greater than the other diets. Skimmed milk diet ranked next. The retention in the calcium supplemented diet was less significant and this might be due to the interference of oxalate and phytate in the absorption of calcium.

Judged by the WHO standards all the target children in the five groups were anaemia to start with. 25 g. of green leafy vegetables were supplemented in all the diets except the control one, which improved the haemoglobin content of the target children. Supplementation of iron in the form of ferrous sulphate to the opaque-2 maize diet improved the haemolglobin content upto 35%.

The results of the iron supplementation showed that there was only very little (2%) increment in the absorption of iron when compared to all the other diets.

The most predominant clinical syndromes were mild forms of protein calorie malnutrition and anaemia. 5 to 7 per cent vitamin A deficiency and B complex deficiency were also present. The target children who had the various symptoms of deficiency diseases had decreased to a great extent at the end of the study especially in the calcium and iron supplemented diet group and skimmed milk diet group in comparison with ordinary maize diet, unsupplemented opaque-2 maize diet and control diet.

In conclusion, this study demonstrates the feasibility of improving the growth of children by supplementing locally available lime powder for a longer period under controlled conditions. Supplementation of prophylactic iron for a longer period to the anaemic children might improve the haemoglobin content of the target children and might prevent anaemia to a considerable extent. Results show that the growth promoting ability of skimmed milk diet in the previous study was due to its better quality protein and not due to its higher calcium content.

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ANNEXURES

ANNEXURE I

Questionnaire to elicit information on Socio-economic background of the families.

I. 1. Serial number: Street:

2. Sample village: House Number:

II. Household particulars:

a. Name of the head of the family interviewed:

b. Person interviewed: (related to the head of the family)

c. Religion

1. Hindu ☐
2. Christian ☐
3. Muslim ☐
4. Others (specify) ☐

d. Caste:

e. Type of family

1. Nuclear ☐
2. Joint ☐

III. Please give the details of your household members and their demographic particulars.

S. No.	Name	Relation to the Head of the family	Age in completed years	Marital status	Age at the first child birth	Age at marriage	Education	Occupation	Income Rs.	No. of children alive	No. of children dead

ANNEXURE II

FAMILY AND INDIVIDUAL FOOD CONSUMPTION SURVEY-WEIGHMENT METHOD

Name of the investigator: Door No:

Name of the head of the family: Address:

Name of the subject: Date:

Age of the subject:

Name of the meal	Menu	Weight of total raw ingredients used by the family (g)	Weight of total cooked food consumed by the family (g)	Amount of cooked food consumed by the individual (g)	Raw equivalents used by the individual (g)
Break fast					
Lunch					
Tea					
Dinner					
Others					

ANNEXURE III

Nutrition Assessment schedule:

S.No:

Name of the village/Block: Date of Assessment

Name of the child: I Initial II Final

Name of head of the family:

Boys/Girls: Age:

- -

Clinical examination	I	II

- -

Anthropometry

1:1 Length (cm)
1:2 Weight (kg)

Clinical examination:

2:1 Loss of lustre
2:2 Thinning
2:3 Sparse
2:4 Discoloured
2:5 Easily plucked

Face:

3:1 Moon face (Puffy cheeks) (Boggy cheeks)

3:2 Naso labial dysebacea

- -

Contd.....

Clinical examination	I	II

Nail:

10:1 Koilonychia finger nails

Subcutaneous tissue:

11:1 oedema
11:12 marasmus

Musculo skeletal system:

12:1 craniotabes

12:2 Beading of ribs

12:3 Knock-knee, Bow legs

12:4 Epiphyseal enlargement

12:5 Frontal and parietal bossing

General remarks:

ANNEXURE IV

HAEMOGLOBIN ESTIMATION BY CYANMATHEMOGLOBIN METHOD

Reagents:

1. Drabkins Diluent solution

Sodium biocarbonate	-	1 g.
Potassium cyanide	-	0.05 g.
Potassium ferricyanide	-	0.20 g.
Distilled water to make	-	1 litre.

This solution should not be used after it forms a precipitate on the bottom of the storage bottle. The solution is preserved in a dark brown bottle and preferably under cold storage. Its preparation and handling should be done with great care.

Procedure:

1. Exactly 5 ml. of Drabkin's diluent solution is measured into a dry test tube from a burette or a pipette with suction bulb.
2. Exactly 0.02 ml. of blood is transferred fom a standardised Haemoglobin pipette into the diluent solution, usual care in filling and cleaning of loaded haemoglobin pipette must be observed.
3. The pipette is rinsed three times with the diluent solution, without allowing the formation of air bubbles in the solution.
4. The blood and the diluent are thoroughly mixed by the rotating the tube.
5. 10 minutes time is allowed for the formation of the cyanmethemoglobin.
6. 5 ml. of diluent solution is used as blank.
7. With green filter No.540 the readings are taken in a photoelectric colorimeter.

Calibration Procedure:

1. Total blood iron is determined by wong's method. This determination would give absolute amount of Haemoglobin.

2. Exactly 0.02 ml. of this known blood sample is measured as above into 5-0, 7.5, 10.0, 12.5 and 15.0 ml. respectively of diluent solution and mixed by rotating the tubes. These solutions are now equivalent to blood samples containing respectively 100, 67, 50, 40 and 30% that of the original solution.

3. The intensity of the colour is read using green filter 540 against diluent as blank set at zero O.D.

4. On a graph paper a standard graph is drawn using these haemoglobin concentration and corresponding density values.

ANNEXURE V

ESTIMATION OF CALCIUM
(Method of clark and collip)

Reagents:

1. Ammonium oxalate 4% solution
2. Ammonia 2% (V/V) solution
3. Potassium permanganate 0.01 N. Prepare freshly before use by diluting stock 0.1 N solution
4. 2 n sulphuric acid.

Procedure:

5 g. of the food sample and 5 g. of faecal matter were transferred into a crucible separately and they were kept over fire and charred. Then the charred samples were kept in a muffle furnace for 4 hours and ashed. The ash thus obtained was dissolved in a few drops of concentrated hydrochloric acid, transferred into a 100 ml. standard flask and made up to the mark with distilled water. This ash solution was used for the estimation of calcium and iron.

One ml. of urine was taken for the estimation.

1. One ml. of the test solution was pipetted out into a clean test tube.
2. Added 2 ml. of distilled water
3. Added a drop of methylorange indicator (The solution turned pink)
4. Then added a few drops of ammonia till the solution turned yellow.

5. Added a drop of concentrated hydrochloric acid. The solution turned pink.

6. The test tube was kept in a boiling water bath and the contents were allowed to boil.

7. After cooling, added 3 ml. of 4% ammonium oxalate and a drop of ammonia

8. Allowed it to stand overnight.

9. Centrifuged the contents.

10. Washed with ammonia, repeated the process till the supernatant was free from oxalate.

11. The oxalate free precipitate was dissolved in 2N sulphuric acid, heated to 70°C and titrated against 0.01 N potassium permanganate. The end point was the appearance of pale permanent pink colour.

ANNEXURE VI

ESTIMATION OF IRON

Reagents:

1. 30% sulphuric acid A. R. (30 ml. Concentrated sulphuric acid diluted to 100 ml.)

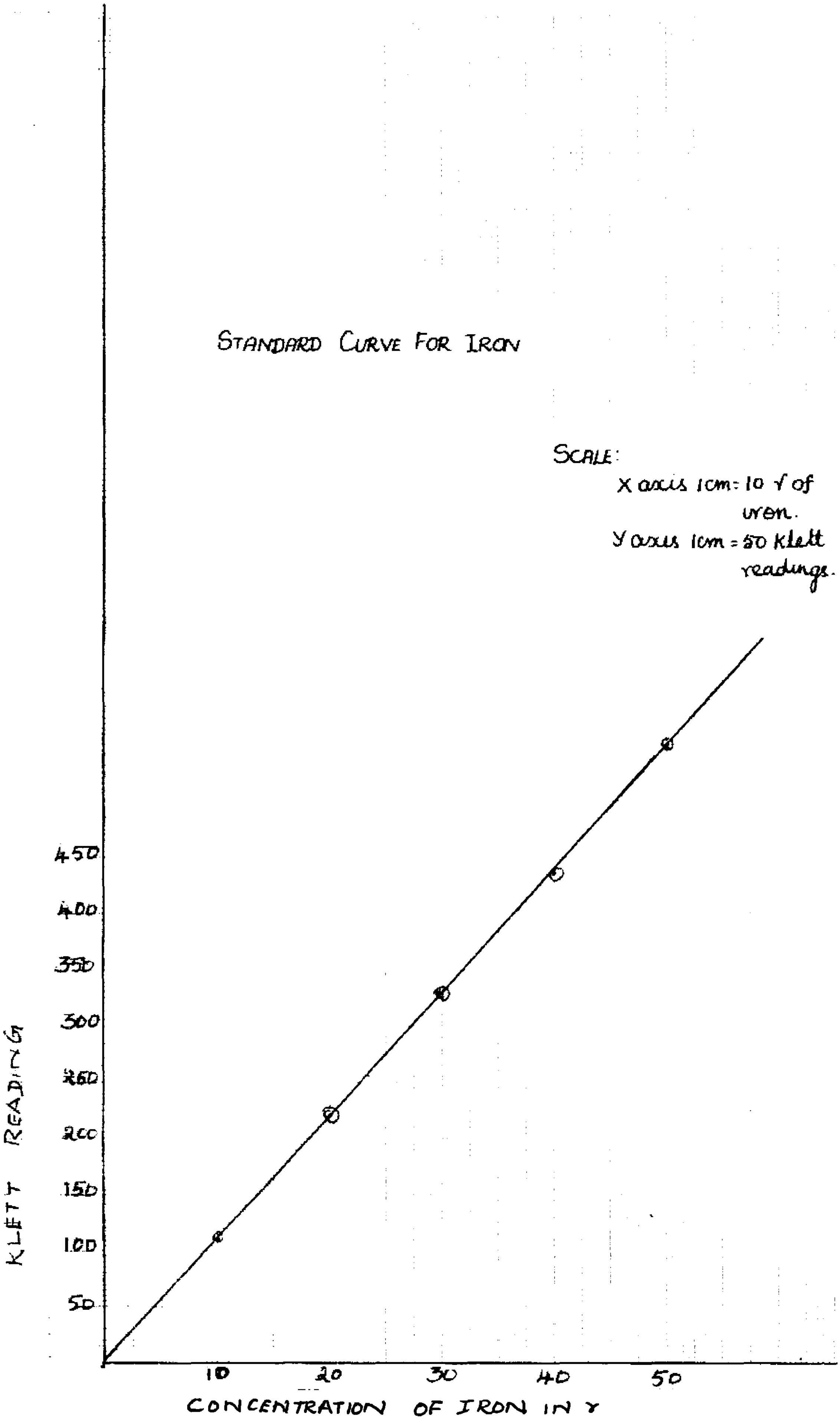
2. Saturated potassium per sulphate solution: 7 g. potassium per-sulphate are dissolved in glass distilled water and the solution made upto 100 ml.

3. Potassium thiocyanate 40% solution: 40 g potassium thiocyanate are dissolved in 90 ml. glass distilled water, 4 ml. acetone added and the volume made upto 100 ml.

4. Standard iron solution: 0.7022 g. ferrous ammonium sulphate is dissolved in 100 ml. glass distilled water, and after the addition of 5 ml. of 1:1 Hydrochloric acid, the solution is made upto 1 litre and mixed throughly (1 ml = 0.1 mg. Fe) The standard solution is prepared fresh once in six months.

   Working standard solution (0.01 mg. iron/ml) is prepared by diluting the above solution ten-fold.

(Note: For iron estimation all the reagents used should be free iron. Use of glass distilled water is preferred. If use of reagents containing traces of iron cannot be avoided, it should be seen that the final solution of standard and test contain identical quantities of those reagents containing iron as impurity).



STANDARD CURVE FOR IRON
SCALE:
X axis 1cm = 10 γ of iron.
Y axis 1cm = 50 Klett readings.
KLETT READING
450
400
350
300
250
200
150
100
50
10
20
30
40
50
CONCENTRATION OF IRON IN γ

Procedure:

To an aliquot (6.5 ml. or less) of the mineral solution enough water is added (if necessary) to make up to a volume of 6.5 ml. followed by 1.0 ml. of 30% sulphuric acid, 1 ml. of potassium per sulphate solution and 1.5 ml. of 40% Potassium thiocyanate solution. The red colour that developed is measured with in 20 minutes at 540 mμ.

ANNEXURE VII

STATISTICAL ANALYSIS (t VALUE)

Diet Groups	Age in months	't' values Weight	't' values Height	Age in months	't' values Weight	't' values Height	Age in months	't' values Weight	't' values Height
Ordinary maize Vs Control	12-17	0.96	1.53	18-24	1.24	4.87	25-30	0.054	2.6**
Opaque-2 maize Vs Control		0.21	1.8		2.49**	10.5*		0.156	3.0**
Supplemented opaque-2 maize Vs Control		6.8*	2.06**		3.4*	3.4*	4.4*	3.45**	3.0**
Skimmed milk Vs Control		4.8*	4.3*		3.3*	12.1*		4.0*	3.4*
Opaque-2 maize Vs Skimmed Milk		1.4	2.1		0.04	14.3*		1.5	0.45
Supplemented opaque-2 maize Vs Skimmed milk		2.1	1.6		0.045	0.5		0.254	0.34

* Significant at 1 per cent level

** Significant at 5 per cent level