AVAILABILITY OF β-CAROTENE FROM PAPAYA TO THE PRE SCHOOL CHILDREN OF LOW INCOME GROUP

By

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A C K N O W L E D G E M E N T

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I. INTRODUCTION

"Darkness is the time for sleep, not for seeing". These words depict the need for eyesight. The eye is a uniquely beautiful and complex organ. The readiness with which its intimate structure and functions can be observed, sets it aside from all other organs, even the skin. Momentary reflection on the structure of this organ leads one to suspect that it might be unusually sensitive to nutritional influences and one could suppose that malnutrition and vitamin A deficiency do not occur in the densely populated fertile parts of the country, as such green areas abound in vitamin A precursors (Emiru, 1971). In man, and particularly in children vitamin A deficiency most commonly and prominently affects the eyes, both externally by disrupting the epithelia of the cornea and conjunctiva and internally by lowering the senitivity of the retina to light. Unfortunately, Vitamin A deficiency is not a rare condition in the world even though scientific knowledge and technical skills could now markedly reduce its prevalence at a relatively modest cost(World Health Organisation (WHO) United States Agency for International Development, (USAID) 1976).

Vitamin A deficiency is a devasting public health problem in India, where malnutrition has taken its severe

form. Venkataswamy (1972) estimated one million cases of blindness in India due to malnutrition. Well nourished children hardly ever die from measles, whereas malnourished children show a mortality rate upto 40% (Gupta and Singh, 1975).

The incidence is particularly high among the preschool children of low income groups. It has been estimated that more than 20,000 children in such East Asia, the Middle East and tropical Africa are permanently blind (WHO, 1974). Very recently, Sauter (1976) has stated that 25% of population were admitted for various forms of malnutrition. Among them, 72% were between 1½ to 3 years old the fact which highlights the susceptibility of preschool children to various deficiency disorders.

Causes of a general nature for this widspread vitamin A deficiency are poverty, ignorance, poor living conditions and large family size (Kanawati 1973 and Morlay, 1973). The poor people in many parts of the country have to subsist on diets providing not much of animal foods and have to derive their vitamin A from carotene present in plant foods. Foods containing preformed vitamin A are expensive and beyond the reach of poor. Carotene derived from the plant foods therefore needs to be the chief source of vitamin A in the diet of many population groups in India (Rao et al, 1970).

Of the various carotenoids present in foods, β-Carotene is the most important both quantitatively and qualitatively due to its presence in common fruits and vegetables and its biological potency (Dam and Sondergaard, 1964). It is clear that the majority of the children in many developing countries mainly have to rely on yellow or orange coloured fruits and dark green leafy vegetables for an adequate intake of vitamin A precursors (WHO, 1974). Srikantia and Vinodini Reddy (1970) have reported that the most rational method for the control and prevention of vitamin A deficiency is the improvement of children's diets through inclusion of yellow coloured fruits and green leafy gvegetables.

A series of investigations have been undertaken on the availability of β-Carotene in different vegetables. But studies on comparative utilisation of β-Carotene by children and adults from fruits are few. Certain tropical fruits, notably mangos and papayas (paw-paws) provide useful amounts of Carotene whereas citrus fruits and non-tropical fruits in general contain very little (WHO/USAID, 1976). Among all fruits, only very few fruits like papaya constitute the cheapest and most easily available source. While 30 grams of leafy vegetable provided 400 µg of β-Carotene and

satisfactorily maintain the normal vitamin A status in young children (Chandrasekaran et al 1972), it is felt that yellow or orange coloured papaya can also supply large amounts of β-Carotene. Sauter (1976) has reported that the more coloured a fruit the richer its carotene content. Wenkam and Miller (1965) reported that this fruit can be an excellent source of β-carotene.

The Protein Advisory Group, PAG (1973) opines that "even among nutritionists, the importance of edible leaves and fruits in human diets seem inadequately stressed". This is a challenge we should take up. The present investigation is an attempt to gauge the availability of provitamin A or β-Carotene from papaya to preschool children, coming from low income group.

II. REVIEW OF LITERATURE

The review of literature of this present study has been given under the following headings.

A. Incidence of vitamin A deficiency.

B. Studies on serum vitamin A levels.

C. Protein deficiency and vitamin A.

D. Massive doses as a preventive measures.

E. Provitamin A.

F. Biological availability and absorption of β-carotene.

G. Various factors affecting the absorption of β-Carotene.

H. Faecal excretion of β-carotene.

A. Incidence of vitamin A deficiency:

Among the nutritional deficiency disorders affecting the children in India and other SoutheEast Asian countries, protein-calorie malnutrition and vitamin A deficiency disorders are the most widespread. Vitamin A deficiency accounts for nearly 25% of all cases of clinical malnutrition in pediatric age group. Maximum incidence is found between 3 and 5 years. (Venkatachalam et al, 1960).

Pathwardhan (1969) has reported that 8.1% of the children below 6 years of age in East Jeruselam and Amman

had clinical manifestations of vitamin A deficiency. In a study around the city of Hyderabad, the prevalence of vitamin A deficiency showed an increase with age. Diet surveys undertaken on a sub-sample showed that the intake of vitamin A was only about 60μg as against a requirement of 250-300μg /day and hence the deficiency (Swaminathan et al, 1970).

Reports of the Indian Ministry of Health based on recent surveys indicate that in the poor section of the population, the prevalence of vitamin A deficiency is about 3-8% in preschool children and 10-15% in school children and there are lower estimates of at least 15 million blind people in the world of to-day (WHO/USAID, 1976)

Night Blindness:

Difficulty in seeing in dim light is the first clinical symptom of vitamin A deficiency in man. (Dowling and Wald, 1958). Recent data from upper Volta show that there is considerable seasonal night blindness (Raoult, 1975)

Bitot Spots:

Bitot spots often have a more or less traingular shape, their base lying parallel to the limbus and after a long time, local metaplasia of epithelium may become irreversible

(Boyd, 1974 as cited by Sauter 1976). In 1974, examination of about 5000 preschool children in refuges camps in Gaza and Jordon revealed none with advanced eye lesions and five with bitot spots (Sauter, 1976) The prevalence Survey Covering 19,508 children in El Salvador did not reveal any cases of active corneal involvement, but five children with bitot spots, three with corneal lesions thought to have been due to vitamin A deficiency (Sommer et al, 1975). In 1974, the eyes of 90 children of 0-10 years old were examined in the Infectious Disease Hospital, Kenya. The result showed that majority of the children had pigmentation of conjunctiva (Sauter, 1976). He reported that in Central Province, among 580 preschool children, 2.6 percent were with bitot spots and among 2117 school age children, 0.9 percent were with bitot sports.

Xerophthalmia:

The amazing fact remains that it is in the green areas of the world, parts of India, Bangladesh, Indonesia and Phillipines that xerophthalmia appears as the real scourage (Pirie, 1970). Xerophthalmia means disease of the dry eye. All epithelial cells which line the surface and cavities of the body and most likely many of the other cells as well, are affected (Boyd, 1973). In the S.O.S. Children's village in Nairobi, fifty healthy children were examined. None

of them fifty showed any signs or symptons of Xerophthalmia. The Tear-film Breaking up Time (BUT) for the above mentioned children was measured and it was more than 10 seconds with ten consecutive measurings per child. (Sauter, 1976) BUT as cited by Sauter (1976) is defined by Lemp et al (1973) and Dohlman (1972) as the interval between a complete blink and the appearance of the first randomly distributed dry sport. It should be measured ten consecutive times and averages between 25 and 30 seconds in normal eyes with a wide fluctuation. The BUT should be longer than ten seconds with ten consecutive measurings (Lemp, 1973).
In 1974, in Kenyan National Hospital, 180 children of 0-10 year old had bitot spots and xerophthalmia. Available data suggest that about 25% of the survivers of severe xerophthalmia remain totally blind, about 50-60% are partially blind and only 15-25% escape with unimpaired sight (WHO/USAID, 1976). Xerophthalmia existing in Thailand is not a problem of major public health importance, whereas it has virtually disappeared from Hong Kong and Singapore. Xerophthalmia is not limited to poor rice consuming communities, but occurs also among people who have white maize for their staple food (Toureau, 1976).

The presumed Xerophthalmia cases in Maua Hospital from 1973 to 1974 given below depict the incidence of vitamin A deficiency.

Age distribution	Number of children	Percentage
6-12 months	17	32
1-3 years	21	40
4-7 years	11	21
8-13 years	4	7

Corneal Ulceration:

It is likely that rapid progression of the stromal ulceration is due to the release of protelytic enzymes from the atered epithelial cells (Dohlman, 1972). Typical of conjunctival xerosis owing to vitamin A deficiency are: The striking decrease of mucus producing conjunctival goblet cells and the keratinisation of the superficial epithelial layers (Dohlman, 1972).

Keratomalacia:

Blind school Report (1970) from Trichy shows that 47% of blindness is due to keratomalacia and the blind people become a liability to the nation. A report from the Government Erskin Hospital, (1971) in Madurai, indicates that 1014 cases of keratomalacia were treated in 1970. Individual reports from Bangladesh refer to a massive pro-

blem of Xerophthalmia and Keratomalacia in that country. Large numbers of cases of Keratomalacia are reported from Dacca and other Large urban centres (WHO/USAID, 1976)

Some available recent data from famous hospitals of South India given below depict the incidence of keratomalacia among the South Indian people.

Government Hospital	Year	Total number of out patients	Cases of keratomalacia
Trivandrum, Kerala	1971	39277	8
Minot opthalmic Hospital, Bangalore	1971	117838	1521
Government Erskin Hospital, Tamil Nadu	1972	47512	142
Sarojini Devi Hospital Hyderabad, Andra Pradesh	1971	75258	204

Sauter (1976) has reported that most of the children with marasmus and Kwshiorkor have keratomalacia in one or both eyes.

The biochemical findings in the yound children of American migrant families evidenced the prevalence of vitamin A deficiency (Larson et al, 1974). Lamb et al (1975) suggested that altered aminoacid metabolism, depressed appetite, in-

creased nitrogen excretion and cessation of growth are interrelated phenomena, closely associated with the onset of vitamin A deficiency.

B. Studies on serum vitamin A levels :

In Indonesia, preschool children whose habitual diets contained adequate amounts of B Carotene, but low levels of fat had low levels of vitamin A in serum (Roels et al, 1974). Pereira and Begum (1969) found that if childfen were given 30 grams of grean leafy vegetables daily for three months, adequate levels of serum retinol were maintained for further six months when the supplement of vegetables was withdrawn. The results of a supplementation study by Lala and Reddy (1974) showed a significant increase in serum vitamin A levels after feeding green leafy vegetables for two weeks. Amen and Lachance (1974) reported that vitamin A significantly depressed both serum and liver cholesterol and B-Carotene acted directly or indirectly to reduce the amount of total cholesterol in the serum.

The Mexican American in Texas had lower vitamin A levels than those in New York City, Children in the South and West of United States had consistently lower plasma retinol levels than those in the north central regions of the U?S. In Cebu, 17% of children had levels below 20 µg/ 100 ml of serum vitamin A and 57% had levels below 20 µg/ 100 ml. (Solon et al, 1975). A study by Dieleman (1975) shows that zinc deficiency lowers the serum vitamin A although liver

vitamin A was not dificient in rats. Pharaon et al (1969) found five per cent of the childrem of Jordan below 5 years of age with deficient levels of serum retinol. A recent study in Kenya, by Sauter (1976) reports that the serum retinol values in twelve healthy Dutch children between 2 and 5 years old are 27.2 to 57.6 µg %. and the serum β-carotene levels of fourteen healthy Dutch children range between 43.9 to 132 µg %.

C. Protein deficieny and Vitamin A:

Among nutritional disorders affecting intants and children, vitamin A deficiency comes second in importance to protein calorie malnutrition. Very often these two occur together with serious consequences. The question of relationship between protein-calorie malnutrition and vitamin A deficiency has attracted attention in recent years. It has been suggested by Arroyave et al (1961) that absorption of vitamin A from the gut is impaired in children suffering from kwashiorker. However Reddy and Srikantia (1966) found that there was a marked increase in serum vitamin A following oral administration of vitamin A even in severe cases of kwashiorkor, provided some fat was also given simultaneously. In a study conducted in children by

Vinodini Reddy (1976), the absorption of vitamin A has been found to be normal, whereas in protein-calorie malnutrition, the absorption was only 90%.

It has been found out that retinol building proteins are very low in kwashiorkor children, but normal in marasmic children (Smith et al, 1973). Feeding of 50,000I.U of vitamin A daily for four days significantly increased the liver microsomal protein in rats (Ram and Misra, 1974).

In Indonesia, 75 per cent of cases of protein calorie malnutrition were found to suffer also from xerophthalmia, whereas such association was rarely seen in Uganda (Food and Agricultural Organisation/World Health Organisation, FAO/WHO, 1967). In Lation America and Caribbean, 1-6 per-cent of malnourished infants and children have occular signs of vitamin A deficiency (Oomen, 1967).

In India, Venkatachalam and Gopalan (1960) reported that 36 percent of kwashiorkor cases in Coonoor and Hyderabad in the state of Andra Pradesh had associated xerophthalmia.

D. Massive does as a preventive measure:

Since vitamin A can be stored in the body for long periods, one possible approach is to administer a massive dose

of vitamin A so as to form a depot which can be used as and when needed for prolonged periods. The WHO Expert Committee on xerophthalmia in 1972, endorsed this method among pre-school children in the rice eating countries of Asia, where xerophthalmia is endemic.

In a study by Srikantia (1970), it was found out that giving oil soluble vitamin A preparation to preschool children showed no increased vitamin A levels in serum and water soluble vitamin A preparation produced higher levels of vitamin A by oral and parenteral route. Whereas, in case of severe gastro-enteritis and stomatitis of fourteen Kenyan children, an instramuscular injection of 1,00,000 I.U in vitamin A in oil given in the buttock proved a quick prompt response in them (Oomen, 1972). This preparation is to be prepared because of quicker and better release from the deport in the gluteal muscles resulting in a higher blood level (Mclaren et al, 1972). But Baurenfeind et al (1974) has reported that oil solutions of vitamin A should never be injected intramuscularly because the vitamin is liberated extremely slowly if al all, from the injection site. For practical consideration, Gopalan (1970) recommends two annual oral doses of 2,00,000 IU of vitamin A each at intervals of six months. One lakh I.U

under the age of one year, 2,00,000 I.U between one and six years and 3,00,000 I.U over six years protects the susceptible, malnourished vitamin A deficient children for about six months (Olson, 1972). The administration of a simple massive dose of 3,00,000 I.U to preschool children could maintain high levels of vitamin A for a period of 6 months and satisfactory levels beyond 6 months (Srikantia et al, 1974). The use of 3,00,000 I.U of and oil soluble preparation of vitamin A, administered once a year, has in fact been reported to bring down substantially the incidence of vitamin A deficiency in children between the ages of one and five years (Swaminathan et al 1970). In Bangladesh and Indonesia, in 1973, Capsules containing 2,00,000 I.U of vitamin A administered by mouth showed lowered prevalence rates.

As a preventive measure in Indonesia, ophthalmogists and pediatricians administer 1,00,000 I.U of vitamin A to all preschool children (Sauter, 1976). A single oral dose of 1,00,000 I.U of vitamin A was administered by Susheela (1969) to children between the ages of two and six years and elevated blood serum vitamin A values were observed at four months after dosing.

Perira (1971) found that a single dose of 50,000 I.U of vitamin A in oil solution given to pre-school children was effective for a lesser period of time.

E. Provitamin A:

Since many carotenoids have no vitamin A activity, the total amount of carotenoids present in food do not indicate the vitamin A value. Xanthophylls are the most commonly occurring carotenoid pigments which have no vitamin A activity; Although more than eighty naturally occurring carotenoids are known, only ten have vitamin A activity (Karrer, 1950). They are;

1. α-Carotene
2. β-Carotene
3. γ-Carotene
4. X-Catotene
5. Cryptoxanthin
6. Citroxanthin
7. Myoxanthin
8. Aphanin
9. Echinenone
10. Terulahodine

Of all these, β-carotene is the most commonly occurring caroteniod having highest biological value as provitamin A. The dietary carotene will not be utilised as such by the body, but converted to vitamin A and absorbed. So far as provitamin A β-carotene is concerned, FAO/WHO, Expert group (1967) and Davidson & Passmore (1973) have assumed that one µg β-carotene has biological potency of only 0.167 µg of retinol. This is based on the theroretical efficiency of conversion

to retinol which is 50 percent and the absorption of β-carotene from foods which is about 33 perwent.

F. Absorption of β- Carotene and biological availability:

The total vegetable and fruit consumption was found to be of the order of 44g among the rural poor in Madras, 10g. in Kerala, 60 g. in a tribal area in Gujarat and 100-150g. in urban Baroda. In all states of India, easpecially in the lower income groups, the highest vitamin A values were found in West Bengal (833.4 µg) and the lowest in Kerala (160 µg),(NIN, 1970).

Negeswara Rao and Narasinga Rao (1968) have found that the absorption of total carotenes from green leafy vegetables and papaya was found to be 58% and 46% respectively ahd β-cartene was found to be 76% and 90% respectively. Rajalakshmi and Ramakrishnan (1969) found that 30g. of green leafy vegetables a day cleared symptoms of vitamin A deficiency in two third of school children. A study by Narasinga Rao and Nagerwara Rao (1970) has proved that β-carotene from papaya is absorbed much better than that from wither carrots or amaranth. The less fibre content of papaya may be the basis ofor thes result. Protein Advisory Group (PAG) (1973) has reported that the use of fruits as a sourve of vitamins in young children feeding has been wisely endorsed.

Sauter (1976) has stated th t upto 1973, the children fed in the soup kitchen of Meriti which is run by National Christian Council of Kenya (NCCK) had night-blindness and bitot-spots. But after 1973, when they were fed thick soup of dried green vegetables every other day and yellow or orange beeries rich in β-carotene, nobody had signs or symptoms of xerophthalmia. Lala and Reddy (1970) found out that healthy but undernourished children fed 40g amaranath a day containing 1.2mg of carotene absorbed. 70 per cent of it and their serum retinol rose from 15 to 28 per 100 ml in two weeks.

Studies done by Rajalakshmi and Nageswara Rao (1970) on human subjects have proved a fairly efficient utlisation of carotene in leafy vegetables.

Vinodini Reddy (1977) proved that in the group supplied with amaranth, the serum vitamin A levels ranged from 21.2 to 31.5 µg/100ml. In a study by Baroda University (1976), the amounts of β-carotene consumed by the adults from green leafy vegetables was 3132 and the serum carotene level was 36 µg/100ml. and serum vitamin A was 15 µg/100ml. In a symposium report by Vinodini Reddy (1977) in NIN, it was quoted that utilisation of β-carotene is not so efficient as

vitamin A. The total carotene absorption from green leafy vegetables was 75% and the faecal excretion of carotene was higher.

Singh (1977) reported that lemon grass, the intermediate which is used for the manufacture of vitamin A is very rich in carotene.

Saroja (1968), Sangheetha (1970) and Jalaja (1971) proved better utilisation of β-carotene from radish tops, Chakravarthy Keerai and Kalavai Keerai . Patel (1972) in his study, found relatively higher vitamin A storage in groups of human beings fed fenugreak leaves, radish tops, corriander leaves, yellow pumpkin and kankoda and very less levels of liver vitamin A in the subject given vitamin A acetate. A similar study done by Mutalik (1972) suggests the greater availability of β-carotene in fenugreek leaves as compared to pure vitamin β-carotene in it.

Bhatt (1973) found 194, 227, 270, 145 and 121 µg of vitamin A in liver of the subjects fed fenugreek leaves, mustard greens, portulace, knol-khol and mint leaves respectively for the percentage of intakes 13,19,18,14 and 13 grams respectively.

A study conducted by Murthy and Geetha (1976) showed 76.7 percentage utilization of β-carotene from leaf protein and the absorption ranging widely between 58.3 percent and 90 percent. The same study reported that the mean utilization of standard β-carotene was 58.4 percent which was the highest utilization compared to leaf protein and amaranth.

In a study by Murthy et al (1976) ten students of Sri Avinashilingam Home Science College of the age group 20-24 years were fed 100 grams of cooked carrots which provide 1890 µg of carotene for 20 days. There was a greater utilisation and a significant increase of 2.8 µg/ml in serum vitamin A, whereas there was an increase of 8.1 µg/ml in serum vitamin A after 20 days of supplementation of 475 µg of retinol.

Storage of β-carotene in the body:

From accident data for young children, the lowest postmortem vitamin A value in the liver was found 15 µg/gram of fresh liver (Mclaren, 1966). In another study, hepatic stores of vitamin A in foetuses born to mothers of poor income groups are considerably lower than stores in foetuses born to wellnourished mothers (Iyengar and Apta, 1972). Liuer levels of vitamin A tend to be equally low in kwashiorkor,

but normal in maras, mug with (Smith et al, 1973). In a recent study by suthutvoravoot et al (1974), normal liver reserves of vitamin A were found in agroup of people with plasma levels of 10-20 µg/100ml.

C. Various factors affecting the absorption of β-carotene:

i. Dietary fat:

(Roels et al, 1963) reported that in Indonesia, pre-school children whose habitual diets contained adequate amounts of B-carotene, but low amounts of fat, had low levels of vitamin A in serum and this was raised by mere supplement of coconut oil.

Studies carried out in Kwashiorkor children showed that dietary fat facilitates the absorption of β-carotene, (Annual Report of Nutrition Research Laboratories, 1968). Further, emulsification increases the absorption of an oral dose of vitamin, A, but does not increase its biologigal utilisation (Ames et al 1969). Senger et al (1975) reported that there was no relation between essential fatty acid uptake and vitamin A levels.

In a study by Rajalakshmi et al (1975), the addition of upto 10 grams groundnut oil to a basal diet composed of

wheat and bengal gram was not associated with ah improved utilisation of β-carotene. The fact needed for efficient utilisation of carotene appears to be supplied by the intrinsic fat present in wheat (1-2%) and bengal gram (5.6%). It is supported by the report of Srikantia (1975) that in kwashiorkor, the vitamin A absorption was being poor in the absence of fat Vinodini Reddy (1976) has reported that dietary fat is essential for β-carotene absorption.

ii. Dietary protein

Esh et al (1962) reported that ingestion of different proteins did not affect vitamin A depletion from liver. But another study by Esh et al (1962) proved that liver vitamin A stores are reflected by the quality of protein.

Mortion (1970) suggested that a better quality of dietary protein increased liver and kidney storage of vitamin A. Serum vitamin A was directly proportional to the quality of dietary protein (Gamguly, 1970). In the fcase of carotene, protein deficiency markedly depresses the intestinal conversion of β-Carotene to retinol derivatives (Deshmukh and Ganduly, 1964). Roels et al (1972) reported that absorption of dietary vitamin is impaired in acute protein malnutrition.

Adikari et al (1970) have reported that the enzymic conversion of carotene to retinol and subsequent esterification, absorption and storage need adequate protein in the diet.

iii. Anti oxidants:

Antioxidents like vitamin E present in the diets also have an important effect on the absorption of β-carotene. Bennet et al (1965) reported that injection of vitamin E with vitamin A in fibrolytic children increased serum vitamin A value. The combination of vitamin A and E shortens the period of vitamin A deficiency treatment and can help in decreasing the use of over dose of vitamin A alone (Hanner, 1965). On the contancy, experiments with growing chicks by Sondergaard (1972) demonstrated with both fat free polyunsaturated fatty acid consuming diets, that added vitamin E shows the decline of hepatic depots of vitamin A. Kusin et al (1974) reports it is best to include 10-40 IU of vitamin E in capsules containing 2,00,000 IU of vitamin A for massive dosing, since it corrects any tocopherol deficiency.

iv. Antibiotics:

Bernard et al (1952) has shown that absorption of β-carotene was promoted on administration of streptomycin.

Seshan (1981) quoted that a few authors have reported that auromycin significantly increased the deposition of vitamin A derived from β-carotene, but it did not show any effect on preformed vitamin A.

v. Vitamin A:

Dua et al (1966) has shown that high dietary level of vitamin A interferes with the absorption of carotenoids from alimentary tract. Since vitamin A and carotenoids are structurally related, this would indicate a competition between the two for absorption from the alimentary tract.

vi. Riboflavin:

In the Africa Inland Mission Dispensary in Loglogo, twenty eye patients were examined in 1974. No one had signs of xerophthalmia, but had riboflavin-vitamin C depletion. But two school boys whose night blindness did not respond to adequate vitamin A therapy, recovered after riboflavin administration (Venkataswamy, 1976)

vii. Infection:

The interrelationship between vitamin A deficiency and infection has long been known (Scrimshaw, 1968).

Sivakumar and Reddy (1972) have proved that during infections, the intestial absorption of both β-carotene and vitamin A are seriously interferred with. Singh et al (1968) have found that mature round worms contained as much as 1.33 IU of vitamin A in contrast to earthworms which had no vitamin A in their bodies. Suringa et al (1970) have shown that the giant cells exereted from naso pharynx in measles patients who have conjuctival xerosis contain virus particles.

The role of infection is further proved in the statement that the giant cell formation in the corneal and conjunctival epithlium is in accordance with the giant cell production in the respiratory and digestive tract in measles patients (Scheifele & Forbes, 1973). Measles often plays 'catalysing effects' in the development of blindness caused by xerophthalmia (Bwiwo, 1970, Emiru, 1971, Jelliffe, 1973 and Gibbs, 1974).

Franken (1972) examined in Kenya and Tanzania many eye patients with corneal scars who blamed measles as the cause of their eye lesions. But Norn (1972) has reported that the extensive keratinisation and desquamation of the superfical layer of conjunctival epithelium which occur in conjunctival xerosis owing to vitamin A deficiency do not appear

in measles kerato-conjunctivities in well-nourished children. Shheifle et al (1972) reported that in protein and energy deficiency there is a close association of measles with xerophthalmia. Sauter (1976) in an intensive study, examined 45 well nourished and 234 malnourished children aged 0-6 years and found that malnourished children had measles and kerto conjunctivitis. Balance studies in using β-carotene and labelled vitamin have clearly indicated that even in injection not involving the gastro interestinal tract, the absorption of vitamin A and β-Carotene is impaired (Vinodini Reddy, 1973). It has been shown that leucocytes obtained from children suffering from kwashiorker, not only have decreased phagocytic activity, but also decreased bactericidal activity (Selvaraj, 1973)

Impaired absorption of labelled vitamin A has recently been reported in children with diarrhoea, repiratory tract infection and ascariasis (Sivakumar, 1975) Defective absorption of vitamin A in protein-calorie malnutrition is not due to severe malnutrition, but due to infections. On the contrary, carotene and vitamin A absorption was found to be normal in severe protein-calorie malnutrition. A marasmic woman of 29 with severe chicken-pox showdd conjunctival and corneal xerosis in the right eye and keratomalacia in the left eye (Sauter, 1976).

viii. Climatic and cultural factors:

Lysenko (1964) found that the β-carotene content of the plants were highest in spring and early summer, fell rapidly in not summer and rose again in autumn. A study by Deosthale (1969) showed that the β-carotene content of sorghum varieties varied considerably when grown at different locations due to the factors like social condition and fertilizer treatment.

India is at the bottom of table for fruit consumption. In south India, food equals rice. Fruits like papaya are considered to be harmful to children in the sense it is a heat-producing substance, causing diarrhoea and other ailments (Mely and Tan, 1970).

WHO/USAID, (1976) stated few taboos among the common ignorant people, some of them are; meat causes worms in young children; eggs g make young girls sterile, green leaves are for the goat; when pregnant again, a mother must immediately stop breast feeding.

ix. Cooking:

It has been found out by Nageswara Rao (1967) that as much as 40 percent of the carotene orginally present in the

in the food was lost because of cooking. Sud (1973) showed changes in ascorbic acid and carotene content of seome green leafy vegetables on cooking. Spencer (1973) reported that high temperatures of fat frying may result in considerable losses of β-carotene from plant foods. Gupta (1974) reported that conversion of butter to ghee resulted in an average loss of 26% in vitamin A and heating of hydrogenated fat to 200°C for five and ten mimutes resulted in 60-75% loss of vitamin A.

Another study by Gupta (1974) showed a loss of 12-30% of β-carotene during the preparation of dough for rotti from wheat, bajra, maize and Jower flours and a further loss of 22-31% occured during baking. Whole legumes and dals were found to loose 8-20% of carotene during pressure cooking and 15-38% during cooking by boiling. The corresponding figures for vegetables by the same author were 9-16% and 18-28%. The cooking of sprouted legumes resulted in 27-34% loss of β-carotene.

While cooking seems to destroy a neglible amount of β-carotene, it has been reported recently by Murthy et al (1973) that cooked carrots increase the β-carotene absorption and elevate the serum vitamin A level in children.

x. Processing:

Central Food Technological Research Institute CFTRI (1970)

has reported that fresh carrots have a high β-carotene content and that the loss of carotene during processing is gradual. The same institute in 1970 has found out the effect of canning on the β-carotene content of mango, papaya and Jack-fruit after storage periods of 3 weeks, 12 months and 6 months respectively at 25-30°c. The result showed no β-carotene loss in canned mangoes and papaya and 52% and 64% retention in Jack-fruit. This is of considerable importance in view of the fact that β-carotene is very easily destroyed in other foodstuffs, especially these containing fats. Sweeny and Marsh (1969) reported that as much 20 per cent of the provitamin A activity of green leafy vegetables and 35 per cent in yellow vegetables may be lost due to sterio isomerization during cooking.

H. Faecal excretion of β-carotene:

Almandinger and Hinds (1969) found that when haylage hay was fed to cattle, excretion of β-carotene and other carotenoids were much greater than the apparant amounts contained in the haylage and these apparent increase were much probably due to a releasing of residual carotenoids not extracted. Faecal excretions of carotene for rats receiving carrots were much higher than for those receiving carotene in cotton seed oil (Sweeny and Marsh, 1969) Reddy (1977) in a Symposium has reported that no vitamin A is excreted in faeces of the human beings.

III EXPERIMENTAL PROCEDURE

To estimate the availability of β -carotene from locally available papaya fruit, pre-school children hailing from low income group were chosen as subjects. The existing carotene intake was assessed and then the effect of addition of papaya as a supplement on absorption and availability was assessed. The experiment was conducted in two phases and the procedure for this study included the following steps.

PHASE I

A. Selection of the subjects.
B. Clinical assessment of the selected subjects.
C. Determination of food and nutrient intake.
D. Collection of a faeces.
E. Estimation of faecal β-carotene.
F. Estimation of haemoglobin levels.
G. Estimation of serum proteins
H. Estimation of serum vitamin A.

PHASE II

The β-carotene content of papaya was determined and 136 grams of papaya, yielding, 1,200 of β-carotene which is the requirement for the preschool children given by Indian Council of Medical Research ICMR (1968) cited by Gopalan et al (1976) was fed to the selected children. The effect of this supplement on absorption of β-carotene was studied.

Phase I:

A. Selection of the subjects:

Nutrition of the preschool child is of paramount importance, since the foundation for life time health, strength and intellectual vitality is laid during this period. In developing countries hunger and malnutrition are widespread among the preschool children. The incidence of vitamin A deficiency is particularly high among the preschool children of low income groups (PAG, 1975 and Brown 1975). Hence preschool children were chosen. Children attending a balwadi were chosen for easy access and for facility in incorporating the supplement later. Ten pre-school children-six girls and four boys of four years age, attending the balwadi at Naickenpalayam (situated in Perianaickenpalayam Block, 15km away from Coimbatore city) were chosen. A socio-economic survey was done so that, children selected came from the same strata of society. The other criteria for selection were, normal health, similarities in height and weight and co-operation extended by the parents. The heights of the ten subjects were taken using a fibre glass measuring tape nearest to 0.1 cm. Weights were taken using the human weighing balance nearest to 0.05kg. Heights and weights were taken at the beginning of the study.

B. Clinical assessment of the selected subjects:

Jelliffe (1968) opines that the cheapness and relatively easy organisation of nutritional assessment by means of clinical examination have led to the assumption that the method is simple, quickly mastered by the beginners and yields results that are easy to interpret. A schedule was formulated (given in Appendix I). Clinical assessment was done for the selected ten subjects using this schedule. In the balwadi, a dark adaptation test was conducted. The children were asked to count the fingers of the investigator in a semi-dark room at a distance of one meter(using the technique of Sauter 1976). All the selected children were able to count the fihgers and none of them possessed any inheranr disorders which might interfere with the study.

C. Determination of food and nutrient ihtake:

Food weighment survey is a positive step toward building better eating habits by finding out the needs. (Ramasastri and Murthy, 1973). Tasker et al (1967) opine that a three day weightment survey would be as efficient as that of the seven day weightment, if the dietary variation in the day to day life is not large as in the case of low socio economic groups in India.

Food consumed by the selected subjects at home and in the balwady was assessed by three day weighment method during the end of the Phase I when the children were not given any carotene rich supplement. The total raw food ingredients were weighed before cooking and the total cooked food was also weighed. The subjects were allowed to eat ad-libitum and their food intake was recorded. During this period, the parents of the subjects were asked not to give any vitamin or vitaminmineral supplement. The energy and nutrient intake were calculated for the raw equivalents, using the food composition tables of Gopalan et al (1976).

D. Collection of faeces:

Faeces was collected for three consecutive days on the days when weighment survey was done. Faeces was collected in weighed containers at the home of the selected children. Another set of weighed containers were kept in the balwadi to collect faeces from the children if there was any daefication. After 24 hours collection, total weight of the faeces was recorded correct to one gram. Aliquots of five grams were weighed and taken up immediately in an equal volume of ethanol and the sample of each child was refrigerated in an acid medium (9N $H_2 SO_4$) and later analysed. Nageswara Rao and Narasinga Rao (1970) have indicated that the values for

carotene absorption levels from different sources were similar whether daily stool samples were analysed for carotene content immediately or after storing in a refrigerator in acid medium for few days and analysed.

E. Estimation of faecal β-carotene:

The β-carotene excreted through faeces was estimated using the procedure of Jangannathan and Patwardhan (1960) The procedure is given in Appendix II.

F. Estimation of Haemoglobin levels:

Twenty cubic millilitres of blood was drawn through finger prick and samples were analysed for haemoglobin using Cyanmethaemoglobin method of NIN (1971). (Appendix III)

G. Estimation of serum proteins:

Because of relationship between proteins and vitamin A (Fry et al, 1975) it was important to obtain an estimate of protein nutrition of the target children. Accordingly, total proteins in serum were determined by the Biuret method (which is given in Appendix IV).

H. Estimation of serum vitamin A:

Five millilitres of blood was drawn from the enticubi-

tal vein of the children and the serum was separated and centrifuged in order to discard the sediments. Serum vitamin A was then estimated following the method of Neeld and Pearson (1963) and the solution was read at 348mμ in Beckman Du. 2. Spectrophotometer following the procedure of NIN (1971) (Procedure is given in Appendix V)

PHASE II

Yamamoto (1964) described the carotenoid composition of papaya as that showing significant amounts of carotenoids with vitamin A activity. Among a few carotene rich fruits like mangos, papaya and jack-fruit, papaya is the only cheapest source of the commonly available fruits in India and since this fruit is suitable for preservation in different forms and the retention of carotene is maximum, it was chosen as a supplement. Since various studies on papaya by Akamine and Goo (1971) and Peleg and Gomez (1974) have shown that fruits with initial colouration covering 61% of outer surface area develop full total solids content and the yellowish orange have intensity provides an indication about the percentage of ripending and carotene content of the fruit, a good yellowish-orange coloured Papaya (Col Carica papaya) was selected and obtained from Agricultural University, Coimbatore. Throughout the study, the papaya was obtained

from the Agricultural University to have uniformity of source. At the end of Phase I, the β-carotene content of the papaya was estimated using the procedure of Nageswara Rao (1967) (Appendix VI)

It was estimated that 136 grams of papaya gave 1,200 kg of β-carotene. From the beginning of the Phase II, 136 grams of ripe papaya were given to each of the selected subjects every morning for a period of one month. The consumption of papaya was supervised and no difficulties were encountered regarding consumption or its effect on digestion (Figure. 1).

At the end of the second phase of the study, the home diets of the selected were surveyed.

Faeces was collected for three days at the end of the phase using fersolate as faecal manker. For accurate work it is necessary to mark the faeces at the beginning and at the end of the collection period with a dye or other detectable marker and to analyse the faecal material collected between two marks (Davidson and Passmore, 1973). The total weight of the faeces collected each day was noted and stored for analysis in the same manner as described in Phase I. The faeces was analysed for β-carotene using the procedure followed in Phase I.

SUPPLEMENTATION OF PAPAYA TO THE SELECTED CHILDREN

FIGURE -1

The amount of β-carotene available to the body was obtained by calculating the β-carotene intake through foods and that exereted through faeces. The difference between these two values gave the β-carohene absorption by the children. From this, percentage of absorption was found out. This may represent only apparant absorption since the possibility of bacterial destruction of carotene in the intestional tract was not taken into consideration.

Using a clinical schedule which was used in Phase , I a clinical examination was conducted to observe superficial changes. Heights and weights of the selected subjects were also taken at the end of Phase II.

Haemoglobin levels, serum protein levels and serum vitamin A levels were determined using the procedures followed in Phase I.

IV. RESULTS AND DISCUSSION

The aim of this present investigation was to evaluate the availability of β-carotene from locally available papaya to preschool children of four years old, hailing from a low socioeconomic group. The children were participating in a balwadi feeding programme and their diets were further supplemented with ripe papaya for a period of one month. The intake of β-carotene and excretion of β-carotene in their faeces were analysed during each phase of the study period. The difference between both gave the biological availability of β-carotene. Haemoglobin levels, serum vitamin A levels and serum proteins were estimated and they served as useful indices of evaluation of this study.

PHASE I

A. General background of the selected children:

Through a socio-economic survey, the socio-economic status of the participating children and their families was elucidated. The families of the ten target beneficaries belonged to nuclear families of poor socio economic group with an income range of Rs. 100-200 per month. According to to Pereira (1971), a family is considered to be poor when

total annual income is Rs. 2,400 or less. The educational status of the family members was poor. The heads of the six families had studied only upto ninth standard and the others were illiterates. The occupation of the heads of five families was colli work. Among the other heads of the families, two were dhobies, one was blacksmith, one a pawn broker and one a lineman.

B. Body heights and weights of the selected children at the beginning of the study:

The selected ten children were all preschool children of age four years, attending a balwadi and free from any known diseases. Their heights and weights were measured. Measurement of weight is the best and easiest way to assess growth and development (WAG, 1971 and Siri, 1972) Table I shows the heights and weights of the selected children at the beginning of the study.

TABLE I

HEIGHTS AND WEIGHTS OF THE SELECTED CHILDREN AT THE BEGINNING OF THE STUDY

Subject	Sex	Height (cm)		Weight (kg)	
		Actual	*ICMR figure	Acutal	*ICMR figure
1	F	82.5	94.5	10.5	12.9
2	F	83.4	94.5	10.5	12.9
3	F	88.5	94.5	12.0	12.9
4	F	98.6	94.5	13.0	12.9
5	F	91.5	94.5	13.5	12.9
6	F	98.9	94.5	14.5	12.9
7	M	84.0	96.0	11.5	13.5
8	M	97.0	96.0	14.5	13.5
9	M	98.2	96.0	12.5	13.5
10	M	98.6	96.0	13.5	13.5
Mean		92.2		12.6	
Standard Deviation		± 5.007		± 1.393	

*ICMR (1971) All India level.

Among the ten selected children, six were girls and four boys. When height was considered on a total, five children (two girls and three boys) were above the ICMR levels. Only the weight of four children (three girls and one boy) was above the ICMR level. According to Gopalan (1972) a recent country wide

nutritional survey of thousands ofpreschool children showed that the height and weight of 90 per cent were below the 10th percentile value of the North American Children of corresponding ages.

C. Clinical assessment of the selected children:

The children were clinically examined and it was found out that they were free from any clinical symptoms.

D. Meal pattern of the selected children and their families :

The families of the ten selected children followed a three day meal pattern. There common breakfast itemswere left over rice, cholakali, ragi, rotti and previous day's sambar. The same items were used for the lunch also. For dinner, the very same items were prepared freshly. Thus the menus had not variety. At the balwadi, the children had only lunch for which they were given bulger wheat vegetable uppuma.

E. Food and nutrient intake of the selected children:

A three day food weighment Survey was carried out among the selected children's families. The intake of home and balwadi diets was noted. The mean food intake of children is presented in Table II (details in Appendix VII) and is illustrated in figure 2.

MEAN FOOD CONSUMPTION OF THE SELECTED CHILDREN

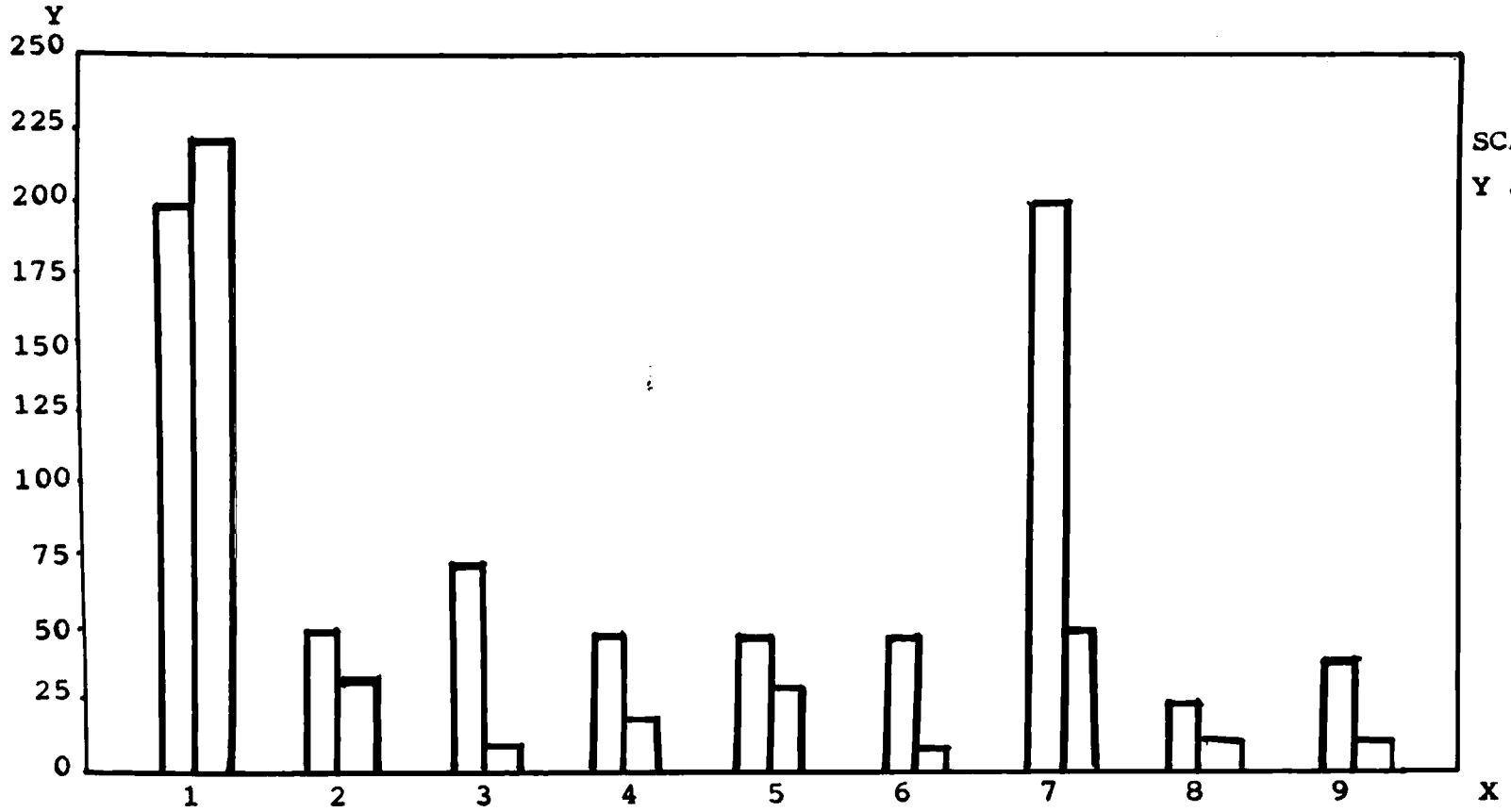


FIGURE - 1

1. Cereals
2. Pulses
3. Green leafy vegetables
4. Other vegetables
5. Roots and tubers
6. Fruits
7. Milk and its products
8. Fats and oils
9. Sugar and jaggery

Recommended allowance

Actual Intake

Apc. 3

TABLE II

MEAN DAILY FOOD CONSUMPTION OF THE SELECTED CHILDREN

No.	Foodstuffs	Acutal intake (g)	R.D.A ICMR (1968) * (g)
1	Cereals (Rice, Jawar, Ragi)	222	200
2	Pulses	29	50
3	Green leafy vegetables	9	75
4	Other vegetables	19	50
5	Roots and tubers	29	50
6	Fruits	6	50
7	Milk & its products	52	200
8	Fats and oils	15	25
9	Meat, fish	—	25
10	Egg	—	30
11	Sugar and jaggery	9	40
12	Nuts and spices	17	—

*Cited by Gopalan et al in Food Composition Tables (1976).

The table indicates that the allowances for cereals only was met by the children. The intake of all other foods especially protective foods was poor. The intake of green leafy vegetables and fruits was absolutely poor. The nutritious green leaves do not find their way into the mouth of the small child (Oomen, 1974). Though all of them were non-vegetarians, they did not consume fleshy foods during the survey. The above findingsare in accordance with Desai et al (1970) who found that on the basis of the trends on food consumption, the diets consumed by a large majority of people of India, based mainly on cereals and are lacking in protective and protein rich foods such as eggs, meat and fish. The reason behind this inadequate consumption of protective foods is that protective foods such as milk, meat, fish and egg are expensive and beyond the reach of the common man (Devadas and Usha Chandrasekar, 1973).

The mean nutrient intake of the selected children was calculated from the three day food weighment survey. The mean nutrient intake of these children is given in Table III. and is illustrated in figure 3. Individual mean values are given in Appendix VIII.

TABLE III

MEAN ENERGY, PROTEIN, IRON AND β-CAROTENE INTAKE OF THE SELECTED CHILDREN

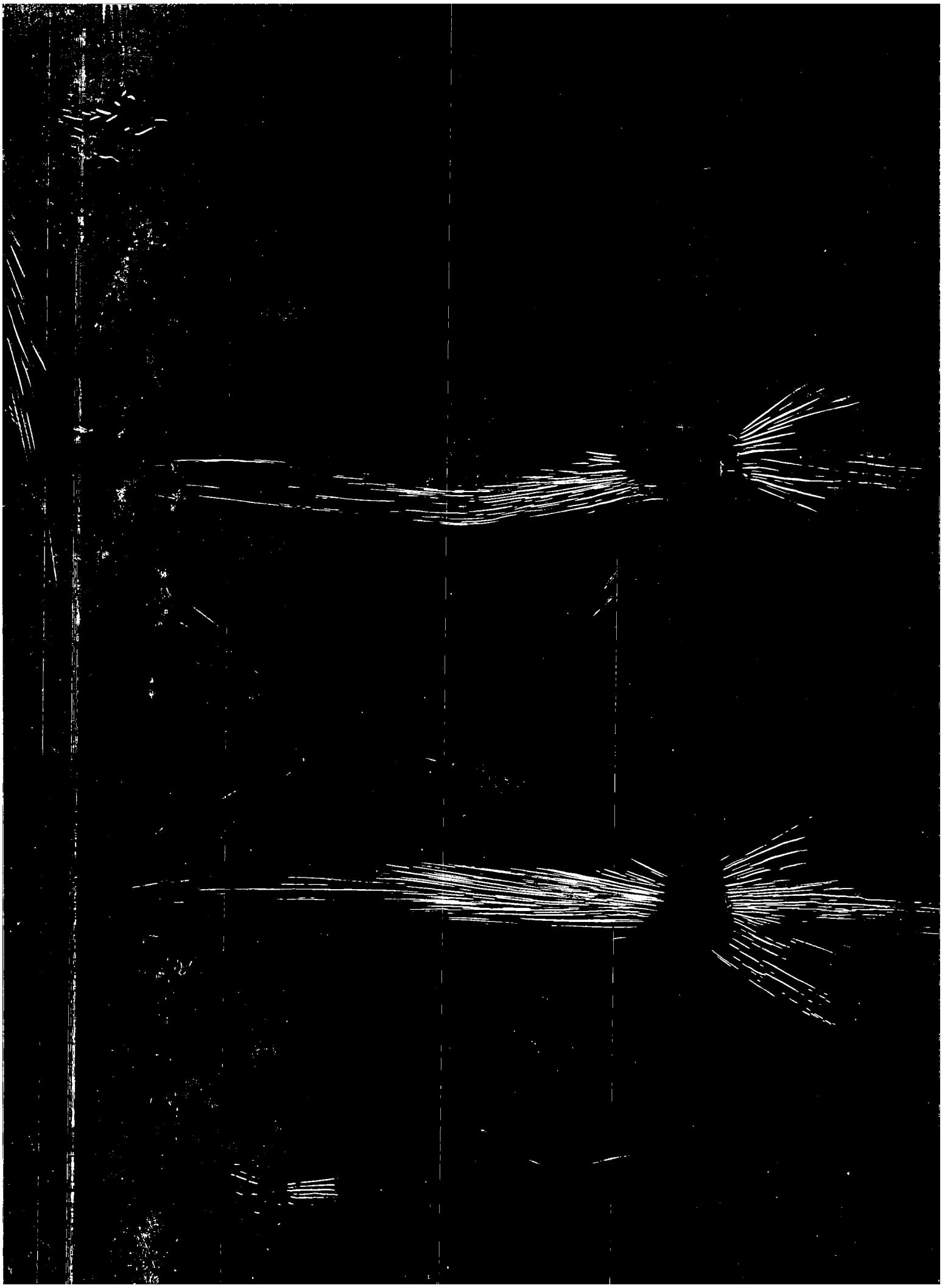
S.No.	Nutrients	Diet		Total	R.D.A ICMR(1968)*
		Home	Balwadi		
1	Energy(Kcal)	779	338	1117	1500
2	Protein (g)	23	6	29	22
3	Iron (mg)	11	5	16	17.5
4	β.Carotene (µg)	313	165	478	1200

*Cited by Gopalan et al in Food Composition Tables (1976)

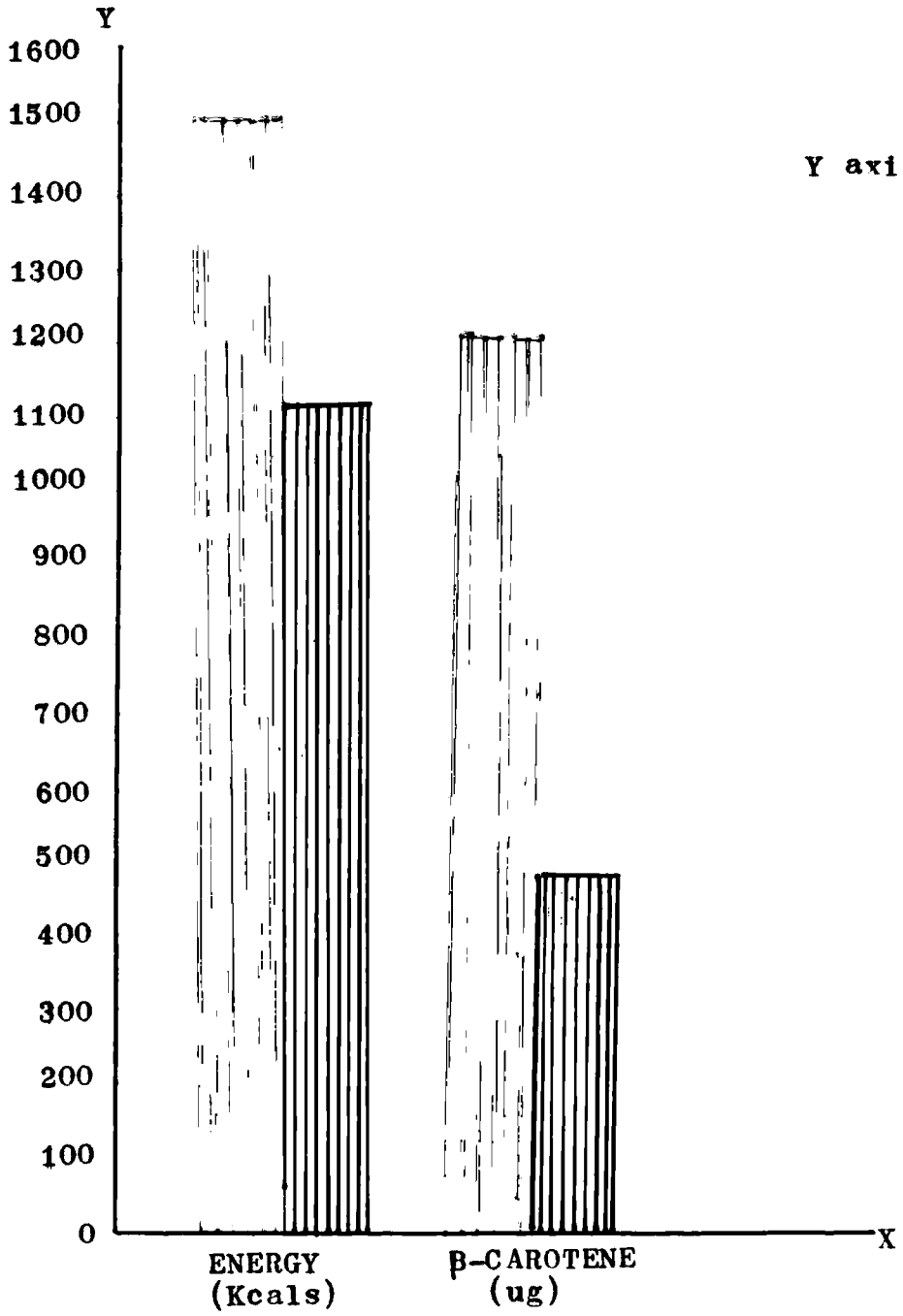
The above table indicates that there is a lacuna in the calorie intakes of the preschool children but there was no deficiency of protein. This finding is in tune with the

findings of Swaminathan (1968), Pereira (1971) and Gopalan (1971, 1973 and 1974). They reported that the primary inadequacy in the dietaries of preschool children is not protein but calories.

The mean intake of protein by the selected children was 29 grams which is comparable to the finding of Hegsted (1970) who reported that the mean intake of protein by preschool children ranged from 19-28 grams daily. The mean intake of iron by the group (selected children) was marginal. But the intake of β-carotene was poor. A study conducted by Venkatachalam and Reddy (1960) in Hyderabad showed that the carotene ihtake of the children were considerably lower than (250-300 IU) recommended allowance.



MEAN ENERGY AND β-CAROTENE INTAKE
OF THE SELECTED CHILDREN



SCALE:

Y axis 1cm = 100 units

FIGURE 3

Recommended Allowance

Actual intake

F. Excretion of β carotene through faeces:

Faeces was collected (during the three days of food weighment) from the selected children and the β-carotene excreted was estimated and the values are given in Table IV.

TABLE IV

PERCENTAGE ABSORPTION OF β-CAROTENE DURING BASAL PERIOD BY THE SELECTED CHILDREN

Subjects	β carotene intake (µg)	Weight of the faeces (g)	Faecal β-carotene (µg)	Percentage	β-carotene absorption (µg)	Percentage absorption of β-carotene ( )
1	361	150	199.0	53.19	169.0	46.8
2	1329	145	556.8	42.00	772.2	57.7
3	521	85	408.0	78.32	113.0	21.7
4	252	180	144.0	57.15	108.0	42.9
5	368	80	240.0	66.67	128.0	34.8
6	377	120	288.0	76.40	89.0	23.6
7	432	74	236.8	54.81	195.2	45.2
8	346	82	177.1	52.37	168.9	48.6
9	317	105	252.0	79.49	165.0	20.5
10	477	98	301.8	63.27	175.2	36.7
Mean	478	111.9	279.5	62.36	208.35	37.75
S.D.	±293	± 34.0	± 55.16	± 12.09	±190.5	± 12.28

The table shows that the faecal excretion of β-carotene by the selected children on basal diet ranged from 144 µg to 556.8 µg . The percentage excretion of β-carotene ranged from 42 to 79.4 per cent, the mean being 62.36 percent. It is in accordance with the report by Hume and Krehs (1949) who reported that 25 to 75 per cent of dietary carotene in man is not lost in the faeces depending on the dietary source. Another study by Nageswara Rao et al (1970) reported that total carotene content in the faeces collected during the first period on basal diet varied from 488-844 µg / day in healthy adults. On the other hand, the percentage absorption of β-carotene ranged from 20.5 to 57.7, the mean being 37.75±12.28. Maximum absorption was found in subject 2. This was later found to be due to the greater intake of green leafy vegetables by this subject than other children.

G. Haemoglobin levels of the selected children:

The haemoglobin contents of the finger prick samples of the blood drawn from the selected children at the beginning of the study are presented in Table V.

TABLE V

HAEMOGLOBIN LEVELS OF THE SELECTED CHILDREN

Subjects	Haemoglobin level	
	Grams/100 ml	Percentage
1	9.190	63.38
2	11.455	79.00
3	12.015	82.86
4	10.035	69.20
5	12.205	84.17
6	11.090	76.48
7	12.205	84.17
8	11.090	76.48
9	10.035	69.20
10	12.590	86.83
Mean	11.19	77.17
S.D	± 1.101	± 7.525

The haemoglobin levels for the selected ten children ranged from 9.190 tp 12.590 grams per 100 ml, the mean being 11.19±1.10. According to WHO (1968) the children upto six years are considered to be anaemic if their haemoglobin

levels are below 11 grams percentage. A study conducted by Baroda university (1976) on preschool children shows that many of the selected subjects had relatively low levels of haemoglobin. Gupta and Agarwal (1972) point out that there is always difficulty in selecting the normal Haemoglobin levels in developing countries.

H. Serum protein levels of the selected children:

Serum total proteins and albumin-globulin fractions were determined for the selected ten children and the values are given in Table VI.

TABLE VI

SERUM TOTAL PROTEINS, ALBUMINS AND GLOBUILINS OF THE SELECTED CHILDREN

Subjects	Total protein g/100 ml.	Albumins g/100ml.	Globulins g/100ml
1	5.8	3.2	2.6
2	7.2	4.6	2.6
3	6.7	3.4	3.3
4	6.2	4.2	2.0
5	6.0	3.0	3.0
6	5.4	2.9	2.5
7	6.4	3.7	2.7
8	5.7	4.1	1.6
9	5.6	3.1	2.5
10	6.5	3.6	2.9
Mean	6.15	3.58	2.55
S.D	±0.5291	± 0.5586	± 0.5383

The above table gives a picture of total proteins that are estimated in the serum of the selected children. The value ranges from 5.4 to 7.2 grams per 100 ml. the mean being 6.15 ± 0.5291. According to West et al (1967) the normal serum levels of total protein, albumins and globuilns are 6.0 to 6.9; 4.7 to 5.7 and 1.3 to 2.5 grams percent respectively. Since there is no protein gap found in the dietary habits of the selected children, majority of them had normal serum protein during the study period.

I. Serum vitamin A levels in the selected children:

The serum vitamin A levels of the selected children were estimated and are shown in Table VII.

TABLE VII

SERUM VITAMIN A LEVELS OF THE SELECTED CHILDREN

Subjects	β-carotene intake (µg)	Initial serum vitamin A levels (µg)
1	361	31
2	1329	33
3	521	23
4	252	24
5	368	25
6	77	28
7	432	30
8	346	32
9	317	21
10	477	26
Mean	478	27.3
S.D.	±293.0	± 2.282

The estimated serum vitamin A level ranged from 21 to 33 µg in 100 ml of the serum, the mean being 27.8 ±2.282. The committee on Nutrition of the American Academy of Pediatrics cited by Sauberlich (1976) considered a plasma vitamin A level of less than 30 µg/100ml as unacceptable. In the present study, subject 1,2,7 and 5 had serum vitamin A levels above 30 µg per 100 ml. Sauter (1976) has reported serum retinol levels ranging from 27.2 to 57.6 µg per cent in twelve healthy Dutch children of two to twelve years old.

J. Correlation coefficient between serum Vitamin A and protein :

Statistical analysis was done to find out the correction between serum vitamin A and protein levels of the selected children. The values are given in Table VIII.

TABLE VIII

CORRELATION COEFFICIENT BETWEEN VITAMIN A AND PROTEIN AT THE BEGINNING OF THE STUDY

No.	Blood picture	Correlation coefficient (r)
1	Vitamin A and total protein	0.1689
2	Vitamin A and albumin	0.4652
3	Vitamin A and globulin	0.4484

There was a positive correlation between vitamin A and total protein, Positive correlation was also noticed between vitamin A albumin and vitamin A and globulin.

K. Indentification of β-carotene from the source - papaya:

At the end of the first place, β-carotene content of papaya was estimated and the differentefractions obtained are depicted in Table IX

TABLE IX

ESTIMATION OF B-CAROTENE FROM PAPAYA

Column chromatography	Colour of bands	Identification	Reference
1st band	Orange yellow	β-carotene	
2nd band	Yellow pigment	Unidentified	(NIN, 1977)*
3rd band	Lemon yellow	∝Carotene	

*Unpublished

Three bands were separated on column chromatography. According to NIN (1967), the first orange yellow coloured band esd wondidered to be the β-carotene which was eluted with 2-3% of acetone in petroleum ether. The eluted β-carotene solution

was read at 540 450 mμ in Klett Summerson Colorimeter. Duplicates were done and the values ranged from 652 to 680 µg, the mean being 66 ± 14.01. This value was used in calculating the amount of supplement that is to be given to the children to contribute the allowances of 1200 µg set by ICMR (1968) cited by Gopalan et al (1976).

L. Comparision of β-carotene value of papaya with other studies:

The β-carotene content of papaya obtained in present study is compared with other studies.

TABLE X

β-CAROTENE CONTENT OF PAPAYA IN COMPARISION WITH OTHER STUDIES

Source	Studies	Amount of β-carotene (µg)	Descrption of the fruit
Papaya	Siddappa et al (1956)	773 ± 20	Large ripe yellow fresh fruit
Papaya	Siddappa et al (1956)	294	Large ripe red fresh fruit
Papaya	Present study (1977)	666 ± 14.01	Large ripe fruit with yellowish orange pulp

In the present study, the β-carotene content of the ripe papaya (col Carica Papaya) with yellowish orange pulp was been estimated to be 666 µg per 75 grams of the pulp. The value thus

obtained was found to coincide with the value given by ICMR (1968) cited by Gopalan et al (1976) in the Food Composition Tables. A study done by Siddappa et al (1956) on the same line reported that the ripe yellow fresh fruit has 773 µg of β-carotene per 100 grams of the pulp. This variation might be due to the variation in ripening process. Mcwilliams (1974) states that different amounts of pigments carten will be found depending on the light.

M. Feeding of the fruit, Papaye:

One hundred and thirty six grams of papaya which supplied the completed requirement of β-carotene was given the children for a period of one month in the second phase. No difficulties were encountered regarding consumption and digestion.

N. Changes in food intake:

At the end of the phase, a survey of food intake was carried out, No significant differences were observed in the ihtake at home or at the balwadi, besides the taking of the fruit.

O. Percentage absorption of β-carotene from papaya:

The faeces was collected for three days at the end of the second phase. Carotene content was analysed and percentage absorption of β-carotene was estimated. The mean values

of β-carotene excreted on the balwadi supplement and home diet was subtracted from the amount of β-carotene excreted in faeces after feeding the papaya along with home diet and balwadi supplement. The different was taken as excretion of β-carotene from papaya. This calculation was done, following the procedure of Reddy (1970). The percentage absorption of β-carotene is given in Table XI and illustrated in figure 4.

TABLE XI

PERCENTAGE ABSORPTION OF β - CAROTENE FROM PAPAYA

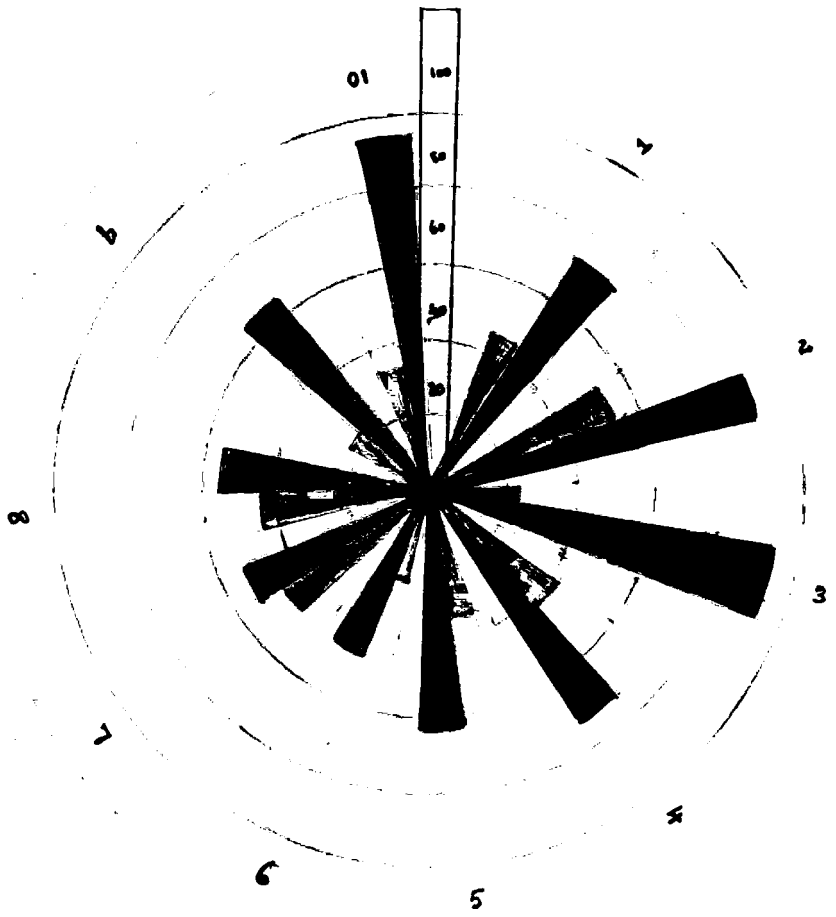
Subjects	β-Carotene intake from papaya (µg)	Weight of the faeces (g)	Faecal β-carotene excretion (µg)	β-carotene absorption (µg)	Percentage absorption β-carotene
1	1200	150	528.0	864.0	72.0
2	1200	175	420.0	1064.0	89.4
3	1200	80	480;0	1128.0	94.0
4	1200	190	496.0	857.6	71.5
5	1200	170	707.2	752.8	61.1
6	1200	200	960.0	528.0	44.0
7	1200	190	790.4	646.4	53.9
8	1200	235	733.2	644.0	53.7
9	1200	175	630.0	812.0	68.5
1010	1200	135	356.4	1145.4	95.5
Mean		170	609.16		70.36
S.D.		± 38.28	± 194.2		± 17.02

The percentage absorption of β-carotene from papaya by the children ranged from 44 to 95.5 per cent the mean being 70.36 ± 17.02. This is in accordance with Sauter's (1976) opinion that under normal conditions, 70 per cent of the ingested carotene is absorbed. A study by Nageswara Rao (1976) on four adult subjects who were fed papaya which contributed 3,139 µg of β-carotene reported that the percentage absorption of β-carotene ranged from 73.3 to 99.4 the mean being 90.3. In the present study, the beneficial effect was observed in a short a period as 30 days. This is in line with the balance study by Reddy (1970) who reported a maximum absorption of β-carotene from amaranath by five children within a short period of 15 days. They also pointed out the β-carotene from soft food like papaya is better absorbed than from fibrous green leafy vegetables and carrots.

In this study, a low absorption of β-carotene by subject -6 was observed. This child developed mumps at the end of the study for a short period. Perhaps the infection was a contributing factor for low levels. This is in accordance with the report of Reddy (1970) who noted that in two children who had acute respiatory infection, absorption of carotene from amaranath was quite low which again is in line with the fact that the infection decreases the absorption of β-carotene reported by Heymann (1941).

PERCENTAGE ABSORPTION OF β-CAROTENE BEFORE AND AFTER SUPPLEMENTATION WITH PAPAYA

SCALE:
1 cm = 20 ug of β-Carotene



——— Before supplementation

——— After supplementation

FIGURE - 4

P. Significance of β-carotene absorption from papaya:

The extent of absorption of β-carotene from papaya was statistically analysed and the values are given table XII

TABLE XII

SIGNIFICANCE OF β-CAROTENE ABSORPTION FROM PAPAYA

Source	Mean β-carotene intake (µg)	Mean percentage absorption	Siginificance (t)
Home diet + balwadi supplement	478	37.75± 12.28	
Home diet + balwadi supplement + papaya	1678 (1200+478)	70.36± 17.02	4.912*

* Significant at one percent level.

When the absorption of carotene from the diet which was consumed at home and balwadi (with papaya) was compared with that before addition of papaya, the difference was statistically significant at one per cent level.

Q. Heights and weights of the selected children after supplementation with papaya:

After the supplementation with papaya was over, the heights and weights were measured to note the changes. The data are given in Table XIII.

TABLE XIII

HEIGHTS AND WEIGHTS OF THE SELECTED CHILDREN BEFORE AND AFTER SUPPLEMENTATION WITH PAPAYA

Subjects	Height (cm)		Increase (cm)	Weight (kg)		Increase (kg)
	Initial	Final		Initial	Final	
1	82.5	83.0	0.5	10.5	11.0	0.5
2	83.4	83.9	0.5	10.5	10.8	0.3
3	88.5	88.9	0.4	12.0	12.5	0.5
4	98.6	99.0	0.4	13.0	13.5	0.5
5	91.5	92.1	0.6	13.5	13.5	..
6	98.9	99.4	0.5	14.5	13.5	-1.0
7	84.0	84.5	0.5	11.5	11.9	0.4
8	97.0	97.6	0.6	14.5	15.0	0.5
9	98.2	98.6	0.4	12.5	12.5	..
10	98.6	99.1	0.5	13.5	14.0	0.5
Mean	92.2	92.71	0.49	12.60	12.82	0.32
S.D.	± 5.007	± 5.889	± 2.0699	± 2.669	± 0.8386	± 0.0690

Comparison of heights before and after supplementation with papaya - t 0.2871

Comparison of weights before and after supplementation with papaya -t 0.2485

The above table indicates that all the children showed an increase in height. But the increase was not statistically

significant. Only one subject (6) showed decrease in weight after two monghts. This child as already mentioned had mumps at the end of the study and perhaps this may have interferred with food intake. In small children, unlike height, weight responds quickly to change in the body. It decreases for instance, when there is a restricted intake of calories and protein which causes a loss of fat and muscles (PAG, 1971). Sauter 1976 reported that the weight of the children of an orphanage in Kenya was 20% to 30%. This was too low for their age which was due to serious illness and malnutrition. The mean increase in weight of the ten selected children was not significant.

R. Changes in the clinical picture at the end of the study:

At the end of the second phase, the same children were clinically examined using the schedule that was used in phase I. Clinically all the children were normal, except the one who had mumps.

S. Changes in haemoglobin levels:

The haemoglobin levels at the end of the study were estimated and changes in haemoglobin picture is given in Table XIV

TABLE XIV

HAEMOGLOBIN LEVELS OF THE SELECTED CHILDREN BEFORE AND AFTER SUPPLEMENTATION WITH PAPAYA

Subjects	Haemoglobin-Initial		Haemoglobin Final		Increase g/100 ml.
	g/100 ml	Percentage	g/100 ml.	Percentage	
1	9.190	63.38	10.035	69.20	0.8
2	11.455	79.00	22.205	84.17	0.8
3	12.015	82.86	12.590	86.83	0.6
4	10.035	69.20	10.035	69.20	-
5	12.205	84.17	12.205	84.17	-
6	11.090	76.48	11.090	76.48	-
7	12.205	84.17	12.590	86.83	0.4
8	11.090	76.48	11.455	79.00	0.4
9	10.035	69.20	10.035	69.20	-
10	12.590	86.82	12.765	88.17	0.2
Mean	11.19	77.17	11.5025	79.325	0.32
S.D	± 1.101	± 7.525	± 2.108	± 8.694	± 0.3124

Comparison of initial and final haemoglobin levels- t= 0.4143.

Six among the selected ten children showed an increase in haemoglobin levels. But the increase was not statistically significant. It is perhaps due to lack of iron in the supplement.

T. **Serum protein levels in the selected children after supplementation with papaya:**

The total protein albumin and globulin levels of the selected subjects are presented in Table XV.

TABLE XV

SERUM PROTEIN LEVELS OF THE SELECTED CHILDREN BEFORE AND AFTER SUPPLEMENTATION WITH PAPAYA

Subject	Total protein g/100 ml.		Albumins g/100 ml		Globulins g/100 ml	
	Before	After	Before	After	Before	After
1	5.8	5.8	3.2	3.4	2.6	2.4
2	7.2	7.2	4.6	4.8	2.6	2.4
3	6.7	6.7	3.4	3.4	3.3	3.3
4	6.2	6.5	4.2	3.8	2.0	2.7
5	6.0	6.3	3.0	3.2	3.0	2.6
6	5.4	5.2	2.9	3.0	2.5	2.2
7	6.4	6.4	4.0	4.0	2.4	2.4
8	5.7	5.8	4.1	3.2	1.6	2.6
9	5.6	5.8	3.1	3.2	2.5	2.6
10	6.5	6.5	3.6	3.6	2.9	2.9
Mean	6.15	6.22	3.58	3.56	2.55	2.61
S.D.	± 0.529	±0.5442	±0.5040	±0.5383	±5383	± 0.3436

Comparison of total protein before and after supplementation with papaya t=0.2925.

The total proteins do not show any significant increase after feeding the papaya. This is due to the fact that the supplement is a fruit which has very little protein.

U. Serum vitamin A levels of the selected children after supplementation with papaya:

The serum vitamin A levels obtained from the selected children after the supplementation with papaya are compared with initial levels of the same children in Table XVI. The same is illustrated in figure 4.

TABLE XVI

SERUM VITAMIN A LEVELS BEFORE AND AFTER SUPPLEMENTATION WITH PAPAYA

Subjects	Initial (µg)	Final (µg)	Increase (µg)
1	31	35.5	4.5
2	33	43.5	10.5
3	23	33.5	10.5
4	24	30.0	6.0
5	25	29.5	4.5
6	28	28.5	0.5
7	30	32.0	2.0
8	32	38.5	6.5
9	21	29.0	8.0
10	26	32.0	6.0
Mean	27.3	33.2	5.9
S.D.	± 2.282	± 5.832	±3.089

Comparison of serum vitamin A levels
before and after supplementation with papaya - t 0.9426

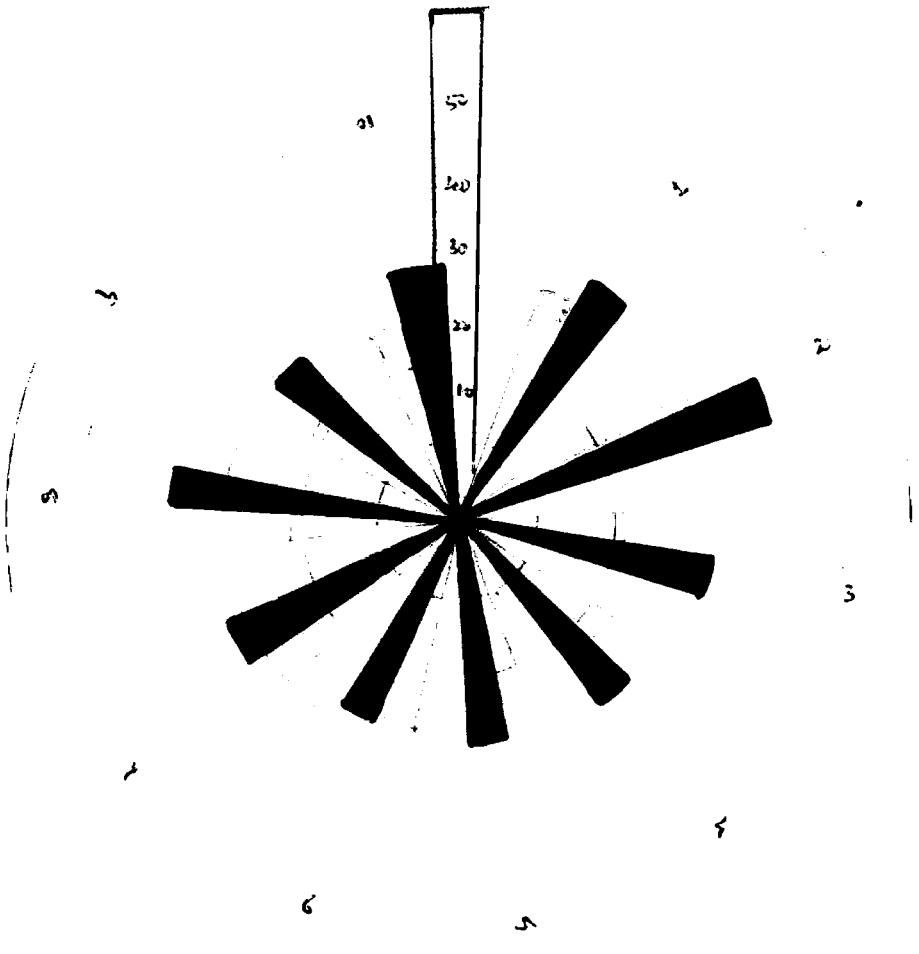
In all the selected children, the serum vitamin A was found to be increased. The mean serum vitamin A level was 33.2±5.832. The committee on Nutrition of the American Academy of Pediatrics, cited by Saubertifh (1976), considered plasma vitamin A levels of 30 per 100ml and above as indicators of vit min A intake to the increasing needs of the growing child and as well for liver storage. The mean increase in the serum vitamin A was 5.9±3.089,. This indicates that in these children, β-carotene was not only absorbed, but also converted to vitamin A and utilized well. The result of a supplementation study by L la and Reddy (1974) showed that there was a significant increase in serum vitamin A levels after feeding green leafy vegetables for the two weeks. A similar observation has been reported by Pereira and Begum (1968) when they fed green leafy vegetables to preschool children.

V. Correlation coefficient between vitamin A and protein :

The correlation coefficient between vitamin A and different protein fractions after supplementation is given in Table XVII.

INITIAL AND FINAL SERUM VITAMIN A LEVELS OF THE SELECTED CHILDREN

SCALE:
1 cm = 10 µg of Vitamin A



Initial level

Final level

FIGURE - 4

TABLE XVII

CORRELATION COEFFICIENT BETWEEN VITAMIN A AND PROTEIN AFTER SUPPLEMENTATION WITH PAPAYA

No.	Blood picture	Correlation coefficient
1	Vitamin A and total protein	0.3441
2	Vitamin A and albumin	0.5002
3	Vitamin A and globulin	0.0496

The table indicates a positive correlation between vitamin A levels and protein levels and protein fractions. A positive correlation between plasma albumin and vitamin A has already been reported in experimental animals by Friend et al (1961) and Ascarelli (1969) as well as in children by Kothari et al (1971). This correlation was done in order to get a complete picture of nutritional status of the subjects and also to investigate any association between protein and vitamin status in the subjects in view of the relations postulated between these two nutrients by Friend et al (1961).

W. Correlation between β-carotene intake and serum vitamin A levels before and after supplementation with papaya:

The correlation coefficient between mean β-carotene

intake and mean serum vitamin A levels was attempted and is given in Table XVIII.

TABLE XVIII

CORRELATION BETWEEN β-CAROTENE INTAKE AND SERUM VITAMIN A

Phase	Mean dietary β-carotene intake (µg)	Mean serum vitamin A levels (µg)	Correlation coefficient (r)
I	478±293	273±2.282	0.4309
II	1678)(1200+478)	33.2 ±5.832	0.6227

The table indicates that there was a positive correlation between the mean β-carotene intake and mean serum vitamin A levels in the selected preschool children in both the phases, the correlation being higher in Phase II. This may be due to the inclusion of carotene rich food-papaya.

V. SUMMARY AND CONCLUSION

A study was undertaken to determine the efficiency of absorption of β carotene and its availability from papaya to ten preschool children of four years old. The selected children attended a balwadi at Naikenpalayam, situated in Perianaikenpalayam Block, 15 km away from Coimbatore city. The children recieved the balwadi supplement of vegetable bulger wheat uppuma along with their home diet.

The study consisted of two phases. During the first phase the selected children were made to consume the balwadi supplement and home diet alone and the food intake was recorded for three days at the end of the phase I. During the following one month in the second phase, 136 grams of ripe papaya at a cost of six paise which contributed 1,200 µg of β-carotene were fed to the children. The faeces was collected for three consecutive days during the dtermination of each phase and was analysed for β carotene. The β-carotene intake and excretion were determined and this facilitated the estimation of absorption of β-carotene from the supplement-papaya by the selected preschoolers. The indices for evaluation indluded the estimation of haemoglobin, serum proteins and serum Vitamin A levels in both the phases.

The findings of the study are summarised as follows:

1. Percentage absorption of β-carotene improved after supplementing the home diet and balwadi diet with papaya. Initially, the mean percentage absorption of β-carotene was 37.75 ± 12.28 and at the end of the study it was 70.36 ± 17.02 The difference in absorption was highly significant at one percent level.

2. There was a mean increase in the serum vitamin A levels from 27.3 ± 2.28 to 33.2 ± 5.83 in the selected children, and the mean difference was 5.9±3.089. There was a positive correlation between initial and final vitamin A levels.

3. Since serum vitamin A levels are influenced by the B-carotene intake of the children correlation between these two factors were searched for. There was a positive correlation between mean β-carotene intake and mean serum vitamin A levels before supplementation (Phase I) and mean β-carotene intake and mean serum vitamin A levels of the children after supplementation with papaya in the second phase.

4. The mean total protein levels was 6.15±0.53 and 6.22±0.54 before and afterrthe study respectively. There

was a positive correlation between the mean initial and mean final serum total protein, but the difference was not significant.

5. There was a positive correlation between serum vitamin A levels and serum total protein levels in the second phase.

6. The mean haemoglobin levels of the children before and after supplementation were 11.19±1.10 and 11.50±2.10 respectively. The mean increase in haemoglobin levels was not significant.

From the study, it can be seen how the addition of such low-cost locally available food-papaya to the home diet and balwadi supplement improves the vitamin A status of the preschoolers. Such economic and low cost source of β-carotene namely papaya which can be given in amounts to provide the required amount of vitamin A can be recommended in the balwadi diets. Consumption of carotene rich foods like papaya by the preschool children along with cereal pulse mixture would certainly relieve them from the haphazards of vitamin A deficiency.

Recommendation:

The availability of carotene from different preparations of papaya and their effects on vitamin A serum levels need to be investigated.

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APPENDICES

APPENDIX - I

CLINICAL ASSESSMENT

Grade I : Healthy and free from any deficiency symptom

Grade II :

(a) Poor musculature

(b) Deficient sub cutaneous fat

(c) Mild anaemia

(d) Lack of interest in surroundings

(e) Mild signs of not more than one of the specific nutritional disorders or deficiences mentioned under group 3 of grade III.

Grade III: Group I

(a) Equal weightage - highly significant of malnutrition

(b) Nutritional oedema

(c) Gross muscular wasting

(d) Marked anaemia

(e) Xerosis of the cornea

Group 2 significant of malnutrition

(a) Xerosis or pi mentation of conjunctive

(b) Bitot spots

(c) Keratomalacia

(d) Caries

(e) Dry/or rough skin

(f) Crazy pavement of skin

(g) Hyperkeratosis

(h) Spoon shaped nails

(i) Dry lusturless hair

General Appearance:

(a) Good erect posture

(b) Neatness and cleanliness

Special remarks if any:

APPENDIX II

DETERMINATION OF FAECAL CAROTENE

The faecal carotene was estimated using the procedure of Jaganathan and Patwardhan (1960).

Reagents:

1. Alcohol - Distilled pure alcohol
2. Petroleum ether
3. Anhydrous sodium sulphate.

Procedure:

The faecal samples were soaked in alcohol over night and they were transferred to a mortar and ground with alcohol repeatedly until the extract had no more yellow colour. The alcohol extracts were then transferred to a separating funnel. An equal volume of petroleum ehter was added, swirled well and the lower layer was discarded. This procedure was repeated until the lower layer showed no trace of yellow. The petroleum ether extracts were then transferred to a separating funnel to which an equal volume of water was added, swirled and the lower layer discarded. The extract was freed from moisture by filtering through a bed of anhydrous sodium sulphate. This was washed with small quantity of petroleum ether until the filtrate was free from traces of carotene. The petroleum ether extract was made upto volume and the absorption of yellow clour was measured in klett Summerson Colorimeter at 540mμ.

APPENDIX III

ESTIMATION OF HAEMOGLOBIN BY CYANMETHAEMOGLOBIN METHOD

Reagents:

1. Drabkin's Diluent Solution:

   Sodium bicarbonate - 1 g

   Potassium cyanide - 0.05 g

   Potassium ferricyanide - 0.20 g

   Distilled water - 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 dark brown bottle and preferably under cold storage. Its preparation and handling should be done with great care since it is poisonous.

Procedure:

1. 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 from a standard haemoglobin pipette into a diluent solution. Usual care in filling and cleaning of loaded haemoglobin pipette must be observed.

3. The pipette is rinsed three times with diluent solution without allowing the formation of air bubbles in the solution.

4. The blood and the diluent are thoroughly mixed by rotating the tube.

5. 10 minutes time is allowed for the formation of the cyanmethaemoglobin.

6. 5 ml of the diluent solution is used as blank.

7. With green filter No. 540, the readings are taken in a photo electric 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 measured as above into 5.0, 7.5, 10.0, 12.5 and 15.0 ml respectively of diluted 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 mu against a blank set at zero O.D.

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

APPENDIX IV

ESTIMATION OF SERUM TOTAL PROTEINS AND ALBUMINS BY BIURET METHOD

Principle:

The (-CONH) groups in the protein molecule react with copper sulphate in alkaline medium to give a purplish violet or pinkish violet colour which is then read at 540 mu.

Reagents:

1. Biurent Reagent:

1.5 g of cupric sulphate and 6.0 g sodium potassium tartarate were dissolved in 500ml of distilled water. To this was added with swirling, 300 ml of 10% (W/V) sodium hydroxide (Co_2 free) and diluted to 1 litre in a plastic bottle. 0.1% potassium iodide was then added and stored at room temperature. This reagent was stable for upto six months.

2. 28% Sodium sulphite solution

3. Protein standard.

The standard protein solution might be either a pooled normal human serum, the protein con ent of which had been accurately determined by a kjeldahl method or solution of pure albumin. Use of albumin solutions permitted concentrations above the normal range. The serum or albumin solutions were diluted

with 0.9% saline to give exact standards of 1.0 to 8.0 per 100 ml.

Procedure:

0.1 ml serum was diluted with 1.9 ml of water. 8 ml of Biuret reagent was added and mixed well by shaking, let stand for 30 minutes and read in a spectrophotometer at 540 mu.

Albumin:

To 0.2 ml of serum in a test tube, added 4.8 ml of 28% sodium sulphite from a burette, (pipette should not be used as the salt crystallizes out easily) mixed and let stand for 10 minutes, filtered with Whatman No. 40 or 44 filter paper. Added 8 ml of Biuret reagent to 2.0 ml of the filtrate and proceeded as for total proteins. Ordinary centrifugation would not yield a clear globulin free supernatent and use of ether reagents at room temperature was not suitable.

Reagent blanks were prepared with 2.0 ml of distilled water and 8.0 ml Biuret reagent for total proteins and with 2.0 ml 28% sodium sulphite and 8.0 ml Biurent reagent for albumin.

APPENDIX V

ESTIMATION OF SERUM VITAMIN A (RETINOL)

The extraction procedure was done, following the method of Neeld and Pearson (1963) and the solution was read at 348 mu (in ultra violet region), following the procedure given by NIN (1971).

Procedure:

Reagents:

1. Alcoholic pottassium hydroxide 0.5%:-

   0.5 gram of potassium hydroxide was dissolved in 100 ml of purified alcohol.

2. N-hexane (HPC) B.P. 65° - 75°C.
3. Chloroform
4. Stock vitamin A solution: 344 mg of vitamin A acetate (300 mg of vitamin A) was dissolved in chloroform and made upto 100 ml. 1 ml of stock contains 3000 mg of retinol.

Intermediate standard:

1. 0.25 ml of stock diluted to 25 ml = 30 mg/ml
2. 0.50 ml of stock diluted to 25 ml = 60mg/ml
3. 0.75 ml of stock diluted to 25 ml = 90 mg/ml
4. 1.00 ml of stock diluted to 25 ml = 120 mg/ml

Working standard:

Each intermediate was again diluted in the ratio 1:9 and from each standard finally 1 ml was taken and 2.0 ml chloroform was added and read at 348 mμ in Beckman Du. Spectrophotometer.

Method:

1.0 ml of serum was placed in a test tube. An equal volume of alcoholic potassium hydroxide was added. 1.5 ml of N-hexane was added to the serum alcoholic potassium hydroxide in the test tube. The test tube was stoppered and the contents were shaken well for 45 seconds. It was centrifuged for 10 minutes at a speed of 3000 revolutions, per minute. The supernatent was evaporated to dryness in a 40°C water bath. The residue was immediately taken up in one ml of chloroform, added 2 ml of choloroform again and the OD of the solution was measured at 348 mμ in the spectrophotometer.

The solution is transfered to a soft glass tube with a stopper and irradiated with U.V. light (kxm the lamp should be turned on 10 minutes before use). the lamp. The OD at 348 mμ is again read and the difference in OD is taken as a measure of vitamin A in the solution.

APPENDIX VI

ESTIMATION OF CAROTENE FROM PAPAYA

PREPARATION OF EXTRACTS

The ripe papaya was cut into small pieces and 75 grams of it was pulverised with 85 per cent acetone. The suspension was refluxed for about 40 minutes over a boiling- water-bath and the supernatent was decanted off. The extraction was repeated twice or thrice and the supernatent was pooled together. It is then diluted with 20 ml of 25 per cent acetone. The carotene was extracted with petroleum ether thre or four times by using 20 ml for each time. The carotene solution was concentrated by evaporating it over a water-bath until the volume was reduced to 1.0 ml and used for column chromotography.

CHROMATOGRAPHIC SEPARATION OF PIGMENTS:

Alumina slurry was prepared by using petroleum ether. Meanwhile a burette with nozzle was taken and a rubber rube was connected to the nozzle. A pinch cock was attached to it. The outlet orifice was plugged with a bit of glass wool to about one centimetre. The alumina slurry was introduced gently to a depth of 10-12 cm. During the preparation of the column, care was taken to see that no air bubble was trapped in between and the top of the column was always kept covered with a layer

of the solvent so that the column does not get dried up.

Poured in the prepared concentrated solution of carotene with a pipette without distributing the upper surface of the column. When most of it had drained, added petroleum ether little by little and allowed it to pass through until discrete bands of the different carotene were obtained.

ESTIMATION OF B-CAROTENE:

A stock standard carotene solution was prepared by dissolving 10 mg of carotene in 10 ml of petroleum ether. From this one ml was taken and diluted to 50 ml with ether so that 1.0 ml of it contains 20 γ of carotene.

Into a series of test tubes, pipetted out 1.0, 2.0, 3.0, 4.0 and 5.0 ml of the working standard solution. The volume was made upto 5 ml in all the tubes with petroleum ether. The separated yellowish orange coloured B-carotene band was eluted using 2-3 per cent acetone in ether. This eluted B-carotene solution of 5 ml was taken and the colour was then read in Klett Summerson colorimeter at 540 mμ against a balnk (5ml of petroleum ether).

APPENDIX VII

*MEAN FOOD INTAKE OF THE SELECTED TEN CHILDREN

No.	Food stuffs	1	2	3	4	5	6	7	8	9	10
1.	Cereals (g)	236	206	170	218	254	250	155	214	251	266
2.	Pulses (g)	42	8	24	35	37	37	29	13	31	47
3.	Green leafy vegetables (g)	5	36	5	5	5	5	5	5	5	5
4.	Roots and tubers	21	18	18	47	51	55	10	17	36	22
5.	Other vegetables	27	-	24	-	34	36	7	-	16	38
6.	Fruits (g)	-	4	20	1	2	2	23	2	4	4
7.	Milk (g)	-	-	114	-	17	17	125	100	17	130
8.	Fats and oils (g)	15	15	15	16	14	15	10	12	10	25
9.	Meat and fish(g)	-	-	-	-	-	-	-	-	-	-
10.	Eggs (g)	-	-	-	-	-	-	-	-	-	-
11.	Sugar and jaggery (g)	5	5	9	13	8	8	11	11	12	12
12.	Nuts and spices (g)	12	10	21	11	20	28	21	8	10	31

*The data collected from Three Day Food Weighment Survey.

APPENDIX VIII

*MEAN NUTRIENT INTAKE OF THE SELECTED TEN CHILDREN

S.No.	Nutrients	1	2	3	4	5	6	7	8	9	10
1.	Energy (kcal)	1202	892	1000	1065	1168	1183	921	1055	1142	1542
2.	Protein (g)	34.0	21.2	25.2	28.2	35.7	36.3	21.5	22.4	32.4	37.5
3.	Iron (mg)	17.5	18.7	12.2	14.4	19.3	19.2	11.6	12.2	16.8	18.1
4.	B-carotene (mg)	361	1329	521	252	368	377	432	346	317	477

* The data collected from Three Day Food Weighment Survey.

29/1/97