

# **NIN**

## **ANNUAL REPORT 1999 - 2000**



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## **HIGHLIGHTS**

The dawn of the new millennium also brings with it more challenges and problems to tackle. The Institute has clearly focused its research efforts to face the twin threat of chronic undernutrition and diet-related chronic diseases, which have emerged as a major public health hazard. Several thrust areas have been identified including maternal child nutrition, body composition and energy metabolism, micronutrient deficiencies, diet-related non-communicable diseases, pre-clinical toxicology and food safety. Due emphasis has also been given to research related to public health nutrition.

The brief write-up presented here in the form of research highlights projects the diverse research activities of the Institute aimed at finding practical solutions to various unresolved issues in the field of nutrition. The research endeavours of the Institute cover hospital, laboratory and community based studies.

### **1. COMMUNITY STUDIES**

#### **1.1 First Repeat Survey of Tribal Population**

The National Nutrition Monitoring Bureau (NNMB) conducted a repeat survey to carry out diet and nutritional assessment of tribal population living in the Integrated Tribal Development Agency (ITDA) areas by visiting the same villages that were surveyed earlier. The results revealed that there was a reduction in the extent of severe undernutrition among preschool children and improvement in their heights and weights. There was also a marginal increase in the intake of cereals and millets at individual level.

#### **1.2 Nutritional Status of Adolescents – NNMB**

The data on adolescent girls, collected through NNMB surveys during 1996-97, was analysed with an aim to assess their current diet and nutritional status. The data was also compared with that obtained in 1975-79 from the same villages. There was an increase in the intake of cereals and millets over the past 25 years among the adolescents. Secular changes in heights and weights were observed with an increase of about 2.5 to 3.5 cm and 1 to 1.5 kg in mean heights and weights respectively. The extent of undernutrition as measured by weight for age and stunting (low height for age) also showed a decline during the current survey as compared to 1975-79.

#### **1.3 Diet and Nutritional Status of the Elderly**

The health and nutritional status of old people has been receiving considerable attention in recent years. In view of this, data collected on elderly during 1996-97 by the NNMB

surveys and the district profile surveys conducted in 1994 in non-NNMB States of Punjab, Haryana, Himachal Pradesh and Assam were analyzed and compared with that of 1975-79 NNMB surveys. The results showed that the consumption of cereals and millets was more than the Recommended Dietary Allowances in all the age and gender groups. Except in the age group of 80 years and above, the consumption of energy has improved over a period of time. The extent of Chronic Energy Deficiency (CED) appeared to be higher among the aged population than their younger adult counterparts. There was a considerable reduction in the prevalence of CED as compared to 1975-79. The study stresses the need for carrying out comprehensive health and nutrition surveys to assess the extent of chronic diseases and behavioural problems among the elderly.

#### **1.4 District Nutrition Project on Micronutrient Deficiency Disorders**

As a part of the nine State multicentric ICMR Task Force Study, a research project was carried out in Mahaboobnagar district of Andhra Pradesh to develop a need-based service delivery intervention model for the control of micronutrient deficiencies and to assess its efficacy. It is envisaged to apply this model at the district level with the use of the existing health infrastructure. Baseline data collected during the Phase-I of the project revealed a very high prevalence of night blindness (5%) among pregnant women and of Bitot's spots (0.4%) among children below 5 years of age. Goitre prevalence of 13% in the children of 6-12 years indicated that IDD was endemic in this district. The prevalence of anaemia among pregnant and lactating women and adolescent girls was above 90% while the prevalence of severe PEM was 3.2% among <5 year old children. The coverage of vitamin A and iron and folic acid supplements was found to be very low.

#### **1.5 Micronutrient Status, Mental and Physical Performance and Bone Health of School Children Supplemented with Special Nutritional Beverage**

A double blind randomised placebo controlled study is in progress to evaluate the impact of supplementation of a cocktail of micronutrients in the form of a specially fortified beverage on physical and mental development of school children with a positive placebo control. Analysis of baseline data indicated that the boys achieved higher IQ as compared to girls.

#### **1.6 Low-Cost Nutritious Supplements**

Since Red Palm Oil (RPO) is a rich source of  $\beta$ -carotene, in collaboration with AP Foods, NIN has developed a RPO (carotino) fortified ready-to-eat food as a nutritious supplement for children. Stability and acceptability of the product were found to be satisfactory according to a preliminary study.

#### **1.7 Low-cost Nutrient Supplement for Malnourished Children - A Biotechno-**

## **logical Approach**

Low-cost nutrient supplements were developed jointly by NIN, CFTRI and NDDB with the financial support of DBT. The nutritive and biological values as well as the organoleptic properties of three sweet and spicy supplements were investigated. Taste, colour, appearance, texture, aroma, after taste and shelf-life of these preparations were studied and their overall acceptability was assessed.

## **2. ENERGY METABOLISM**

### **2.1 Sports Nutrition/Work Physiology**

Intensity of training has been found to determine the energy expenditure of sportspersons involved in different sports events (sprinting, middle- and long-distance running). As the energy requirements were significantly influenced by variation in training load, appropriate dietary recommendations have to be made to achieve optimal body weight, body composition and peak performance level in a desirable way during actual competitions.

## **3. NUTRITION AND INFECTION**

### **3.1 Serum Transferrin Receptor in Children Suffering from Infections**

To define iron status in children, normal levels of serum transferrin receptor (STR) were determined. In addition, the effect of infections on STR was also evaluated. The mean serum STR was  $6.08 \pm 0.48$  mg/L in children who had haemoglobin levels more than 11 g/dl and this value is similar to the value for healthy children reported in the literature. Using this value as cut off level, it was observed that more than 50% of children having Hb more than 11 g/dl also had elevated levels of receptor, thus identifying the wide prevalence of iron deficiency in children. Ferritin, which was also estimated, failed to identify even severe anaemia. Mean receptor levels showed a significant negative correlation with Hb levels while ferritin had no correlation with Hb or STR. Hence, unlike Hb which diagnoses only anaemia, STR appears to be a good indicator to assess iron stores in preschool children.

Effect of infection on STR was determined in 119 preschool children attending the hospital for various infections who were followed up at 15-30 days after the infection. There was no difference in mean receptor levels in children with or without infection with similar haemoglobin levels, thereby indicating that in general, STR is a good indicator of iron status even in the presence of infection unlike ferritin which shows an acute phase response.

The percentage of children having anaemia and elevated STR levels was similar.

However, STR values returned to basal level by 30 days after infection. These observations suggest that anaemia during infection is due to iron deficiency which could be due to lack of storage iron or non-mobilization of stored iron in the non-anaemic children. These results emphasise the importance of controlling infections while improving iron status of children.

### **3.2 Diarrhoea due to *E.coli* 0157 H7 in Children**

In a pilot study conducted on 100 children, *E.coli* 0157 H7, which often causes acute haemorrhagic diarrhoea, was isolated from the stools of two children. The diagnosis was confirmed by sero typing and also by referring to CRI, Kasauli. This study indicates the existence of *E.coli* 0157 H7 as a causative factor for acute diarrhoea in children in this part of the country and suggests the need for more indepth studies to determine its prevalence, nature and outcome of infection in relation to nutritional status.

### **3.3 Outcome of Acute Bacterial Meningitis in Relation to Nutritional Status in Children**

The role of nutritional status and secretion of TNF- $\alpha$  in children suffering from acute bacterial meningitis and the outcome of the disease were determined. Bacterial isolation was found to be positive in 76% of CSF samples with *S.pneumoniae* being the most common organism. *N.meningitis*, *H.influenzae*, *E.coli* and *Staphylococci* also contributed to meningitis. TNF- $\alpha$  was detectable in 69% of all cases and children with *S.pneumoniae*, *N.meningitis* and *H.influenzae* as causative organisms had higher levels of TNF- $\alpha$  compared to the others. Though TNF- $\alpha$  levels were similar among various nutritional grades, the percentage of children producing the cytokine was higher in severe PEM group. Twenty-two percent of the children had died and 40% recovered with sequelae. Severely malnourished children had significantly higher adverse outcome compared to better nourished groups. *S.pneumoniae* contributed to nearly 50% of deaths. These observations suggest that nutritional status and causative organism appear to contribute to the outcome of acute pyogenic meningitis in children. However, TNF- $\alpha$  did not influence the outcome significantly. Similarly, nutritional status had no effect on levels of TNF- $\alpha$ .

## **4. MICRONUTRIENTS AND TRACE ELEMENTS**

### **4.1 Safety of Administering Vitamin A along with Polio Vaccine on National Immunization Days in the State of Orissa**

A rapid safety survey was carried out in Orissa to study the feasibility of administering both vitamin A and oral polio vaccine simultaneously to 12-42 month old children. It was found that this twin-drug administration approach improved the coverage of the national programme against vitamin A deficiency and was also found to be feasible.

#### **4.2 Trends of Xerophthalmia from 1980-1999 - A Retrospective Hospital-based Study**

The data collected from the records of Niloufer Hospital for Women and Children during 1980-99 on trends of severe vitamin A deficiency manifesting as blinding or potentially blinding corneal lesions revealed that a total of 540 children with corneal lesions attended the hospital. On an average 22 to 28 cases were observed per year between 1980 and 1999. The occurrence of corneal lesions was maximum among children aged 1-3 years followed by those between 3 and 5 years. Since vitamin A deficiency is still prevalent in some parts of the country, there is a need to target 1-5 years old children and control the scourge of vitamin A deficiency using innovative methods.

#### **4.3 Effects of Food-based Vitamin A Supplementation during Pregnancy on Maternal and Child Health**

With increase in realization of the importance of vitamin A status in maternal and child health, a cross sectional study was conducted among pregnant women during their third trimester to assess the extent of the problem of clinical and subclinical vitamin A deficiency and relate it to the pregnancy outcome. Night blindness was observed in 5% of the women, and about 60% of them had serum retinol levels less than 30  $\mu\text{g}/\text{dl}$ . Women with night blindness and those with low retinol levels had a significantly higher occurrence of pregnancy induced hypertension, pre-term deliveries and asphyxiated babies. These results suggest the need to undertake indepth studies to explore the role of vitamin A status on maternal health and pregnancy outcome.

#### **4.4 Beneficial Role of *Solanaceae* Family Plant Leaf Powder in Poultry Feed**

Earlier studies have shown that leaves of *Cestrum diurnum* (CD) belonging to *Solanaceae* family are rich sources of vitamin D<sub>3</sub>, the vitamin responsible for monitoring calcium utilisation. The effectiveness of adding these leaves to poultry feed was studied. Optimal use of calcium in chicks may augment egg production and improve egg-shell thickness. Broilers fed CD leaf powder showed better feed efficiency [Feed efficiency ratio = Feed intake (Kg)/Body weight (Kg)] and improved bone weight and bone density. The CD leaves may thus prove to be a cost-effective solution to boost poultry farming.

#### **4.5 Relationship Between Plasma Homocysteine Level and Folate and Vitamin B<sub>6</sub> Status**

Elevated plasma levels of homocysteine are associated with an increased risk of coronary heart disease. Since folate and vitamin B<sub>6</sub> are related to homocysteine metabolism and their deficiencies are also common in our population, a study has been undertaken to examine the inter-relation between plasma homocysteine levels and folate and vitamin B<sub>6</sub> status in normal subjects. Sub-clinical folate deficiency has been found to

be a significant factor contributing to the higher plasma homocysteine level among Indians as compared to the western population.

#### **4.6 Molecular Mechanisms of Immunosuppression in Severe Protein Energy Malnutrition**

The mRNA gene expression for cytokines IL<sub>2</sub> and IL<sub>4</sub> and cytokine profile of lymphocyte culture supernatants for IL<sub>2</sub> were examined in children suffering from anaemia and compared with healthy non-anaemic children aged 1-5 years. Normal children had mRNA expression for IL<sub>2</sub> and IL<sub>4</sub>, whereas anaemic children had only for IL<sub>4</sub>. The IL<sub>2</sub> levels were undetectable in their culture supernatants. Since anaemia is widespread among children, these observations are important, particularly in relation to the ongoing immunisation programmes involving newer vaccines.

#### **4.7 Effect of Different Cereals on Fluoride Deposition in Bone**

Fluoride retention in the body and progression of fluorosis are reported to be influenced by the type of cereal used as staple diet. In a rat experiment carried out to study the effect of jowar, wheat and rice-based diets on fluoride retention and deposition in bones, it was observed that as compared to rice and wheat-based diets, jowar-based diet led to higher fluoride retention.

#### **4.8 Transferrin Receptor**

The indigenous sandwich ELISA developed for the quantitation of serum transferrin receptor (STR) has been validated in adolescent girls receiving 60 mg of iron supplement for 100 consecutive days. The changes in iron status correlated well with STR and not with serum ferritin. The results of the study confirmed that the indigenous sandwich ELISA technique developed for STR is a good indicator of iron status.

### **5. DIET AND NON-COMMUNICABLE DISEASES**

#### **5.1 Studies on Fenugreek Seeds**

Galactomannan, the soluble dietary fibre present in the fenugreek seeds, was isolated and its effect on the blood glucose and cholesterol levels of obese rats (WNIN/GR-Ob) was studied. It was observed that incorporation of galactomannan into the control diet of male and female rats at 2.5 and 5.0% levels could significantly decrease plasma glucose and cholesterol levels at the end of 9 weeks, while a significant increase in the above parameters was observed in animals receiving control diet alone.

#### **5.2 Effect of Spice Principles on Cataractogenesis**

As increased oxidative stress in the lens is implicated in cataractogenesis, a study was undertaken to assess the protective effect of eugenol (active principle in cloves) on biochemical changes associated with opacity in rat lens. It was found that eugenol protects rat lens against  $\text{FeSO}_4$ -ascorbate-induced biochemical damage *in vitro*.

### 5.3 Biochemical and Metabolic Studies with Sesame Lignans

The high oxidative stability of sesame oil is attributed to the lignans namely, sesamol (S1), sesamin (S2) and sesamol (S3). The S2 and S3 are present in seeds and oils. On heating the oils to frying temperature, S1 is formed. The antioxidant properties of S1 (Sigma Co.), S2 and S3 (isolated and crystallized in our lab) in comparison to BHT and tocopherols ( $\alpha$  and  $\gamma$ ) were studied in *in vitro* enzymatic (cumene hydroperoxide induced) and non-enzymatic (iron-ascorbate induced) lipid peroxidation systems. The antioxidant activity in cumene hydroperoxide system was in the following order : BHT> $\alpha$ T,  $\gamma$ T>S1>S2>S3. The antioxidant properties of lignans in combination with tocopherols showed enhanced inhibitory effects. In the non-enzymatic system, only S1 had inhibitory effects. However, sesame lignans in combination with tocopherols enhanced the inhibitory effects in the non-enzymatic peroxidation system. These findings suggest that sesame lignans can potentiate the antioxidant properties of tocopherols and contribute to their beneficial role as dietary antioxidants.

## 6. FOOD CHEMISTRY

### 6.1 Nutrient Composition of Foods

The analysis of total lipid and individual fatty acid contents of selected fresh fruits showed that they contain <1 % total lipids. The  $\alpha$ -linolenic acid (ALNA) content is higher in plum, papaya and guava (0.4%) as compared to other fruits (0.2%). On an average, about 400 g of fresh fruits provide ~ 0.1 g ALNA. It was reported earlier that on an average ~ 60 g of green leafy vegetables or ~400 g of other vegetables provide 0.1 g of ALNA. Thus, fruits and vegetables can further the n-3 PUFA intake, in addition to providing several other nutritional benefits.

### 6.2 Fats in Bakery Products

*Vanaspati* is widely used in the preparation of bakery foods. The data obtained shows that biscuits furnish ~ 12% lipids, ~ 5% saturated fatty acids (SFA) and 4% *trans* fatty acids (TFA). Thus, consumption of 7-10 biscuits would furnish about half of the upper safe limit of SFA and TFA put together.

### 6.3 Mineral and Trace Element Contents of Foods

Apart from reassessing the mineral and trace element composition of commonly consumed plant foods, 20 foods of animal origin were also analysed using atomic absorption spectrophotometer. It was found that country hen eggs had significantly higher amounts of trace minerals like calcium, phosphorus, copper and manganese as compared to poultry hen's eggs.

## **7. EDUCATION AND EXTENSION**

Recently published "Dietary Guidelines for Indians - A Manual" as well as the multicoloured handy booklet "Dietary Guidelines" continued to be popular among the public. Based on this, a special publication "Dietary Guidelines for Vulnerable Groups" was brought out in Telugu, in collaboration with CARE for free distribution to health functionaries. In addition, translation work of the publication in other Indian languages is in progress.

Besides the regular training courses, two new short-term training courses were conducted this year. Since iron deficiency anaemia is widespread, a training course on "Techniques for Assessment of Nutritional Anaemias" was organised in collaboration with Biophysics Division for teachers of medical colleges and public health personnel. In addition, ICMR sponsored Workshop on "Information, Education and Communication in Biomedical Sciences" was conducted for 17 scientists representing various ICMR Institutes and Centres. The Institute continued to attract WHO Fellows from neighbouring countries.

## **8. FOOD & DRUG TOXICOLOGY**

### **8.1 FOOD SAFETY**

#### **8.1.1 Application of HACCP to *Paneer***

The Hazard Analysis Critical Control Point (HACCP) approach which is now well accepted as the best preventive strategy in food safety and quality to prevent foodborne hazards was used for the toxicological evaluation of *Paneer* (Cottage Cheese). After identifying the Critical Control Points (CCPs) in the manufacturing process, it was found that the microbiological contamination of the final product could be considerably reduced by intervention strategies like improving the personal hygiene of the food handlers.

#### **8.1.2 Ochrotoxin in Coffee**

Studies on occurrence of ochrotoxins in Indian coffee had revealed that although ochrotoxin A is present in Arabica, Cherry and monsooned coffee samples, its level is well below the limit of 5 µg/kg level which is considered by the European Union countries as a safe limit.

#### **8.1.3 Antinutritional Factors in Wild Legume *Cassia tora***

Studies on the seeds of wild legume *Cassia tora* indicated the possibility of extracting gum from the seeds using a simple process. This gum can be used as a thickener in food products. The seed germ meal obtained as a by-product of gum extraction though rich in protein is found to contain antinutritional factors. A detoxification method to remove antinutritional factors has also been evolved. The PER and short-term toxicity studies indicated that the detoxified germ could be used as an ingredient in the animal feed.

#### **8.1.4 Analysis of Argemone Oil in Adulterated Oils**

A collaborative study on validation and harmonization of analytical method for quantitation of argemone oil in adulterated vegetable oil revealed the efficacy of the method in detecting adulteration of argemone oil in commonly used edible oils. However, in the case of non-edible oil like Karanjia oil, the analytical method gave false positive results.

### **8.2 NUTRITION AND CANCER**

#### **8.2.1 Biomarkers of Genotoxicity : Antimutagenicity of Heated Garlic**

Our earlier studies have demonstrated antimutagenic property of garlic in rats exposed to Benzo(a)pyrene (Ann.Rep. 1996). In order to verify whether this property is still retained after subjecting the garlic to cooking, a study with cooked, boiled and unboiled garlic was undertaken with *S. typhimurium* TA 98 as tester strain. The results showed that the antimutagenic principles in garlic are not destroyed by heat treatment.

#### **8.2.2 Studies on Certain Medicinal Plants from North-Eastern Region of India**

In collaboration with IASST, Guwahati, some plants used by tribals in North-Eastern India were tested for their ability to counter hepatotoxic effect in a paracetamol-induced liver toxicity model. Aqueous and methanolic extracts of the plants *Costos* and *Leucos Levandafolia* were tested. Extracts of both plants have been shown to exert hepatoprotective effect as evidenced by a decrease in SGOT and SGPT enzyme activities and on the basis of histopathology examination.

Further studies in medicinal plants revealed that the methanolic or aqueous extracts of two plants - *Leucos levandafolia* and *Costos levandafolia* of the north-eastern region of the country confer protection against liver toxicity effectively.

In order to understand the underlying mechanism involved, the antioxidant potential of the extracts of the above plants, besides others like C-Cajan, glycosmis and sida rex, were tested using *in vivo* linoleic oxidation assay. Many plant extracts exhibited significant antioxidant potential as demonstrated by a reduction in MDA formation in *in vitro*

assay. The hepatoprotective properties of the extracts of the plants studied may probably be related to their antioxidant properties.

### **8.2.3 Drug Metabolism in Obese Rats**

Since excess body fat is known to be associated with increased risk of cancer, it was thought that obese rat model developed at NIN could be a suitable model for fat-cancer studies. Before undertaking such investigations, the information on the levels of drug metabolising enzymes, namely, Benzo(a)pyrene hydroxylase, microsomal epoxide hydrolase (Phase I), uridine diphosphoglucuronyl transferase (UDPGT) and glutathione-s-transferase (GST) (Phase II) were estimated in homozygous obese, heterozygous obese and lean rats. The activities of conjugating enzymes GST and UDPGT, which play an important role in detoxifying the xenobiotics, were significantly low in the homo and heterozygous obese rats as compared to lean control, thereby suggesting that the elimination of ingested xenobiotics may be impaired in such animals. These results suggest that obese rat model could be useful in studying xenobiotic metabolism.

### **8.2.4 Effect of Varying Carbohydrate Diets on Glucose Intolerance in WNIN/GR-Ob rats - Histopathology Study**

The effect of 20% stock protein, glucose-, sucrose- and starch-based diets on fasting glucose levels of obese rats was studied. Histopathological studies done to assess organ changes occurring due to varying carbohydrate diet revealed fatty changes in liver of all obese animals while no significant changes occurred in their pancreas and kidneys. The composition of the diet thus had no adverse effect on organ morphology.

### **8.2.5 Chlorella and $\alpha$ -interferon**

Toxicological evaluation on chlorella has been completed. The results in general do not indicate any serious toxic effect.

## **9. NATIONAL CENTRE FOR LABORATORY ANIMAL SCIENCES (NCLAS)**

NCLAS is involved in both service and research activities. Although breeding and supply of laboratory animals came down by 30% during 1999 due to the delay in the registration of the Centre with CPCSEA, research activities related to obese rats continued. These studies have shown that leptin gene amplification was highest for obese rats as compared to lean and carrier animals. However, nucleotide sequencing of leptin gene isolated from obese animals did not show any change/abberation indicating that the mutation in these animals is not with respect to leptin. The melanocortin receptor (MCR-4) expression in obese rat was found to be low as compared to lean and carrier animals. The random primer OPB<sub>15</sub> (Operon Technologies Inc., USA) was able to distinguish WNIN/Ob rats from WNIN stock rats as indicated by PCR based DNA fingerprint. The obese mutant stock (including lean, carrier and obese) as a whole was found to be at a greater risk of oxidative damage compared to parental WNIN rat stock, as shown by high levels of

protein carbonyls and MDH. Correspondingly, the GSH levels were found to be significantly low in these animals. Amongst the tissues studied, brain showed consistent aging pattern with respect to all parameters. The opioid receptor levels (Delta, Kappa, Mu) were significantly reduced in obese animals compared to WNIN control. Binding studies showed lack of leptin receptors in obese animals.

Preliminary studies on spices revealed hypoglycaemic effect of curry leaves and hypolipidemic effect of turmeric in WNIN/GR-Ob rats.

A judicious blend of basic and applied research is the hallmark of the Institute's research endeavours. The rapidly changing nutrition scenario in the country is posing a challenge to the nutritionists today. The Institute is gearing itself to accept the challenge and with the able support of its staff, it is evolving suitable strategies to combat malnutrition in the country.

I sincerely thank the dedicated staff for their co-operation and conscientious efforts to enhance the content and quality of research projects, the results of which will have a positive impact on malnutrition.

**Dr.Kamala Krishnaswamy**  
Director

## **I. COMMUNITY STUDIES AND OPERATIONAL RESEARCH**

### **1. Diet and nutritional status of tribal population - Results of repeat surveys (NNMB)**

The National Nutrition Monitoring Bureau (NNMB) which has been carrying out annual diet and nutrition surveys of rural population in ten States of the country, had carried out diet and nutrition assessment of tribal populations living in the Integrated Tribal Development Agency (ITDA) areas during the years 1985-87. The same tribal villages, that were covered during 1985-87 were resurveyed during the period 1998-1999. The objectives were : (i) to assess the food and nutrient intake of tribal population, (ii) to assess their nutritional status in terms of nutritional deficiency signs and anthropometry, and (iii) to assess the changes, if any, in dietary pattern and nutritional status over the period of time.

The survey was carried out in the States of Andhra Pradesh, Gujarat, Karnataka, Kerala, Maharashtra, Madhya Pradesh, Orissa, Tamil Nadu and West Bengal. In each State, a total of 120 villages were selected for survey, of which 75% were from those surveyed in 1985-87, while the rest were new villages. From each selected village, 40 households (HHs) were covered for anthropometry and clinical examination of all the available individuals. The information on socio-economic and demographic particulars was also collected from these HHs. A sub-sample of 10 HHs was selected by adopting systematic random sampling for conducting 24-hour recall diet survey at the individual level. Data on food and nutrient intake was collected from 32,023 individuals from 8,036 HHs. About 90,885 individuals were covered for anthropometry and clinical examination from 30,390 HHs.

Results indicated the following:

#### **Food and Nutrient Intake**

The intakes of all foods, except cereals and millets among adults, were less than RDA in all the age groups. Only in the States of Orissa and West Bengal, the mean intake of green leafy vegetables was higher than RDA. The intake of protective foods such as milk and milk products, fats and oils and sugar and jaggery was much below the suggested levels in all the age groups in all the States surveyed. There was no perceptible difference between the tribal population and rural communities with respect to the percentage of individuals consuming less than 70% of RDA with respect to cereals, millets and pulses, except GLV.

#### **Time Trends**

1. During the repeat survey, a marginal decrease in the intakes of cereals and millets at the individual level was observed, while the consumption of other foods remained essentially similar as in 1985-87 in all the age groups.

2. The intake of various nutrients such as protein, energy, iron, thiamin, riboflavin, niacin and vitamin C decreased marginally in various States and in most of the age groups in 1998-99 as compared to the levels observed during 1985-87. However, there was no perceptible change in the intake of vitamin A, vitamin C and calcium among all the groups.

3. The per cent prevalence of frank signs of protein energy malnutrition among preschool children declined to 0.5% in 1998-99 from about 4% in 1985-87. There was also a decline in the prevalence of Bitot's spots (0.5%) in 1998-99 as compared to 1985-87 (2.1%). The same trend was observed in other age groups.

4. The percentage of severe undernutrition among girls (7.8%) was marginally low as compared to boys (9.3%), while that of normals among girls was higher (6.5%) as compared to boys (4.4%).

5. The proportion of 1-5 year children with severe degree of undernutrition (<60% weight for age of NCHS standards) decreased from 19.8% in 1985-87 to 8.6% during the present survey with concomitant increase in the proportion of normal and grade-I (mild) undernutrition (**Fig.1**). The reduction was observed in all the States, except in Tamil Nadu and Maharashtra.

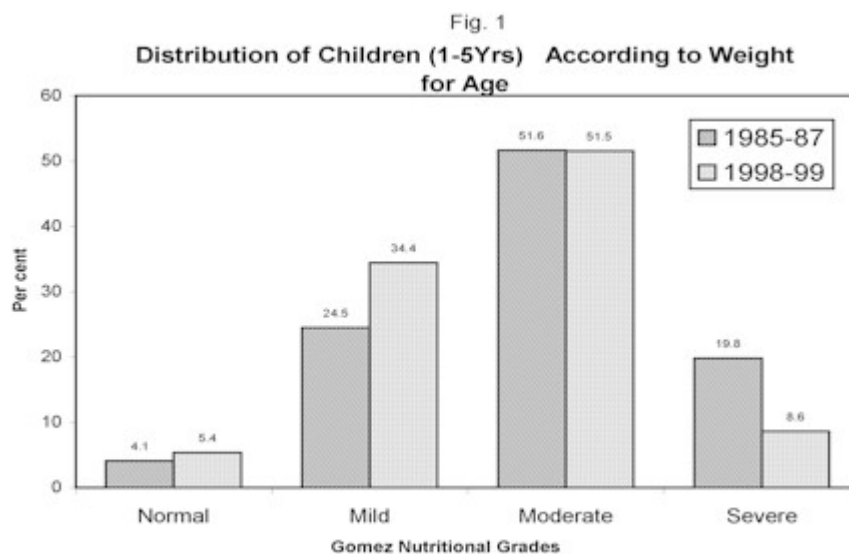
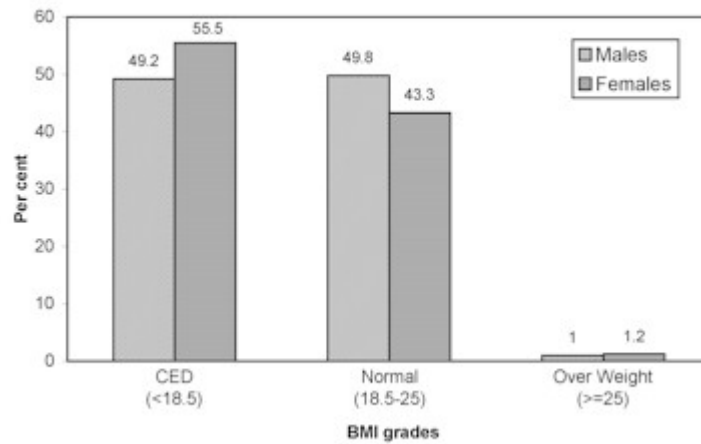


Fig. 2  
Distribution (%) of Adults by BMI



6. About 72% of preschool children had weights less than Median-2SD of NCHS standards. Their proportion was higher than reported by the NNMB in the rural areas (61%). Almost two-thirds of the children (63%) had stunting.

7. The prevalence of chronic energy deficiency (BMI <18.5) in adult males was lower (49%) than in females (56%) (**Fig.2**).

Thus, the results of the repeat surveys in the tribal areas showed a reduction in severe undernutrition among preschool children and improvement in heights and weights, despite marginal decrease in the food and nutritional intake. This, perhaps, may be attributed to the role of non-nutritional factors like improvement in the housing conditions, safe drinking water and better out-reach of health care services coupled with improvement in socio-economic conditions.

## 2. District nutrition project for prevention and control of micronutrient deficiency disorders

The National Nutrition Policy envisages (i) eradication of nutritional blindness in children, (ii) eradication of iodine deficiency disorders, and (iii) reduction in the prevalence

of anaemia in pregnant women to about 25%. The ICMR undertook a 9 State Multicentric Task Force Study including Andhra Pradesh. The project in Andhra Pradesh was carried out by the NIN. The aim was to develop a need-based service delivery intervention at district level through the existing district health system and to assess its efficacy and examine the feasibility of its replication for prevention and control of micronutrient deficiencies. The project was (proposed to be) implemented in 3 phases. In the phase I, baseline data was collected to find out the extent of the micronutrient deficiencies and to assess the current status of ongoing programmes. While the phase II was an intervention phase to strengthen the national programmes and the IEC programmes in the community, the phase III involved evaluation of the impact of intervention.

### **Area and Methods**

The district of Mahaboobnagar was selected in consultation with the officials of the Department of Health of the Government of Andhra Pradesh. For the purpose of baseline survey, a 30-cluster sampling was adopted. Village and household information, clinical assessment for vitamin A deficiency (VAD), PEM and goitre, estimations of haemoglobin (Hb) and urinary iodine were carried out. Body weights of infants and preschool children were recorded.

### **Sample**

About 10,300 preschool children and 10,000 children of 6-12 years were surveyed for VAD and iodine deficiency disorders (IDD) respectively from 10,521 households from 29 selected clusters. Haemoglobin estimation was done on 189 pregnant women, 200 lactating mothers and 294 adolescent girls. Forty-eight functionaries of different National Nutrition Programmes for iron deficiency anaemia (IDA), VAD and IDD were interviewed for their knowledge and perceptions about the micronutrient deficiency disorders and the status of the national nutrition programmes.

Results are summarised below:

### **Prevalence**

The prevalence of Bitot's spots was 0.4% among children of 1-3 years. The prevalence of night blindness was 1.1% among children of 2-3 years, while it was to the extent of 5.2% among the pregnant women. Goitre was observed in 13% of 6-12 year children and the proportion of children with low urinary iodine excretion levels (<10 mg/dl) was 38%, thus indicating high endemicity of IDD in the district. The prevalence of anaemia among pregnant (<11 g/dl) and lactating women (<12 g/dl) was 92.1% and 90.5% respectively. Among the adolescent girls, the prevalence of anaemia (< 12 g/dl) was 92%. The prevalence of severe anaemia was 12.7% in pregnant women (<7 g/dl), 10.5% in lactating mothers (<8 g/dl) and 9.2% in adolescent girls (<8 g/dl). Protein Energy Malnutrition, as assessed by weight for age (<75% of NCHS standards), was observed among

32.6% of the children of 0-6 years, while 3.2% of the children were in severe grades of PEM (<60% of NCHS standards).

### **Status of Intervention Programmes**

The coverage for vitamin A supplementation was very low (11.2%) in children (<3 years). It is intriguing that in spite of the low coverage under the massive dose vitamin A programme, the prevalence of signs of vitamin A deficiency is low. Logistic management with respect to massive dose of vitamin A and iron and folic acid (IFA) tablets distribution in the Department of Health was unsatisfactory. The coverage for iron and folic acid tablets distribution by pregnant women was 52%, and 95% of them stated that they were consuming the tablets. However, only 15.7% of them consumed 90 or more tablets. Only 4.2% of the households in the area used iodized salt. The analysis of salt samples revealed that only 8.3% samples had 15 ppm of iodine, the recommended level of iodine, in the salt at the household level. Most of the functionaries interviewed were aware of the night blindness, Bitot's spots and details of vitamin A prophylaxis programme. Their awareness about iron deficiency anaemia, IFA tablet distribution, iodine deficiency disorders was satisfactory. However, the awareness of the programmes among the community was poor.

The study has since been terminated after Phase I was completed as per the instructions of ICMR.

### **3. Nutrition profile of Indians (NPI) - A district level survey in Orissa**

Earlier, results of nutrition profile of population of the States of Assam, Haryana, Himachal Pradesh and Punjab were presented (NIN Ann. Repts. 1995-96 & 1996-98). The survey was extended to the state of Orissa to assess the food and nutrient intake of rural and urban communities and to assess the nutritional status of the representative segments of populations in terms of anthropometry and clinical status.

#### **Sample**

The survey was carried out in all the thirty districts and twenty urban wards from the selected towns which formed the urban sample equivalent to the sample size covered in the rural areas of one district. Twenty villages were selected giving due representation to all the blocks/taluks in the district by systematic random sampling procedure, coupled with probability proportion to size (PPS). Within each selected village, twenty households (HHs) were selected by cluster sampling method. A total of 5931 HHs were covered for household particulars and family diet survey. About a half of the HHs were covered for individual diet survey by 24-hour recall, and nutritional assessment was done on 51934 subjects of different age and sex groups.

Results are summarised below:

### **Food Intake (CU/day)**

The average intake of rice (585 g), the staple, was well above the RDI of 460 g. However, the intake of pulses (31 g) was below the RDI of 40 g. Consumption of income elastic foods such as milk (17 ml), visible fat (8 g), sugar and jaggery (11 g) was very low compared to RDI.

### **Nutrient Intake**

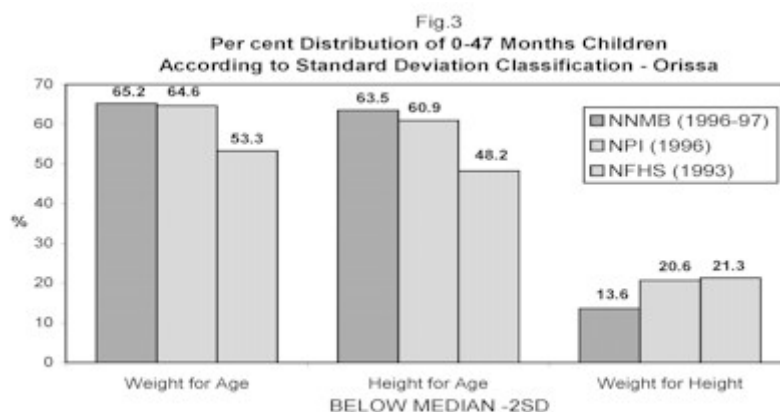
The intakes of energy (2419 Kcal), protein and calcium were comparable to the recommended intakes, while the intake, especially of micronutrients such as iron (14 mg), vitamin A (382 µg) and riboflavin (0.6 mg) were well below the RDI. About 68% of the HHs had adequate dietary energy intakes, while protein adequacy was observed in about 81% of the HHs surveyed. About 18% of the households have inadequate intake of both protein and energy.

### **Nutritional Status**

The anthropometry of individuals surveyed revealed that the community was considerably lighter and shorter than the international (NCHS) standards. The proportion of severe undernutrition (< 60% weight-for-age) in preschool children was about 8% with no gender differences. However, the proportion of normal girls (>90% weight-for-age) was slightly higher (10%) than in the boys (8%). The proportion of moderate and severe undernutrition (<75% weight for age) was higher among 1-3 year old children (55%) than among the 3-5 year old children (47%). About 59% of the children (0-47 months) underweight (weight-for-age <Median-2 SD of NCHS standards) with about 25% of the children having severe undernutrition (weight-for-age < Median-3 SD). About 61% of children had stunting (< Median-2 SD of height-for-age), while 32% had severe stunting (< Median-3 SD height-for-age) indicating chronic undernutrition among children. The reported figures by National Family Health Survey (NFHS) for stunting (48%) and underweight (53%) are lower (**Fig.3**). Around 21% of the children had wasting (<Median -2 SD of weight for height) which is comparable to the NFHS (21.3%).

In the case of adults, about 45% were in different grades of chronic energy deficiency (CED) as measured by BMI (< 18.5). It was lower in males (38%) than in females (52%). Less than one per cent of the adults were obese.

Thus, these results indicate that the undernutrition is widely prevalent in the State both in preschool children and adults. Such data on diet and nutritional status of population, available for the first time at the district level, would enable the planners to prepare plan of action on nutrition at district level.



#### 4. Diet and nutritional status of the elderly - NNMB

The elderly population (60 years and above) in India, at present, is estimated to be 65 millions and is expected to cross 177 million by the year 2025. They have a host of problems, particularly with regard to socio-economic, health and nutrition. In India, there is paucity of data on health and nutritional status of the elderly. The year 1999 has been declared as the Year of the Elderly. Hence, the data on dietary pattern and nutritional anthropometry of elderly collected by the National Nutrition Monitoring Bureau (NNMB) was analyzed to assess their current diet and nutritional status. For the purpose, the dietary data collected by 24-hour recall method and information on nutritional status of elderly as assessed by nutritional anthropometry in the rural areas of Kerala, Tamil Nadu, Karnataka, Andhra Pradesh, Maharashtra, Madhya Pradesh, Gujarat and Orissa during 1996-97 were utilized. The results were also compared with similar data collected by NNMB in the same villages during 1975-79 to assess time trends, if any. In addition, the data collected in the year 1994 on Nutrition Profile of Indians (NPI) at the district level in the States of Assam, Haryana, Himachal Pradesh and Punjab was also utilized and compared.

About 922 elderly individuals were covered for 24-hour recall diet survey and 3646

for nutritional anthropometry (height and body weight) during 1996-97, while the coverage was 858 and 3659 respectively during 1975-79. The data was analysed according to the age groups (years) of 60-69, 70-79 and >80.

Results can be summarised as follows:

The average daily consumption of cereals, in general, was more than the RDA, except in the age group of >80 years among both the sexes. The consumption of pulses (29 g) was less than the RDA (40 g) in all the age groups. In fact, the intakes seem to have declined from 33 g in 1975-79. In States of Haryana, Himachal Pradesh, Punjab and Assam (NPI) surveyed during 1994, the pulse intake was 31 g.

The consumption of green leafy vegetables, which are rich sources of micronutrients and antioxidants, was much below the suggested level of 40 g in all the age groups, (1996-97: 16 g; 1975-79 : 13 g; and NPI : 27 g).

During 1996-97, the intake of milk and milk products (82 g) was below RDA (150 g) in all the age groups and among both the sexes except in the NPI (Males: 250 g; Females: 226 g).

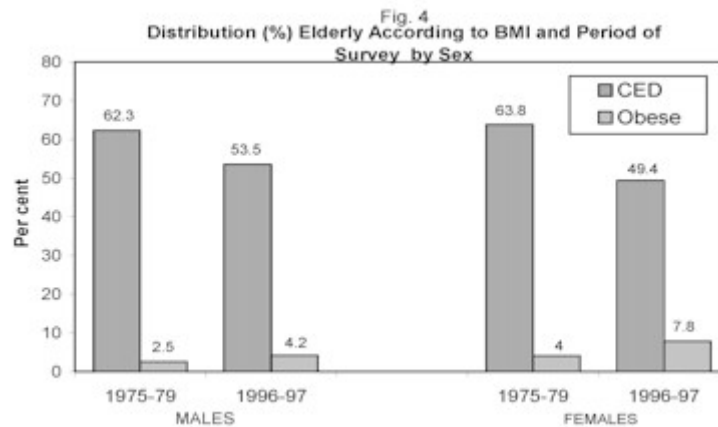
In about 71% of males and 60% of females, the consumption of energy was more than the RDA during 1996-97 (RDA for energy among elderly was calculated according to their body weights). There appeared to be an improvement from that in 1975-79 (55% and 51% respectively). The mean intake of micronutrients like iron, vitamin A was below the RDA in all the surveys.

During the period 1996-97, the mean weights and heights were 48.1 kgs and 160.4 cms among males and 42.5 kg and 148 cm among females. The corresponding figures for 1975-79 were (males: 47.2 kg. and 161.4 cm; females: 39.3 kg and 147.4 cm).

The extent of Chronic Energy Deficiency (CED) (BMI <18.5) during 1996-97 (52%) was lower than that observed in 1975-79 (63%). In the NPI States it was about 39%.

In general, the extent of CED was marginally higher among males (53.5%) than among females (49.4%) during 1996-97. There was considerable reduction in the prevalence of CED as compared to 1975-79 both in males (62.3%) and females (63.8%). However, during 1994, similar trend was observed among both the sexes (Males: 39.6%; Females: 39.2%). The distribution of BMI at two points of time is given in **Fig.4**.

The prevalence of CED among SC and ST communities was higher (65%) in the present survey than that (50%) observed during 1975-79. Similarly, the proportion of CED was higher (61%) in agricultural labourers and those living in kutcha houses during both



the periods (60% and 68%).

From these data, it was observed that with increasing age, there was a decreasing trend in consumption of food and nutrients, which may be attributed to decrease in appetite with increasing age, reduced physical activity and loss of teeth. There is also a need to collect comprehensive information on the prevalence of chronic diseases and the behavioural problems among the elderly.

## 5. Nutritional status of adolescents - NNMB

Adolescence is a period of rapid growth and maturation in human development after infancy. The nutritional status of adolescent girl, the “future mother”, contributes significantly to the nutritional status of the community. According to WHO (1986), individuals between 10 and 17 years are considered as “adolescents”. It is only recently that efforts, though small, are made to include adolescent girls in some of the intervention programmes. The information is scanty about dietary and nutritional status of adolescents from different parts of the country.

### Sample and Methods

The NNMB, through its annual surveys, since 1972, established a large database on different representative segments of population belonging to different States. It has also

conducted repeat surveys in 1988 and 1996, in the same villages that were surveyed in 1975-79. In the present report, results of analysis, carried out on the data on adolescents collected during 1996-97 are presented. These are compared with those obtained in 1975-79 to find out whether there were any time trends in the dietary pattern and nutritional status.

The anthropometric data obtained on 12,124 adolescents and 24-hour recall dietary information on 2,579 individuals in 1996-97 were analyzed to assess (i) the diet and nutritional status of adolescents from eight States, and (ii) the role of socio-economic factors on nutritional status. These results have been compared with the anthropometric data obtained on 24,683 adolescents and dietary information on 3,313 individuals obtained in the same villages in 1975-79.

Results can be summarised as follows:

The average consumption of cereals and millets increased slightly (379 to 427 g in boys; 348 to 391 g in girls) between 1975-79 and 1996-97. While the consumption of pulses and other vegetables though remained same over the period, there was a marginal increase (10-16%) in the intake of green leafy vegetables and fruits (10-20%). The proportion of adolescents consuming more than 170% RDA increased in 1996-97 as compared to 1975-79 in both boys and girls in all the food stuffs.

There appears to be secular changes in height of the adolescents. The distance charts for height and weight showed an increase to the extent of 2.5 to 3.5 cm in height and 1 to 1.5 kg in weight between 1975-79 and 1996-97 in both the boys and girls (Fig.5).

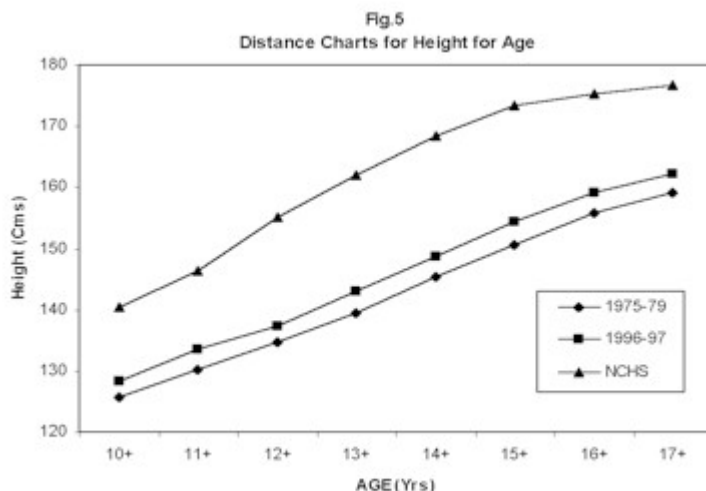
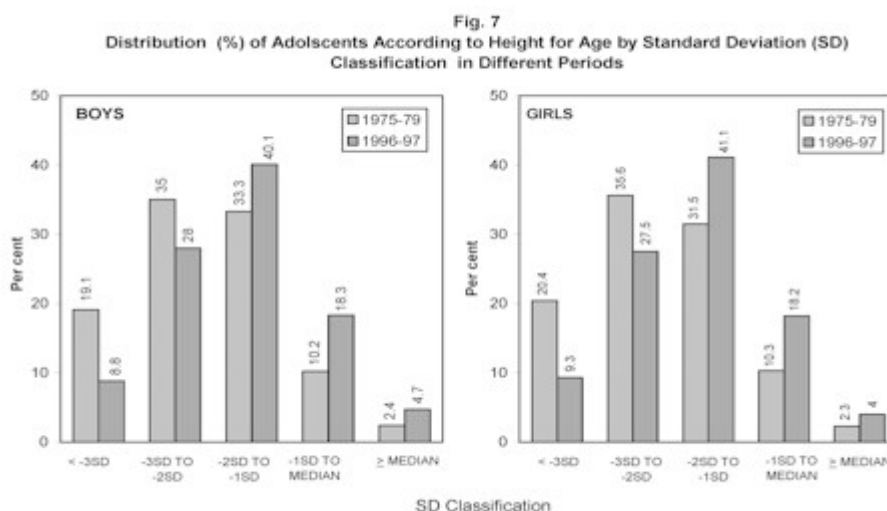


Fig. 6  
Distribution (%) of Adolescents According to Weight for Age by Standard Deviation (SD) Classification in Different Periods



In general, the distribution of heights and weights indicated that the adolescents measured during 1996-97 were taller and heavier than those in 1975-79. The extent of undernutrition (<Median -2 SD NCHS weight-for-age) in 1996-97 declined from 66.5% in 1975-79 to 51.6% in boys and 52% to 38% in girls (Fig.6). The prevalence of stunting (<Median -2SD for NCHS height for age) also showed similar changes in boys (1975-79: 54.1%; 1996-97: 36.8%) and girls (1975-79 : 56.0%; 1996-97 : 36.8%) (Fig.7).



The prevalence of Bitot's spots declined from 2.9% in 1975-79 to 0.8% in 1996-97.

These results thus indicate that the extent of stunting, wasting and undernutrition is high in adolescents indicating the urgency to develop suitable strategies to reach the adolescent girls and control undernutrition among them.

## 6. Long-term impact of ICDS on school-going rural children

One of the important components in the package of services of the ICDS programme is the non-formal preschool education for children between the ages of 3 to 6 years. An earlier evaluation indicated higher intellectual benefits to children exposed to the programme. The benefits persisted even in the face of socioeconomic deprivations (NIN Ann. Rep. 1992-93). Since it would be of considerable significance if these benefits persist even during their school age, a follow-up study was undertaken in 23 villages of Tirupati ICDS project area which was one of the centres where beneficiaries and controls had been studied earlier (NIN Ann. Rep. 1992-93).

The objectives of the study were to assess the long-term benefits, if any, on the intellectual development, cognitive functions, classroom behaviour and scholastic achievement of school children with or without early exposure to the ICDS programme and to assess independent association of other factors influencing intelligence and scholastic achievement of these children, such as, socioeconomic status, demographic features,

home environment and nutritional status.

### **Area and Methods**

A list of children from 23 villages in Tirupati block who had participated in the earlier study was used to locate the subjects for the present study. In a single blind design, the available children were tested in their homes using the following tests:

1. Binet-Kamat Intelligence Scale for Indian Children
2. Draw-man-test
3. Raven's Coloured Progressive Matrices
4. Behavioural check list for teachers' for ranking the students
5. Scholastic Achievement Test
6. Socio-economic Status Rating Scale.

In addition, information on demographic profile of all children and their heights and weights were taken. After the completion of data collection on all the children, the erstwhile ICDS beneficiaries were identified. Children not exposed to the programme were from the same villages and shared similar socio-economic and home backgrounds as reported earlier. The investigator, collecting the data, was not aware of the beneficiary or control status of the children.

### **Results**

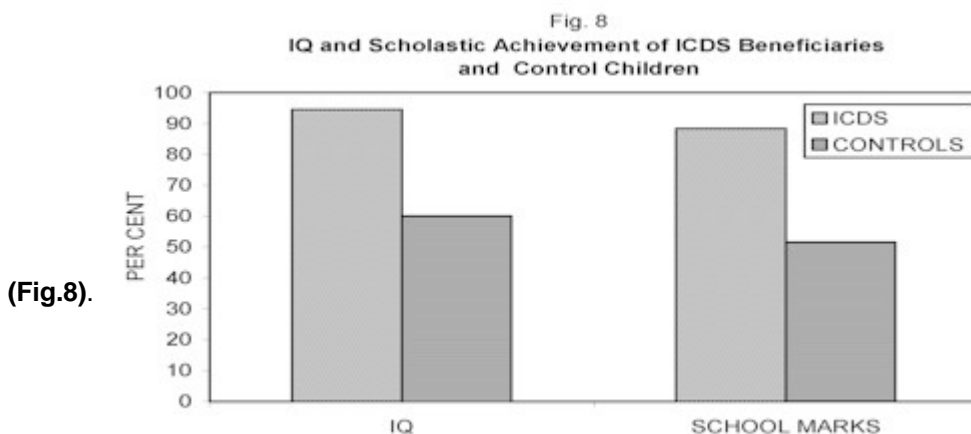
Of the 271 ICDS beneficiaries who had enrolled in school, 259 were available in the same villages for psychological testing. Among the 110 non-ICDS controls who had enrolled in school, 107 were available. The distribution of the boys and girls in both the beneficiary and control groups was similar in the age groups of 7-9 years.

The percentage distribution of children according to Gomez classification in the two study groups was comparable to the total sample studied during 1992-93 indicating that

both the groups in the present study were representative of the earlier sample. The proportion of children with less than 75% of NCHS (Grades II and III) was higher ( $P < 0.05$ ) in the erstwhile non-ICDS (66.4%) than in the erstwhile ICDS (52.1%).

Distribution according to the three IQ categories, using the Binet-Kamat test, revealed that 69% of children in the erstwhile ICDS group had normal average IQ as compared to 41% in the non-exposed group. In fact, nearly 60% in this group had below average IQ (Dull category), as compared to 23% in the ICDS exposed group ( $P < 0.01$ ). Among the erstwhile ICDS beneficiaries, about 8% had above average IQ while none of the control children had above average IQs.

Results of the Draw-a-man and Binet-Kamat IQ tests indicated that the erstwhile ICDS beneficiaries achieved significantly higher mean IQs ( $P < 0.001$ ) compared to the erstwhile non-ICDS controls across age groups



(Fig.8).

Bivariate analysis of IQ scores in relation to nutritional status (%weight for age) indicated that the erstwhile ICDS beneficiaries achieved higher scores, irrespective of nutritional status, than their erstwhile non-ICDS counterparts. In fact, the undernourished erstwhile ICDS beneficiaries achieved higher scores than even the well nourished erstwhile non-ICDS counterparts across the three age groups. Similar findings were observed in relation to height for age percent and IQ. These findings persisted even after controlling the factors such as parental education and occupation, family income and other home environmental factors.

Stepwise multiple regression analysis, carried out to find out the best set of factors explaining the variation in IQ of the study children, indicated that being a beneficiary of the ICDS during preschool years, father's occupation (middle income), presence of a TV at home, higher per capita income, presence of hygienic toilet facilities and socialization of child explained 39.3% of variation in the IQ of 7-8 year old children. For 8-9 year old children, factors such as being an ICDS beneficiary, maternal education, per capita income, socialization of the child, and safe water supply explained 52.2% of variation in their IQs. Factors including presence of TV at home, father's education, being an ICDS beneficiary and per capita income explained 57.2% variation in the IQs of 9-10 year old children.

Results on the scholastic achievement indicated that the erstwhile ICDS beneficia-

ries achieved significantly higher percentage of marks compared to the erstwhile non-ICDS controls in all the age groups ( $P < 0.001$ ).

Age-wise multiple regression analysis for the 7-8 year olds indicated that child's IQ, maternal education and being a beneficiary of the ICDS explained 57.9% of variation in the scholastic achievement. For the 8-9 year olds, none of the variables studied entered the equation. However, for the 9-10 year olds, one variable i.e., father's occupation (middle income yielding) entered the equation and explained 11.7 % variation in children's scholastic achievement. These results suggest that being a beneficiary of the ICDS is third in importance as a variable influencing scholastic achievement, the first and second being IQ and maternal education respectively. Also, the effect of the ICDS on scholastic achievement seems to last only for a year or two after joining formal school unlike its effect on IQ which was seen to last even up to three years or more.

### **Conclusion**

The children who have had the early experience of ICDS tended to have long-term benefits on their cognitive development and limited effects on their scholastic achievement. Results of the study show that the benefits were not only immediate (NIN Ann. Rep. 1992-93), but also lasted up to three years follow up. Efforts are therefore needed to upgrade the preschool educational curriculum of the ICDS to attract wider coverage and communicate its benefits to the community using social marketing techniques to make it more cost-effective.

## **7. Effect of supplementation of nutritional beverage on micronutrient status and mental and physical performance of school children**

A new blend of nutritional beverage with micronutrients was developed by a company and a sponsored project for the product evaluation was entrusted to NIN. Two phases were envisaged in the study, the first regarding formulation, the second on impact of the product.

As per the agreement, the beverage was to be tested for bioavailability of iron and calcium and impact on certain functional characteristics in children. These studies formed Phase I studies of the project. Screening different chemical sources of iron for their suitability in fortification of the nutritional beverage was one of the important points of reference in the sponsored project. Also, the hypothesis that addition of zinc compromises the absorption of iron was tested in the first set of experiments.

Therefore, the relative bioavailability of iron from the nutritional beverage prepared with five selected chemical sources of iron and the effect of added zinc on both iron and zinc absorption was carried out in a double-blind study in rats. Coded beverage samples were supplied by the Company and their key was provided by the Company after a report was received.

### **Study-1: *In vivo* bioavailability screening for chemical sources of iron**

The *in vivo* bioavailability screening for iron was based on the AOAC method of haemoglobin regeneration in iron-deficient anaemic rats.

#### **Study design**

Eighty 25-day old Wistar NIN rats of both sexes individually housed in polypropylene cages were fed *ad libitum* with a low-iron diet for 40 days so as to decrease the haemoglobin levels to  $7.6 \pm 2.28$  g/100 ml from the initial value of  $10.9 \pm 0.42$  g/100 ml. They were randomly allocated into 8 groups of 10 animals each and were rehabilitated for 4 weeks with the same diet containing the beverages with different sources of iron (14 mg iron/54 g of beverage powder which was used in the rat diet to provide 3.5 mg iron/100 g). The codes of 5 sources of iron salts tested were: (i) 101; (ii) 110; (iii) 220; (iv) 404; (v) 303, and (vi) 330. Two prospective iron salts (ii) and (iv) with additional zinc of 7 mg/54 g beverage (code 202 and 440, respectively) were also tested. Thus, a combination of 8 beverage preparations were studied.

Overall growth of the rats and the rate of recovery of haemoglobin (Drabkin's method) were monitored at 2, 6, 9, 13, 17 and 28 days of dietary repletion.

### **Study-2: Effect on inclusion of zinc on absorption of iron and Zn**

The absorption of zinc in the preparation fortified with both iron and zinc was studied using radio-labelled  $^{65}\text{ZnCl}_2$ . The animals belonging to the test groups were fed on diets containing 202 and 110 and 440 and 404 with and without zinc respectively.

The animals were acclimatized to consume a small amount of the diet as a lump with the radioactive Zn for 3 days before metabolic experiment was initiated. A dose of 2 mci of  $^{65}\text{Zn}$  equivalent to 4.9 mg of Zn was spiked with the diet and ensured its complete intake in the first three hours. The rest of the diet was fed to the animals subsequently. The animals were housed individually in metabolic cages for collection of urine, stool and food left for 6 consecutive days.

Radioactivity due to  $^{65}\text{Zn}$  was measured using a gamma scintillation counter with approximately 6% efficiency. The cumulated amount of radioactivity excreted in the faeces over four days was subtracted from the dose to obtain apparent absorption. The radioactivity that was left out in the diet cups, if any, was counted in the washing after each radioactive bolus feeding to correct for the intake.

The results on Hb and body weights were evaluated by Analysis of Variance with repeated measures. The increments in Hb were correlated with time and the slopes and elevations were compared by Analysis of Covariance. The slope of the increment is considered to reflect the relative bioavailability of iron.

Results are summarized as follows :

Body weights and weight gains were not significantly different between any of the groups.

#### **A. Iron bioavailability**

1. Three types of responses : good 101, 110 and 330, intermediate 440 and 220 poor 303 bioavailability were obtained with nutritional beverage containing different iron sources.
2. The best response with an increment of 6.7 g of Hb/100 ml over the experimental duration and a rate of 0.25 g/100 ml mean increment in Hb/day was observed with 101.
3. Beverage containing 110 showed an increment of 6.4 g/100 ml and the rate of increase in haemoglobin was comparable to that of 101 which was used as a reference.
4. The response with one supply source of the same iron salt 220 was lower than that of the other 110, emphasising the importance of supply source and quality of iron salt used.
5. Iron from 330 also gave comparable bioavailability to 110.
6. Iron from 404 and 220 gave intermediate rates of increments in haemoglobin.
7. Beverage 303 as a source of iron gave the poorest mean increment in Hb (0.087 g/100 ml).

#### **Effect of zinc on iron bioavailability**

Addition of zinc at the level used in the study (7 mg/54 g beverage) did not alter the response in haemoglobin regeneration with the two iron salts tested with 202 and 440. Thus, iron bioavailability of the beverage supplement was not influenced by the inclusion of zinc.

#### **B. Zinc absorption**

1. In general, excretion of Zn in stool was maximal on the second day, accounting for 26-37.5% of the dose except for one animal which showed the peak on the first day (30%). Subsequently, the combined excretion was between 9.6-19.0% and less than 3% on days 3 & 4 and 5 & 6, respectively.
2. The proportion of absorbed zinc excreted in urine was negligible.
3. The overall absorption of zinc (considering 4 day faecal excretion) ranged between 31% to 46.4% with a mean of 36% in the animals receiving 202. On the other hand, in animals receiving only 110, the absorption ranged from 32% to 43.6% with a mean value of 37%.

4. In the same way, the absorption of Zn was  $34.2 \pm 4.2$  with 404 and  $32.1 \pm 5.0$  with 440 containing both iron and zinc.

## **Conclusion**

Beverage preparations with three different chemical source of iron 110, 202 and 330 had good iron bioavailability.

Different market sources of the iron salt as in 202 and 220 showed varying bioavailability indicating that quality of supply source is important.

Addition of zinc to two different chemical sources of iron did not alter the availability of iron.

Also zinc absorption was not affected in the beverages containing additional amounts of zinc as compared to only iron fortified ones.

Thus, for iron and calcium balance studies in children, beverage containing a particular market source of iron salt, 110, was chosen.

## **Part - B**

### **Iron and calcium balance studies from a special nutritional beverage code No.110 in children**

The nutritional beverage with different salts of iron and zinc were screened to obtain the combinations with optimal absorption for iron and zinc using experimental rats. The preparation with code No.110 was selected for evaluating the bioavailability of iron and calcium in children using 24 h chemical balance methods. A pilot study was carried out as a part of this study to test the comparability of a 4-day equilibrium to that of 7-day, for the possibility of shortening the duration of the study. Usually chemical balances are carried out over one week equilibration and 3-4 day test points.

#### ***Study design***

A metabolic experiment was performed in 8 children of 4-7 years age group at 2 points of time. On admission at the hospital, all the children were subjected to a thorough clinical examination by a paediatrician. All of them were dewormed with a single dose of albendazole (400 mg).

The first set of balances were performed in 4 children during a 10 day equilibrium on a hospital diet. The hospital diet consisted of breakfast, lunch and dinner. Bread and tea was given for breakfast while lunch and dinner consisted of rice, *dhal* and vegetable curry. In order to keep the calcium content of the diet close to the habitual intake, consumption of egg was omitted from the diet while milk intake was kept minimal. The total weight of the duplicate diet, on an average varied between 1250 to 1015 g. This was followed by additional intake of nutritional beverage for the next 10 days. During this period, the designated beverage was given at the recommended level of iron of 14 mg/day on the same

diet (from 54 g of the beverage). During experimental period, the beverage was given in the morning during 10 am to 10.30 am. In the case of one child who was not able to take it in one sitting, the drink was given in two portions with in half-an-hour. Twenty four-hour urine and stool and food left were collected for each child for a period of 6 days (4 to 10 days) during each metabolic study period. Duplicate diet collected during the balance period was processed for iron and calcium. The absorption and retention of both iron and calcium was determined.

Based on the results of the balance study in the first set of 4 children, a minimum equilibration period of 7 days was found to be essential. In the second set of 4 children, the balance study was carried out exactly in the same way as in the first set, except that 7 days of equilibration followed by 3 days of collection was employed.

About 10 g of the homogenized diet or stool samples were processed by dry digestion and ashing. Suitable aliquots were used for the estimation of iron by Wong's method. The calcium content was determined in an atomic absorption spectrophotometer.

Salient findings of the study are given below :

1. The children belonged to the low socio-economic group. Based on the  $wt/ht^2$  (% BMI) ( $<0.150$ ), 3 out of the 8 children studied exhibited general undernutrition.
2. All of them were anaemic and the mean haemoglobin was 95 g/L.
3. The coefficient of variation (CV) for iron balance for the first 3 days was generally much greater than that for the second 3 days period indicating insufficient stabilization in 4 days as compared to that in 7 days.
4. The mean (SE) intake of dietary iron of the eight children on basal diet was  $18 \pm 0.32$  mg/day. With the inclusion of special nutritional beverage, it was  $28.9 \pm 0.64$  mg/day in the second part. Thus, all the children received additionally 11 mg of iron per day during the experimental period.
5. Children on beverage period excreted higher amounts of iron compared to that on basal diet. The iron balance were  $1.15 \pm 2.61$  and  $7.2 \pm 2.71$  mg per day on basal and experimental periods, respectively. The differences in the intake, excretion and balance were significantly higher during experimental period compared to the basal.
6. These results suggest that additional amounts of iron (12 mg) given through the special nutritional beverage is bioavailable and provided a significantly higher iron retention. Intake of calcium was higher (by about 400 mg) on beverage compared to basal

### **Calcium balance**

period. Though faecal excretion of calcium increased on beverage intake, urinary excretion was similar during both the periods of observation. Due to low intake of calcium as seen in basal diet, the balance in all children except in one child was negative. On taking the beverage, all except one child, exhibited a fractional negative balance. On an average, the children showed an absorption of 32% and retention of 23% in the balances carried out in the first set of children. The dietary calcium varied between 72-78 and 157-179 mg/L in the first and second set of children. The differences were mainly due to the differences in the nutrient components of ingredients used in the beverage. One child (No.8) with severe round worm load, exhibited very adverse calcium balance.

## **Conclusion**

The extra amounts of iron (12 mg) and calcium (400 mg) per day provided through the special nutritional beverage is bioavailable when included on a basal rice-based diet. The absorption increased from 6.4% to 24.8% with iron, from a negative value to 31.7% with calcium. The absorption of iron by chemical balance is usually known to be over-estimated. Therefore, the present values are in the acceptable range set by NIN (3.0% iron and 30% calcium absorption) for the beverage before-hand and the beverage could be further tested in the clinical trial.

## **II. ENERGY METABOLISM**

### **1. Physiological energy transfer mechanisms of athletes**

The energy needs of various sport activities depend on several factors such as age, sex, body size and composition, climate, level of fitness, apart from type of sport and phase of training, intensity and duration. In order to maintain desirable body weight, composition and peak performance levels, adequate nutritional support needs to be provided. Sound nutrition forms the foundation of sports performance and any imbalance between energy expenditure and intake impairs the performance. Therefore, this study was carried out to identify the manifestations of incremental training load on physique, physiological energy mechanisms and energy requirements in three groups of athletes from Transition Phase (TP) to Competition Phase (CP) of training.

To achieve this objective, national level male sprinters (n=12), middle-distance runners (n=14) and long-distance runners (n=16), aged between 18 and 22 years were recruited from Sports Authority of Andhra Pradesh (SAAP), Hyderabad. The anthropometric measurements were recorded using standard procedures. The Lean Body Mass (LBM) and Fat Mass were derived from skinfold thickness measurements using age and sex matched equations of Durnin & Womersley (1974). The Basal Metabolic Rate (BMR), energy cost of various activities and at different loads of Graded Exercise Tests (GXT) were measured by open circuit indirect calorimetry. Bruce protocol of GXT was adopted to evaluate physiological efficiency and maximal work performance. Quantification of training and energy expenditure was done by 24-hours Time Allocation Pattern (TAP) combined with oxygen consumption levels and heart rates. The same protocol was given in

the three phases of training.

The results of the study are as follows:

It was observed that the training duration was increased by two-folds, (88 Vs 171 min/day in sprinters, and about 100-204 min/day in distance runners) and intensity by 1.2 folds in sprinters (826 Vs 995 kpm/min), 1.5 folds in middle (672 Vs 1096 kpm/min) and long-distance (727 Vs 1138 kpm/min) runners from TP to CP. The increment in training load might have manifested the following changes.

a) The anthropometric profile revealed a significant increase in height (0.5-0.9 cm) and weight (1.5-2.5 Kg) from TP to CP in all the three groups of athletes. The sprinters were found to be heavier with comparable stature than the middle and long-distance runners.

b) The LBM was significantly increased about 3% in sprinters (55.2 Vs 56.8 Kg), 4.6% in middle (45.4 Vs 47.5 Kg) and 3.4% (46.7 Vs 48.3 Kg) in long-distance runners with progression of training without any remarkable change in fat content.

c) Based on BMI, the sprinters were found to be normally nourished while the middle and long-distance runners were found to be moderately undernourished in any given phase of training.

d) The 24-hour BMR was enhanced by about 9% in sprinters (1452 Vs 1584 Kcal), 16% in middle (1216 Vs 1404 Kcal) and 11% (1282 Vs 1423 Kcal) in long-distance runners from TP to CP.

e) The basal heart rate of three groups of athletes was significantly decreased with progression of training reflecting the cardiac adaptation to training.

f) The maximal work performance increased by 15% in sprinters (2013 Vs 2319 kpm/min), 37% in middle (1571 Vs 2147 kpm/min) and 32% (1676 Vs 2211 kpm/min) in long-distance runners with progression of training.

g) The aerobic capacity increased by about 10% (47.6 Vs 52.1 ml/kg/min) in sprinters and about 13% in both middle (52.1 Vs 58.8 ml/kg/min) and long-distance (51.2 Vs 57.9 ml/kg/min) runners with training.

h) The total daily energy expenditure increased by about 30% in sprinters (2630 Vs 3410 Kcal), about 48% in middle (2360 Vs 3483 Kcal) and 41% in long-distance (2554 Vs 3597 Kcal) runners with increase in training load, even though no significant change was observed in non-occupational (other than training) energy expenditure of three groups of athletes between the phases of training.

Based on these observations, the following can be concluded:

1. There was a larger inter-individual variation in training intensity received by the athletes in a given training schedule. In view of this, it would be better to construct individualised progressive program of training considering the athletes body weight, LBM and initial fitness levels to provide optimal/crest load to all athletes to achieve optimal work performance.
2. The energy requirements were significantly influenced by variation in training load. Therefore, periodical evaluation of energy needs is necessary to recommend energy allowances to monitor desirable weight, composition and peak performance levels.
3. Even though all the physical and physiological parameters improved with incremental training load, these athletes are par below the international standards. Therefore, it is essential to select the athletes based on scientific evaluation with better physique, endowed with innate physiological efficiency and nurture them with proper scientific training and adequate nutritional inputs to achieve high level of athletic performance in years to come.

Similar studies are planned in different athletic events like hockey, football, volleyball, basketball etc., so as to identify the energy requirements of athletes.

## **2. Relationship of women's work and energy expenditure to anthropometry and body composition**

Body mass index (BMI) is highly correlated with body weight and is known to be independent of stature across populations worldwide. Therefore, BMI is normally used for the assessment of nutritional status of adults for epidemiological studies.

Nutritional status is known to affect work output and total energy expenditures during increased demands of work. During periods of low energy intakes, several scientists have observed changes in work patterns in terms of time and energy spent on different productive and leisure time activities. On the other hand, studies from this Institute have shown that conditions of increased energy demands, such as increased housework, child care, illness, etc., resulted in women's attempt to negotiate between heavy tasks by very often opting out of paid work (Ann. Rep. 1993-96). The relationship between low BMI and low productivity has been clearly demonstrated in an urban industrial situation. Studies from Ethiopia and India have highlighted very low levels of physical activity in adult men and women with low BMI (Ferro Luzzi *et al.*, EJCN. 1992,46,173-186). Studies on rural Guatemalan populations showed that malnourished men carried out specific agricultural tasks allocated to them, but took much longer time (Torun *et al.*, Proc. XIV Int. Cong. Nut, Seoul, 306-309, 1989). Data collected for the National Food Consumption and House-

hold Budget Survey in Rwandan Women revealed that physical activity levels (PAL) in women of low BMIs are significantly lower (Francois P, FAO Report, 1990). It is obvious that both undernutrition in terms of low BMI and increased demands of work output force population to negotiate energy expenditure. It was therefore considered necessary to study the impact of under nutrition on the activity profiles and total energy expenditures of poor undernourished women belonging to the unorganized sector.

Thirty women with  $\leq 16.5$  BMI, and forty-three women with  $\geq 18.5$  BMI from the low socio-economic group were randomly selected for the study. Background information including family size, age, education etc., was collected. Body composition was measured by skinfold thickness at 4 sites.

Time disposition studies of the women were undertaken to record the activity profiles on a typical day. Basal metabolic rate and energy cost of household, childcare, occupational and other activities were measured under standard conditions. Using time data and energy cost of activities, the energy expenditures were calculated and the total energy expenditures per day on each of the activities were arrived at.

The results can be summarised as follows:

1. The mean (SD) age of the overall study group was 26 (5.0) year and it was not different in the 2 BMI groups. The mean (SD) weights of the two BMI groups (low BMI 16.5 vs normal BMI 18.5) were 35.0 (3.8) kg and 48.8 (7.0) kg, the mean fat per cent being 17.6 (3.37) and 26.9 (5.69) and the mean BMI being 15.5 (0.72) kg/m<sup>2</sup> and 21.4 (2.26) kg/m<sup>2</sup> which were significantly different ( $P < 0.05$ ). However, the mean heights were not different between the 2 groups. The social indicators of the two groups were as follows, mean family size 4.8 and 5.1, the mean number of children <5 years of age was 1.55 (0.60) and 1.67 (0.65) and they were not significantly different. The mean basal metabolic rate of the 16.5 BMI group was 0.66 (0.08) Kcals/min, which was significantly lower than that of >18.5 BMI group 0.77 (0.06) Kcal/min ( $P < 0.05$ ). This may be explained by the increased body weight of the 18.5 BMI groups. The mean energy expenditure of the two BMI groups in terms of BMR factor during the standard activities such as sitting and standing were 1.14 (0.09), 1.30 (0.08) in the <16.5 and it was 1.15 (0.08) and 1.32 (0.09) in the 18.5 BMI group, which were not different.

2. Energy expenditure during the household activities ranged from 1.7 (0.27) BMR to 3.86 (0.73) BMR in the 16.5 BMI group and 2.01 (0.30) to 4.34 (0.82) BMR in the 18.5 BMI group of women. Childcare activities consumed energy ranging from 1.5 (0.42) to 2.39 (0.46) BMR in the low BMI group, and 1.5 (0.42) to 2.7 (0.49) BMR in the normal BMI with bathing the child requiring the highest amount of energy and breast-feeding the least. Among all the activities measured, swabbing consumed the highest amount of energy and sitting the least. Beedi-making and tailoring by <16.5 BMI groups consumed 1.5 (0.15) BMR and 1.7 (0.01) BMR and 1.5 (0.115) BMR and 2.08 (0.30) BMR in the >18.5

BMI group.

3. There was no significant difference between the 2 groups in the energy cost of activities in terms of BMR factors excepting for sweeping, washing clothes and cooking. As the BMI increased, the amount of energy spent on these activities increased significantly in terms of BMR factors. Even the energy equivalents of walking in the 2 BMI groups was 2.45 (0.34) and 2.96 (0.36) in terms of BMR factors, which were significantly different ( $P < 0.05$ ). It is possible that women with higher BMI have to strain themselves during heavy activities resulting increased energy expenditures.

The total BMR were 969 Kcal/d (<16.5 BMI) and 1109 Kcal/d (>18.5 BMI). The mean total energy expenditures during the day of the <16.5 BMI group and >18.5 BMI group women were 1596 Kcal/d and 1868 Kcal/d which were significantly different ( $P < 0.05$ ). The mean total energy expenditure per day in terms of BMR factor were 1.68 (<16.5 BMI) and 1.73 (>18.5 BMI) which were not significantly different. The total energy expenditure in terms of unit body weight were 46 Kcal/day (<16.5 BMI) and 38 Kcal/d (>18.5 BMI), suggesting that work may be the cause of lower body weights in the <16.5 BMI group, and women spending less energy during the day had higher weights.

### **III. NUTRITION AND INFECTION**

#### **1. Safety of administering vitamin A along with polio vaccine on national immunization days in the State of Orissa**

In view of high prevalence of vitamin A deficiency and low coverage of vitamin A administration to 1-5 year old children through the National Vitamin A Supplementation Programme in Orissa, the State Government initiated a strategy of administering vitamin A to children between 12-42 months on a campaign basis. The first round was given during the month of March 1999. The second round of administration was combined with National Immunization Day (NID) of IPPI Programme held in October. WHO suggested a rapid safety survey and also assess the feasibility of such combined schedule.

Five districts in Orissa with 2 blocks in each district and 2 immunization booths in each block were randomly selected. Ten teams consisting of a Medical or Technical Personnel from NIN and RMRC, Bhubaneswar, with 10 local volunteers administered a precoded and field tested questionnaire to the mothers of children below the age of 5 years on 25th and 26th October following the NID on 24th October 1999. The sample size proposed for this was 160 children per district.

The results are as follows:

A total of 879 children were covered for the survey. Polio coverage was 97%, while 72.4% of the children received vitamin A on NID. Among these, 22% were infants be-

tween 9-12 months. Not a single infant below 6 months received vitamin A on NID in the survey region, while 92% of the target group of infants (12-42 months) received vitamin A. A total of 26 children complained of ill health like diarrhoea, vomiting, nausea, fever and excessive crying. However, these symptoms were not different between the children who received vitamin A or not along with oral polio vaccine (OPV) on 24th October.

This approach of administering vitamin A on NIDs appears feasible and safe and improves the coverage and may be beneficial in regions where vitamin A deficiency is still prevalent and national programme has low coverage.

## **2. Serum transferrin receptor (STR) in children suffering from infections**

An ELISA technique has been developed to estimate circulating transferrin receptor (STR) in children. Using the technique, an attempt was made to (a) arrive at the normal values for serum transferrin receptor (b) develop cut-off values to define iron status of healthy children, and (c) study the effect of infection on STR.

Forty-four healthy children with percentage weight for age more than 90 of NCHS standards with Hb levels ranging from <9.0 to >12.0 g/dL from a Social Welfare Hostel were recruited. Haemoglobin, serum ferritin and STR were determined according to the methods described in our earlier reports.

The results are as follows:

1. The receptor levels (mg/L) were  $14.8 \pm 3.65$ ,  $9.1 \pm 1.16$ ,  $6.3 \pm 0.52$  and  $5.5 \pm 0.89$  in children with Hb (g/dl) 9-10, 10-11, 11-12 and  $>12.0$  respectively while their mean ferritin levels ranged from  $19.6 \pm 3.81$  to  $27.1 \pm 3.11$  mg/L.
2. There was a significant negative correlation between haemoglobin and STR ( $-0.81$ ;  $P < 0.01$ ).
3. STR, however, failed to show any correlation with serum ferritin value. As the mean in healthy children  $6.1 \pm 0.44$  mg/L (Brit. J. Nut. 1997; 77:390) it was used as normal receptor value in preschool children to define iron status. As this value is similar to the reported mean STR value ( $6.1 \pm 0.48$  mg/L) was obtained.

### **Determination of iron status of children using STR**

In an earlier study, iron status indicators viz., haemoglobin, ferritin, STR in 46 apparently normal preschool children who were siblings of children hospitalized for anaemia were determined.

Mean values of  $6.1$  mg/L for STR was obtained by pooling data from these two studies. Using this value, as determined for STR, and  $20$  mg/L for ferritin as cut off levels, the iron status of children was defined.

Distribution of children having STR less than  $6.1$  mg/L in various Hb groups is shown in **Figure 9**.

The results can be summarised as follows:

The results suggest that iron stores are absent by the onset of even mild anaemia. More than 50% of children with haemoglobin more than  $11$  g/dl also have elevated levels of the receptor, thus, identifying the wide prevalence of iron deficiency status in children. Ferritin, however, failed to identify even severe anaemia.

### **Effect of infection on STR**

Changes in STR and ferritin were measured in 119 preschool children attending hospital for upper respiratory infections/pneumonia/chicken pox. Among them, 74 and 29 children could be reinvestigated on 15th and 30th day of the infection, respectively. Normal children recruited for assessment of iron status served as control group.

The results were as follows:

1. Mean haemoglobin (g/dl) was  $9.8 \pm 0.14$  during infection which was significantly lower ( $P < 0.001$ ) than that observed in children without infection ( $10.4 \pm 0.20$ ).
2. There was a significant drop in Hb to  $9.3 \pm 0.22$  g/dl by day 15 which tended to

increase by 30th day ( $9.9\pm 0.33$ ) in children with infection.

3. Mean STR levels were  $14.3\pm 1.38$  mg/L in children without infection and they ranged from  $13.3\pm 1.17$  to  $17.1\pm 1.02$  mg/L in the infected children on different days of investigation.
4. STR had a significant negative correlation with Hb even during infection ( $-0.71$  on day 1,  $-0.61$  on day 15, and  $-0.77$  by 30th day).
5. Mean ferritin was significantly elevated during acute infection which tended to decline only by 30th day.
6. Frequency of children having anaemia increased with a corresponding rise in the proportion having elevated STR values during infection which was almost 100% by 30th day after infection (**Fig.10**).

The following conclusions may be drawn from these studies:

Infection does not appear to significantly alter mean STR in populations having widespread iron deficiency as the mean STR is already high. Depletion of iron stores in children having normal receptor levels in the anaemic group and inability to come back to normal levels, even 30 days after infection, suggest that anaemia in infection is due to iron deficiency. This could be due to lack of iron in the anaemics or non-mobilization of iron from the storage pool in non-anaemics. The fairly prolonged period of this phenomenon raises the important issue of control of infections in improving the iron status of children exposed to frequent infections.

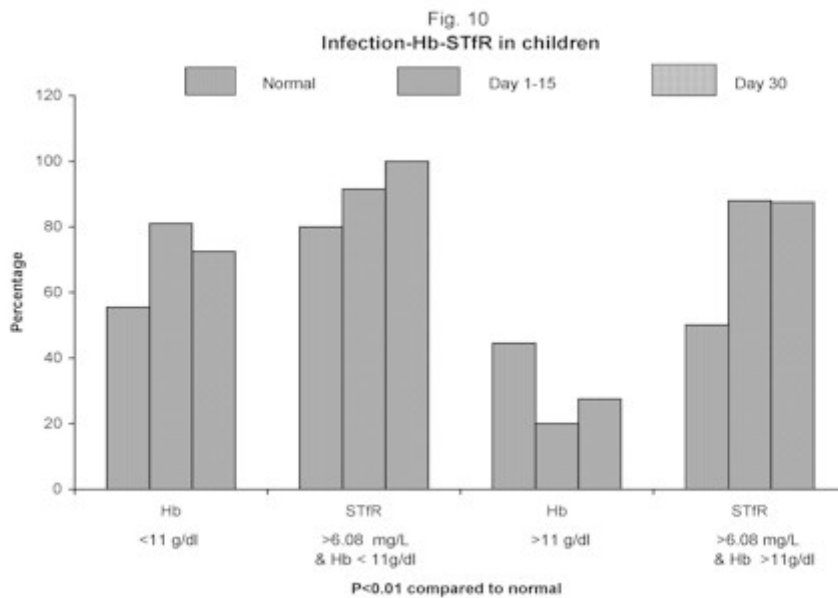
### **3. Acute bacterial meningitis and TNF- $\alpha$ in malnourished children**

Pyogenic meningitis is an important contributor to <5 year childhood mortality and morbidity due to neurological sequelae. Tumor Necrosis Factor (TNF- $\alpha$ ) is implicated in determining the severity of the disease. TNF- $\alpha$  is a cytokine produced in response to infections and inflammations and modulates host tissue response. Sustained over production of this cytokine contributes to local tissue damage, the severity of which determines the outcome of the disease.

In the present study, the role of nutritional status and TNF- $\alpha$  secretion in children suffering from acute bacterial meningitis and the outcome of the disease were determined.

One hundred and forty children diagnosed to be suffering from acute bacterial meningitis based on the standard criteria of CSF changes (elevated protein, fall in glucose levels and predominance of neutrophils) were recruited for the study. Weight for age was

The results may be summarised as follows:



1. Of the children recruited, 85 were males and 55 females. Eighty three of them were under 1 year and 57 between 1-5 years. Severe malnutrition was significantly higher in children above 1 year of age.
2. Bacterial isolation was positive from 76% of CSF samples, the organisms isolated being *S.pneumoniae* (20), *N.meningitidis* (6), *H.influenzae* (7), *E.coli* (10), *Staphylococci* (6) and *Enterococcus* (1). Bacterial isolation rate was higher in older children (92%) compared to infants (67.5%).
3. TNF- $\alpha$  was detectable in 67 per cent of all the cases. Children who were culture positive for *S.pneumoniae*, *N.meningitidis* and *H.influenzae* had higher levels of TNF- $\alpha$  compared to the others though the differences were not significant.
4. Mean TNF- $\alpha$  levels were similar in all nutritional grades. However, percentage of children producing TNF- $\alpha$  was higher in the severe PEM group.
5. There were 22 deaths (15.7%) and 40 (28.5%) children recovered with sequelae. Deaths and sequelae were similar between males and females and in infants and older children.
6. Children with normal nutritional status registered maximum recovery (84.6%) compared to all the children and CSF culture for bacterial isolation was carried out on a random sub-sample of 65 children using standard microbiological procedures. TNF- $\alpha$  was measured in all CSF samples by developing sandwich ELISA. Recombinant TNF- $\alpha$  was used as the standard. Recovery and sequelae were assessed by clinical examination. Data was analysed using SPSS PC software.

pared to 31.6% in severely malnourished children while the percentage recovery was 62.7 and 50.9 respectively in the mild and moderately nourished groups.

7. Highest percentage of deaths occurred (50%) with *S.pneumoniae* infection followed by 18.7% with *N.meningitidis*, 12.5% each with *H.influenzae* and *E.coli*. Infection with *S.pneumoniae* and *H.influenzae* was more common in severely malnourished children.

The results can be concluded as follows:

1. Nutritional status and causative organism appear to determine the outcome of pyogenic meningitis in children.
2. TNF- $\alpha$  concentrations do not appear to have any significant influence on the outcome nor its levels are influenced by nutritional status.

#### **4. *Escherichia coli* in children with acute diarrhoea - A pilot study**

*Escherichia coli* (*E.Coli*) 0157:H7 is a recently identified foodborne pathogen responsible for outbreaks of haemorrhagic diarrhoea with high mortality rate and is often associated with development of haemolytic uraemic syndrome. A preliminary study was conducted to determine whether this pathogen is responsible for acute diarrhoea among children in our community during the diarrhoea season (June, July, August) of 1999.

One hundred children (<5 years of age) having acute diarrhoea, were investigated; faecal samples were cultured and screened for enteric bacterial pathogens including 0157:H7 strain. This strain was isolated by culturing the faecal samples on Macconkey Agar. Different strains of *E.coli* were identified from gram negative catalase positive cultures, further processed by entero rapid test followed by serotyping. The identified strain of *E.coli* 0157:H7 was confirmed further by referring to Central Research Institute, Kasauli.

The preliminary work may be summarised as follows:

1. Mean age of diarrhoea in affected children was 10.2 $\pm$ 2.01 months with 70% of cases being less than one year.
2. Pathogenic *E.coli* were isolated from 27 cases. Of these, enteropathogenic *E.coli* (EPEC) was the commonest (16), followed by enterotoxigenic *E.coli* (ETEC) (6) and enteroinvasive *E.coli* (EIEC) (3).
3. From 2 children EHEC (0157:H7) was isolated. These results suggest that acute diarrhoea due to EHEC 0157:H7 strain is existent in this part of the country and is one of the causes for diarrhoea in infants.

There is a need to conduct indepth studies to determine the prevalence, nature of infection and outcome in relation to nutritional status.

#### IV. MICRONUTRIENTS AND TRACE ELEMENTS

##### A. VITAMINS

##### 1. Severe xerophthalmia in children attending Niloufer Hospital, Hyderabad, between 1980-1999

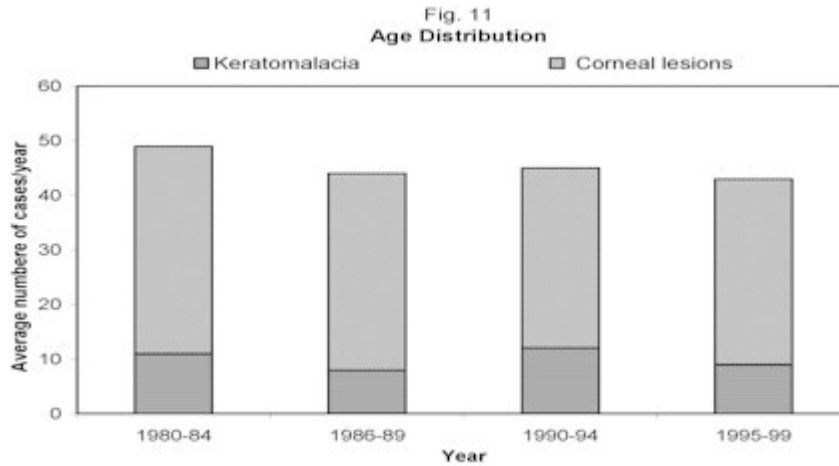
To observe the changing trends of severe vitamin A deficiency, data collected on children with corneal lesions from Niloufer Hospital from 1980 to 1999 were analysed.

At Nutrition Unit of Niloufer Hospital, rehabilitation services to children with nutritional deficiencies are routinely extended by NIN. Cases of corneal xerosis/ulcer/ keratomalacia, attending out-patient department or admitted to Paediatric wards, are identified, both in-patient and out-patient departments of all Paediatric Units every day. They were appropriately treated and details entered into well maintained records. In effect, no child with corneal lesions was missed.

A total of 540 children with corneal lesions attended the hospital during this period.

The results are as follows:

1. On an average, 22-28 cases of corneal lesions were observed per year from 1980-1999. The lowest average was observed (22) between 1990-1994.
2. Average number of children with keratomalacia resulting in total blindness ranged from 9-12/year (**Fig.11**). Distribution of the cases according to age showed that 66% of the children were between 1-3 years, while 25% were between 3-5 years. A small percentage of 3 and 6 were observed in the age groups above 5 years and below 1 year respectively.
3. Distribution of cases under one year of age again demonstrated that 86% were infants above 6 months of age. Fourteen percent were between 3-6 months while no case of corneal lesions under 3 months of age was recorded.



parts of the country. In view of this, there is an urgent need to introduce innovative strategies for improving vitamin A intakes in children.

**Conclusions**

1. Severe clinical vitamin A deficiency is still a problem in the slums and nearby rural areas of Hyderabad which are catered by Niloufer Hospital.

2. Severe xerophthalmia does not seem to be a problem in young infants (<3 months)

**2. Relationship between plasma homocysteine level and folate and**

**vitamin B<sub>6</sub> status**

This analysis by no means attempts to project the incidence of vitamin A deficiency in the community. But, it does suggest that severe clinical vitamin A deficiency is still alive in poor communities and there is a need to search for pockets of such cases in other  
Several studies carried out in western population have shown that elevated plasma levels of homocysteine are associated with an increased risk of early onset of cardiovascular disease. Folate status is the major determinant of plasma homocysteine level and vitamin B<sub>6</sub> and B<sub>12</sub> dependent enzymes are required for homocysteine metabolism. Since

biochemical deficiencies of folate and vitamin B<sub>6</sub> are common in our population, a study was undertaken to examine the correlation between plasma total homocysteine level and folate and vitamin B<sub>6</sub> status in apparently normal male subjects.

Forty apparently normal male staff members of the institute in the age range of 40-58 years belonging to the middle and high income groups participated in this study. Fasting blood sample was collected for the measurement of the following parameters:

- (1) Plasma total homocysteine by HPLC method
- (2) Erythrocyte and plasma folate concentrations by microbiological assay
- (3) Erythrocyte aspartate aminotransferase activation coefficient (EGOT-AC) as an index of vitamin B<sub>6</sub> status.

The results are as follows:

1. Plasma total homocysteine concentration (Mean±SD 19.83±7.9  $\mu\text{mol L}^{-1}$ ) was negatively correlated ( $P < 0.001$ ) with erythrocyte folate (164.78±73.5 ng/ml) and plasma folate levels (5.26±2.8 ng/ml).
2. There was no significant relationship between plasma total homocysteine level and EGOT-AC (Mean±SD 1.73±38).
3. About 42% of the subjects had biochemical deficiency of folic acid as judged by red blood cell folate level (<140 ng/ml). The mean erythrocyte folate (ng/ml) and plasma homocysteine ( $\mu\text{mol/L}$ ) levels in these subjects were 100.6±28.85 and 26.81±6.7 respectively. Whereas in subjects with normal erythrocyte folate level (212.2±58.70), the mean plasma homocysteine concentration was 14.66±3.474  $\mu\text{mol/L}$ .

The results of the study suggest that subclinical folate deficiency is a significant factor contributing for the higher plasma homocysteine level observed in our population compared to the western population.

## **B. MINERALS**

### **1. Validation of serum transferrin receptor (STR) as a specific indicator of iron status**

Circulating serum transferrin receptor (STR) concentration is reported to be a sensitive and specific indicator of iron status. A two step sandwich ELISA technique was developed last year to assess the levels of STR (Ann.Rep. 1998-99). This indicator needs validation in health and disease. In the present study, the validation of STR with other iron status and haematological parameters in normal healthy adolescent girls is reported.

## Methodology

Thirty four healthy adolescent girls from an orphanage school in Hyderabad were recruited for the study. Oral consent was obtained before obtaining their basal blood sample. They were given a tablet containing 60 mg iron and 500 mg of folic acid daily for 100 days. Five ml fasting blood samples were obtained from all of them during supplementation (50 d, 100 d) and after its withdrawal (90 d and 180 d).

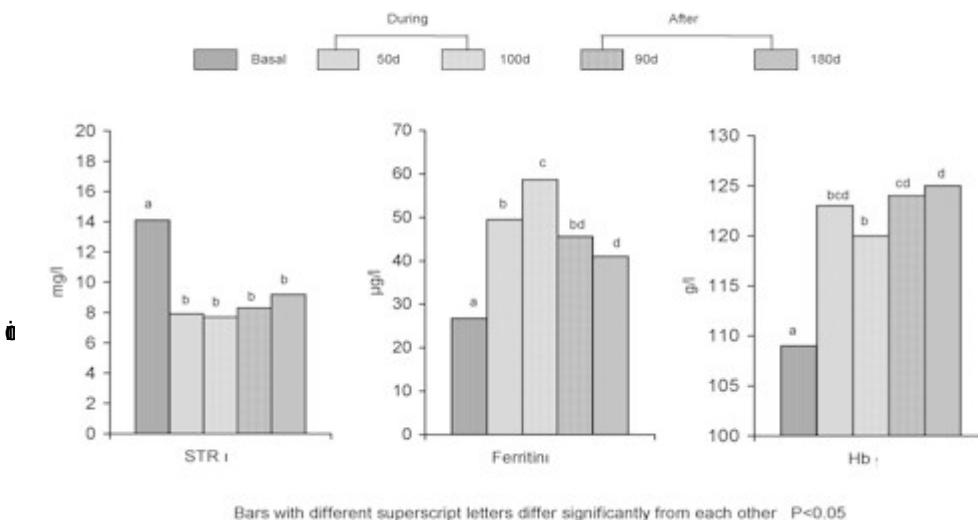
Blood haemoglobin (Serono Baker Counter System 9000 Rx), serum ferritin and transferrin receptor (indigenous ELISA techniques) concentrations were estimated. The sequential changes in the above parameters in response to iron treatment was compared by repeated measures ANOVA. The sensitivity and specificity of the STR as a measure of iron-status was calculated. Also, the response of parameters to treatment in normal (Hb >120 g/L and ferritin >15 mg/L), iron deficient (Hb <120 g/L and ferritin <15 mg/L) and anaemic (Hb <120 g/L and ferritin >15 mg/L) subjects were compared.

The salient findings are summarized as follows :

1. Based on the basal iron status, the STR levels (Mean  $\pm$  SD) were  $9.5 \pm 1.07$  mg/L,  $19.9 \pm 1.98$  mg/L and  $17.1 \pm 2.45$  mg/L respectively in normal (Hb,  $124 \pm 1.6$  g/L and ferritin  $25.4 \pm 1.92$  mg/L), iron deficient (Hb,  $96 \pm 5.4$  g/L and ferritin  $11.6 \pm 0.75$  mg/L) and anaemic (Hb,  $95 \pm 1.9$  g/L and ferritin  $28.2 \pm 4.6$  mg/L) subjects.

2. The sequential changes in the indicators of iron status are presented in **Figure 12**. Haemoglobin concentration increased significantly ( $P < 0.05$ ) from (Mean  $\pm$  SD)  $109 \pm 1.3$  g/L to  $123 \pm 0.58$  g/L in 50 days with no further increase at the end of 100 days of tablet intake ( $120 \pm 1.0$  g/L). The Hb concentration maintained at this level upto 180 d after the withdrawal of iron supplement, possibly due to the mobilization of storage

Fig 12  
Sequential Changes in Serum Ferritin and STR During Iron Supplementation and After its Withdrawal in Adolescent Girls



3. Serum ferritin concentration was significantly higher at 50 and 100 d compared to basal value (26.8±19.2 to 49.5±20.4 to 58.7±26.2 mg/L).

Serum ferritin concentration lowered significantly from the peak value of 58.7±26.3 mg/L to 45.6±24.6 and 41±23.7 mg/L respectively at 90 and 180 d after the withdrawal of iron supplement. This indicates mobilization and adequacy of iron stores by 180 d after withdrawal of iron supplement.

4. Serum transferrin receptor concentration decreased significantly from 14.1±8.5 mg/L by 50 d to 7.9±4.1 mg/L and stabilized. STR levels did not change neither with increasing intake of tablets (100 d) nor after its withdrawal (90 or 180 d).

5. STR failed to show any correlation with serum ferritin.

6. There was a significant negative correlation between haemoglobin and STR concentration ( $r = -0.816$ ).

7. A cut-off value of 8.2 mg/L is obtained for a normal haemoglobin value of >120 g/L (WHO) for adolescent girls by fitting into a regression model. This normal mean value is higher than that reported in literature (5.6 mg/L) for US normal female. (J Lab Clin Med, 1989, 114: 368).

8. The sensitivity of STR as a measure in identifying iron deficient individuals is 0.87 with a specificity of 0.63. The specificity increased to 0.81 with normalisation of iron status. The sensitivity and specificity remained same (0.71) when tested with 180 days value

(after withdrawal of supplementation).

Thus, it is concluded that STR detects latest stage of iron deficiency. Similar studies in adolescent boys are in progress to derive their normal STR levels.

## **2. Epidemiological survey of endemic fluorosis in Mandla district of Madhya Pradesh with reference to intake of Ca<sup>++</sup>, vitamin C and fluoride**

Following an unexpected outbreak of dental and skeletal fluorosis along with genu valgum deformities in some villages of Mandla District of Madhya Pradesh, an extensive survey for endemic fluorosis was initiated by RMRC, Jabalpur. Food and water samples were collected and analysis for fluoride and trace elements (Cu, Ca, Zn, Mg) was undertaken. The values for fluoride, Ca<sup>++</sup>, Mg<sup>++</sup>, Cu<sup>++</sup> and Zn<sup>++</sup> in drinking water have been reported earlier. The fluoride and mineral content of food materials has been determined.

The results are as follows:

1. There is no definite pattern or change seen among the trace element contents of different food stuffs from the study and control areas.
2. None of the locally grown food stuffs were found to contain high fluoride. The fluoride content of different food materials ranged between 0.1 ppm to 0.87 ppm.
3. Water fluoride is mainly responsible for the fluorosis in the affected villages.
4. Chakoda (*Cassia tora*), chech and mustard leaves consumed by the local people were rich in calcium. Hence regular consumption of these leaves by people throughout the year has been recommended.

## **3. Effect of different cereals on fluoride deposition in bone**

Fluoride retention in the body and progression of fluorosis are reported to be influenced by the type of cereal used as staple diet. Hence, the present experiment on rats was initiated to find out the influence of jowar, wheat and rice as staple cereals on fluoride retention and deposition in bones.

The analysis of all the femur samples for fluoride, Ca<sup>++</sup> and Mg<sup>++</sup> has been completed. Fluoride contents of femur (mg/femur) were (Mean±SE) 0.595±0.0272, 0.560±0.0278, 0.571±0.0403 and 0.585±0.0426 in rats fed starch, wheat, rice and jowar based diets respectively. The differences in fluoride content were not statistically different. Calcium contents of femurs (mg/g bone ash) were (Mean±SE) 340±21, 349±26, 328±26 and 303±19.7, while magnesium contents of femurs (mg/g bone ash) were

(Mean±SE) 5.55±0.205, 6.54±0.214, 5.83±0.205 and 6.93±0.205 in starch, wheat, rice and jowar fed animals respectively. Calcium and magnesium contents were not statistically different among groups.

Fluoride content of mandibles (mg F/mandible) was 0.597±0.0555 for wheat group, 0.668±0.0555 for rice group and 0.772±0.06964 for jowar group. Fluoride content of wheat group is significantly lower than that of jowar and rice groups. Calcium and magnesium values did not differ significantly.

These results support the earlier observations that fluoride retention on jowar diet is higher than that on rice diet.

#### **4. Mineral and trace element composition of foods of animal origin**

Human beings require sufficient amounts of proximate principles, vitamins and minerals in their daily diet. Although many elements are known to be present in tissues, their precise role in human nutrition is not known clearly with respect to many of them. In planning dietaries for communities, it is therefore necessary to aim at an adequate well-balanced diet. Also for conducting dietary surveys to estimate nutrient intake, it is essential to have authentic scientific data about nutrients in commonly consumed foods.

Earlier, NIN has screened most of the plant foods of Indian origin for their mineral content and their profile and incorporated in the book "Nutritive Value of Indian Foods".

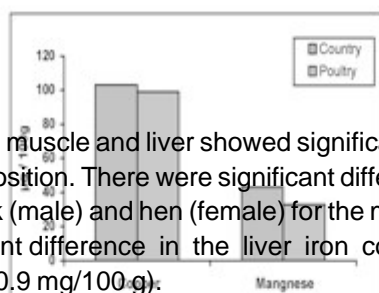
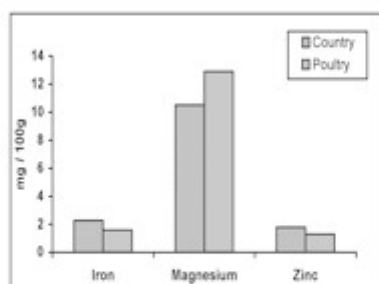
There is no authentic data for minerals and trace elements in the foods of animal origin based on modern techniques such as atomic absorption spectrophotometer (AAS). Hence, to generate the data, commonly consumed animal foods were screened for mineral and trace element content.

Foods of animal origin like eggs (hen, duck), milk (buffalo, cow, goat, ass, market milk), milk products (butter, cheese, cream, khoa, ghee), liver and muscle from goat, lamb, pork, pig (country), hen (country and poultry), cock (country), chinese bird, duck, pigeon were analysed for their mineral content and individual elements like calcium, phosphorous, iron magnesium, copper, zinc and manganese. For each foodstuff, three samples were collected and analysed.

The study revealed that:

1. Eggs of hen (country, poultry) and duck are good sources for minerals (Ca, P, Fe, Mg) and trace elements (Cu, Zn, Mn). Between country and poultry eggs, it appears that country egg contains higher quantities of these mineral and trace elements than poultry

Fig. 13  
Mineral Profile of Country & Poultry Eggs



2. Chicken muscle and liver showed significant differences in the mineral and trace element composition. There were significant differences between the muscle and liver of country cock (male) and hen (female) for the mineral and trace element profile. There was a significant difference in the liver iron content of country hen (3.6 mg/100 g) and poultry (10.9 mg/100 g). (Fig.13).

3. The other bird varieties studied like duck, pigeon, chinese bird for their liver and muscle also showed significant differences in the mineral composition. Particularly, duck liver appears to be superior than other bird livers in the quantities of minerals and trace elements with the exception of pigeon liver which contained highest amount of iron (28.1 mg/100 g).

4. Animal foods like goat, lamb, pork, pig (country) were also screened for their mineral composition in liver as well as muscle. Mineral composition of goat and lamb liver revealed that they are very good sources with similar quantities, except in the magnesium content (goat liver 91.7%, lamb liver 17.7 mg/100 g). There were significant differences between country pig and pork for all the mineral and trace elements studied. Country pig, both liver and muscle, contained higher quantities Ca, P, Fe, Mg and other micro elements than pork. Country pig liver, particularly, is the richest source for Zn (4 mg/100 g).

5. Milk of buffalo, cow, goat, ass and market samples have shown significant differences in their mineral content. Buffalo's milk is the richest source for most minerals and trace elements studied. Between goat milk and ass milk, goat milk had higher quantities of Ca, P, Fe, Mg where as ass milk contained higher quantities of trace elements like Cu, Zn and Mn.

6. Milk products like curd, butter, cream, cheese, *paneer* and *khoa* samples showed significantly higher content of mineral and trace elements than milk samples due to lower content of water. Among the milk products, *khoa* was found to be the richest source for calcium (744 mg/100 g), iron (2.3 mg/100 g) and magnesium (54.2 mg/100 g). Ghee did not contain any minerals.

## V. DIET AND NON-COMMUNICABLE DISEASES

### 1. Biochemical and metabolic studies with sesame lignans

The non-glyceride fraction of edible oils is a good source of natural antioxidants. While tocopherols are present in all oils, the lignans (sesamol - S1, sesamin - S2 and sesamol S3) present in sesame oil impart greater stability to the oil. Current evidences indicate that high intake of antioxidants can prevent diet-related chronic diseases.

Earlier, lignan profile and tocopherol levels in sesame seeds, isolation and crystallization of S2 and S3 and the effects of varying concentrations of alpha-tocopherol ( $\alpha$ T), gamma-tocopherol ( $\gamma$ T), BHT, S1 and S2 were studied in *in vitro* lipid peroxidation systems (Ann.Rep.1996-98 & 1998-99). The results showed that S1 has antioxidant effects in both cumene hydroperoxide (CumOOH) and iron ascorbate mediated lipid peroxidation systems, whereas S2 showed inhibition only in the CumOOH system. When varying levels of tocopherol were added to 40 nmoles of either S1 or S2, the inhibition of lipid peroxidation in both the systems was found to be greater than the calculated additive effect, thereby indicating that S1 or S2 have synergistic effects with tocopherol.

During this year, lignan and gamma-tocopherol concentrations were determined in 10 commercially available oils and the effects of S3 was evaluated in both the lipid peroxidation systems. The synergistic effects of S1 and S2 with tocopherols were evaluated at lower concentrations (20 nmoles) and that of S3 was done at both 20 and 40 nmoles.

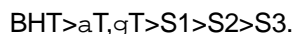
The results are as follows:

1. Total lignan content in oils ranged between 7-16 g/kg oil and gamma-tocopherol concentrations were between 400-800 mg/Kg oil. Total lignan content in oils was similar to the data reported on 19 sesame seed varieties but were lower as compared to the two Assam seed varieties (28 g/kg oil).

### 2. Lipid peroxidation

a) **CumOOH system**: Based on the entire data the activity of various compounds re-

quired to bring about 30% inhibition of lipid peroxidation is as follows:



The antioxidant properties of lignans (20 nmoles) of S1 or S2 or S3 with varying concentrations of alpha or gamma tocopherols showing 30% inhibition of lipid peroxidation was found to be 8 fold high with S1, 6 fold high with S2 and two fold high with S3 as compared to either alpha or gamma tocopherols. With higher concentrations of lignans (40 nmoles), the magnitude of synergistic effects further enhanced.

**b) Iron ascorbate system:** S3 did not show any inhibition in this system but in combination with either alpha or gamma tocopherols it enhanced the inhibitory effects of the tocopherols. These findings are similar to those observed with S2 (Ann. Rep. 1996-98).

These findings suggest that sesame lignans can potentiate the antioxidant properties of tocopherols and contribute to their beneficial role as dietary antioxidants. Therefore, sesame lignans can be recommended as natural antioxidants in edible oil and food industry. Further, the use of sesame oil in combination with other oils will increase the antioxidant potential of oils. This work has provided a scientific basis for the health claims of sesame seeds and oil in ayurvedic medicine.

## 2. Lipids, fatty acid composition and tocopherols in foods

The Institute's publication "Nutritive Value of Indian Foods" does not give data on fatty acid composition, cholesterol and tocopherol contents of foods. In view of their importance for normal physiological function and in the prevention of diet-related chronic degenerative diseases, it is important to generate this data-base. The total lipid and fatty acid composition of some commonly consumed foods (cereals, pulses, spices and vegetables) was published earlier (Eur. J. Clin. Nutr. 1989 and Food Chem. 1993). During this year, analysis of total lipid and fatty acid composition was carried out in fruits and biscuits.

Three market samples of banana, chikoo, grape, guava, papaya and plum and seven varieties of mangoes were purchased and analysed for total lipid fatty acid composition.

The results are as follows:

1. The total lipid content in fruits ranged between 0.22 to 1.1%.
2. The mean percentage of fatty acids were: SFA 0.15%; MUFA 0.1% and PUFA 0.17%.
3. The ALNA levels were higher in plum, papaya and guava (0.4%) as compared to banana, grape, mango and chikoo (0.2%).
4. Out of the seven different varieties of mangoes analyzed, the mean levels of ALNA in three varieties (alphonso, rasalu and raspuri) was 0.03% as compared to 0.01% in the

other four varieties (jehangir, benisha, badami and totapuri). On an average, about 400 g of fresh fruits provide about 0.1 g of ALNA. It was earlier reported that about 400 g vegetables on an average provide 0.1 g ALNA but green leafy vegetables contain seven times more (Food Chem.1993). Thus increasing the consumption of fruits and vegetables can contribute to increasing n-3 fatty acid intake in addition to providing several other nutritional benefits.

Indian vanaspati is prepared by partial hydrogenation of vegetable oils. It contains >20% *trans* fatty acids (TFA) and >60% of saturated fatty acids (SFA). As SFA and TFA raise LDL cholesterol, one of the dietary recommendations for prevention of CHD is that the total intake of SFA and TFA should not exceed 8 en% (20 g/d). Since vanaspati is used in the preparation of bakery products, the total lipid and fatty acid composition of five different types of biscuits (3 market samples of each) were analysed. The total fat content ranged between 11 to 13% and the mean percentage of fatty acids were : SFA, 5%; Cis-MUFA 3%, *trans*-MUFA 4% and PUFA 1%. Consumption of 100 g of biscuits (7 to 10 pieces) in a day provides ~4 en% (9 g) of SFA and TFA i.e., half of the upper safe limit of SFA and TFA together.

### 3. Hypoglycaemic and hypocholesterolemic properties of galactomannan isolated from fenugreek seeds in obese rats (WNIN/GR-Ob)

Fenugreek seeds are rich sources of total (48%), insoluble (28%) and soluble (20%) dietary fiber. The hypoglycaemic and hypocholesterolemic properties of fenugreek seeds in animals as well as in normals and diabetic patients are well demonstrated. It is suggested that galactomannan, a polysaccharide, is responsible for its antidiabetic properties.

In the present investigation, galactomannan (soluble dietary fiber) was isolated from fenugreek seeds and its hypoglycaemic and hypocholesterolemic properties were studied in obese rats (WNIN/GR-Ob) of Institute's colony. Fifty day old obese rats (WNIN/GR-Ob) were randomly divided into five groups of six males and five females each. The design of the experiment was as follows:

Group	Diet	No. of animals	
		Male	Female
I	Control	6	5
II	10% Fenugreek	6	5
III	2.5% galactomannan	6	5
IV	20% Fenugreek	6	5
V	5.0% galactomannan	6	5

All the above diets were made isonitrogenous and they were fed to animals for a period of 9 weeks and the following parameters were studied:

1. Fasting plasma glucose levels at 0,3,6 & 9 weeks.
2. Fasting plasma insulin levels at 0 & 9 weeks.
3. Fasting erythrocyte insulin receptors at 9 weeks.
4. Serum total cholesterol levels at 0, 3, 6 & 9 weeks.
5. Serum triglycerides levels at 0, 3, 6 & 9 weeks.
6. Faecal bile acids and neutral sterols at 9 weeks.

The results of the experiment are summarised as follows:

### **Hypoglycaemic effects**

1. Addition of 2.5 and 5.0% galactomannan as well as 10 and 20% fenugreek seed powder to the control diet brought about a significant ( $P<0.05$ ) decrease in the fasting plasma glucose levels of male and female rats at 6 and 9 weeks, while a significant ( $P<0.05$ ) increase was observed in control male and female rats (**Fig.14**).
2. Supplementation of control diet with two levels of galactomannan and two levels of fenugreek seed powder brought about a decrease in fasting plasma insulin levels. However, due to large variation, insulin values were not significantly different. On the other hand, fasting plasma insulin levels showed a significant increase ( $P<0.05$ ) by 9th week in control rats.
3. Fasting erythrocyte insulin receptor levels were significantly higher ( $P<0.05$ ) in all the four groups of experimental animals as compared to the corresponding control animals.

The results indicate that supplementation of diets with galactomannan to obese rats improved their glucose utilisation.

### **Hypocholesterolemic effects**

4. Feeding of diets containing 2.5 & 5.0% galactomannan and 10 & 20% fenugreek seed powder to obese rats significantly ( $P<0.05$ ) decreased the serum total cholesterol levels after 9 weeks in both males and females, while a significant ( $P<0.05$ ) increase was observed in control males and females (**Fig.14**).
5. Addition of galactomannan or fenugreek seed powder to the diets of rats resulted in significant ( $P<0.05$ ) decrease in serum triglyceride levels in experimental males and females from 3rd week onwards, while significant ( $P<0.05$ ) increase was observed in control males and females at 9th week (**Fig.14**).
6. Supplementation of stock diet with either galactomannan or fenugreek significantly increased faecal excretion of bile acids and neutral sterols in experimental rats.

Thus, fenugreek seed powder, a rich source of soluble fiber, decreased the serum

cholesterol and triglycerides with a concomitant increase in faecal bile acid and neutral sterol excretion in experimental rats.

#### 4. Effect of spice principles on cataractogenesis

Increased oxidative stress in the lens is implicated in cataractogenesis. Recent evidence suggests that optimal intake of antioxidants may prevent/delay, the onset of several age dependent diseases, including cataract.

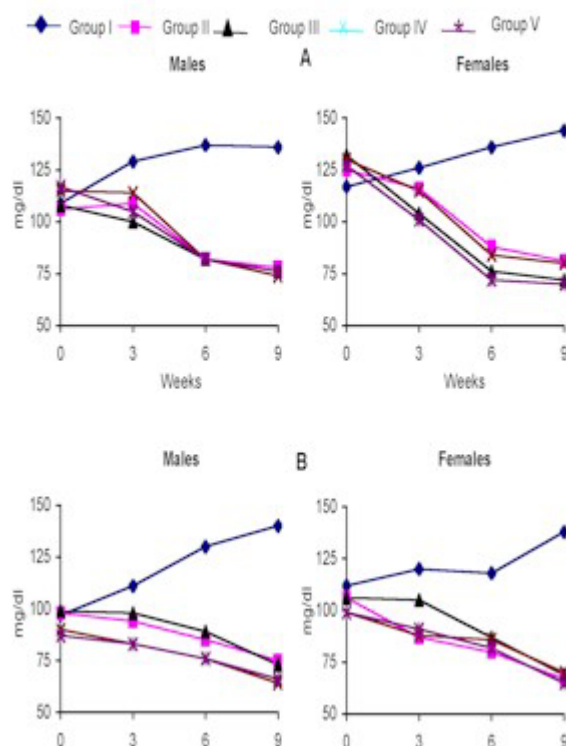
Eugenol is an active principle in cloves, which is shown to act as an antioxidant by lowering lipid peroxidation in rats with iron induced hepatic toxicity *in vivo* and inhibiting the non-enzymatic lipid peroxidation in rat liver mitochondria *in vitro*.

Studies were therefore conducted to assess the protective effect of eugenol addition on biochemical changes associated with opacity in rat lens during FeSO<sub>4</sub> (100 mM); Ascorbate (2 mM) induced damage *in vitro* in Tris (20 mM)-KCl (125 mM) buffer pH 7.4.

The following biochemical changes were observed.

1. During FeSO<sub>4</sub>-Ascorbate induced oxidative stress, MDA (Mean±SE) levels (24.25±1.65 nmoles/g tissue, n=4) were increased significantly (P<0.05) as compared to the controls [11.76 ± 0.85 nmoles/g tissue, n=4], indicating increased lipid peroxidation. Upon co-incubation with eugenol at 100 mM and 150 mM levels, there was a significant (P<0.05)

Fig. 14  
Effect of Fenugreek Seeds and Galactomannan Isolated from Fenugreek Seeds on (A) Fasting Plasma Glucose (B) Serum Total Cholesterol, and (C) Serum Triglycerides Levels in Obese Rats



decrease in the MDA levels [ $16.37 \pm 1.61$  nmoles/g tissue and  $13.73 \pm 1.75$  nmoles/g tissue], in a dose dependent manner. However, eugenol at 50 mM concentration did not show any significant effect ( $18.23 \pm 1.67$  nmoles/g tissue, n=4) on MDA levels.

2. The tryptophan fluorescence (area under the peak) in soluble protein fraction significantly ( $P < 0.01$ ) decreased ( $21.82 \pm 3.35$ /mg protein) as compared to control ( $42.4 \pm 4.47$ ). Co-incubation with eugenol at 150 mM concentration significantly ( $P < 0.05$ ) improved tryptophan fluorescence ( $33.14 \pm 5.01$ , n=5).

3. Hexokinase activity was significantly ( $P < 0.05$ ) decreased ( $2.032 \pm 0.376$  mmoles NADP reduced/hr/100 mg protein, n=5) as compared to the controls ( $3.35 \pm 0.704$  mmoles NADP reduced/hr/100 mg protein, n=5). Co-incubation with eugenol at 150 mM concentration significantly improved the enzyme activity ( $2.87 \pm 0.18$  mmoles NADP reduced/hr/100 mg protein, n=5).

4. Glucose-6-phosphate dehydrogenase activity also decreased ( $6.04 \pm 0.627$  mmoles NADP reduced/hr/100 mg protein, n=5) significantly ( $P < 0.05$ ) as compared to controls ( $8.35 \pm 0.850$  mmoles NADP reduced/hr/100 mg protein, n=5). Co-incubation with eugenol at 150 mM concentration significantly ( $P < 0.05$ ) improved the enzyme activity ( $7.55 \pm 0.174$  mmoles NADP reduced/hr/100 mg protein, n=5).

5. However, there was no change in total as well as soluble protein content before and after oxidative stress.

The results suggested that eugenol protects rat lens against  $\text{FeSO}_4$ -Ascorbate induced biochemical damage *in vitro*. Protective effect of eugenol on biochemical changes during  $\text{FeSO}_4$ -Ascorbate induced changes will be studied next in animal model of cataract.

## **5. Studies on food intake regulation, lipid metabolism in WNINOb/WNIN GR-Ob rats**

The aim of the study was to understand the mechanism of food intake regulation and obesity in WNIN/Ob & WNIN/GR-Ob. With this broad objective, experiments were designed to achieve the following specific objectives:

i) To determine the levels of leptin a cytokine which is known to play a role in food intake regulation.

ii) To study the activity of acetyl cholinesterase (Ach E) which is known to be altered by feeding behaviour.

iii) To study the activities of sodium and potassium pumps of these animals which reflect

the cellular energy utilization and thereby affect the energy homeostasis of these animals.

### **Studies on leptin levels of WNIN/Ob and WNIN/GR-Ob**

Among these three above mentioned objectives, the first one has already been studied and reported (Ann. Rep. 1997). The obese rats of WNIN/Ob and WNIN/GR-Ob have very high levels of immuno reactive leptin. However, whether this leptin is biologically active or these animals have receptor deficiency has to be established.

### **Studies on acetylcholinesterase**

Apart from hormone leptin, which is known to regulate food intake, the activity of serum acetylcholinesterase (AChE), which is known to be altered by feeding behaviour and hyperlipoproteinaemic conditions, was studied during this year. Although the precise function of plasma cholinesterase is not known, it has been implicated in lipid metabolism. Increased activity of this enzyme has been reported in hyperlipoproteinaemia, obesity, diabetes, CVD and also in genetically obese mice. Fasting and PEM are known to decrease AChE activity.

The results can be summarised as follows:

- 1) Lean male rats of WNIN/Ob and WNIN/GR-Ob did not show any increase in the activity of AChE by 200 days compared to their 100 days controls.
- 2) On the other hand, obese male rats of WNIN/Ob and WNIN/GR-Ob showed a significant increase in the activity of this enzyme at 200 days compared to that of 100 days old obese counterparts of the two strains (1.53  $\mu$ moles of acetylcholine released/min/ml at 200 days in male obese rats of WNIN/Ob as against 0.64  $\mu$ moles/min/ml at 100 days old male obese rats of WNIN/Ob strain).
- 3) Lean as well as obese female rats of WNIN/Ob and obese female rats of WNIN/GR-Ob showed almost double the activity by 200 days compared to their respective controls of 100 days age. However, lean females of WNIN/GR-Ob did not show any such change.

Although at 100 days obese rats of both strains had lower activity, by 200 days the activity of this enzyme in obese rats was much higher than the lean animals. The higher AChE activity at 200 days age was similar to the one reported for genetically obese mice compared to their lean counterparts. The AChE is known to be an inducible enzyme and perhaps the flux in fatty acyl COA due to hyperphagia may be responsible for this induction. In addition, there are also reports to suggest that hyperinsulinemia occurs much before hyperphagia becomes evident in the obese mice which are leptin-deficient. Leptin is known to constrain the acetylcholine-mediated insulin secretion from pancreatic islets in WNIN/Ob and WNIN/GR-Ob animals. In addition to non-functional leptin (whether it is a

defect in leptin or receptor), acetylcholine levels could be high in obese rats at 100 days ~~due to lower AChE activity which was getting reversed by 200 days. A study of this enzyme below 100 days of age may throw some light on the early event that leads to hyperinsulinemia in these animals.~~

### **Studies on erythrocyte membrane Na<sup>+</sup>, K<sup>+</sup>-ATPase Ca<sup>2+</sup>, Mg<sup>2+</sup>-ATPase of WNIN/Ob and WNIN/GR-Ob rats**

In simple terms, the genesis of obesity may be due to a positive energy balance, input exceeding the output. It involves excess storage of food derived calories as triglycerides in adipose tissue depots due to either excess calorie intake or decreased calorie expenditure or both. The obese rats of WNIN/Ob and WNIN/GR-Ob strains are hyperphagic. However, the animals may also have defects in energy utilization. Therefore, studies were carried out to understand energy utilization at the cellular level. Na<sup>+</sup> pump Ca<sup>2+</sup> pump in the form of ATPases together contribute to the 25-35% of cellular energy utilization. ~~Therefore, the activities of erythrocyte membrane Na<sup>+</sup>, K<sup>+</sup>-ATPase and Ca<sup>2+</sup>, Mg<sup>2+</sup>-ATPase which are reflections of the activities of these pumps were studied.~~

The results are as follows:

1. It was observed that ouabain sensitive-ATPase (Na<sup>+</sup>, K<sup>+</sup>-ATPase) and Ca<sup>2+</sup>, Mg<sup>2+</sup>-ATPase) were significantly lower in male obese rats of WNIN/Ob strain compared to their lean counterparts (Na<sup>+</sup>, K<sup>+</sup>-ATPase activity 0.84  $\mu$ moles Pi/mg/hr and Ca<sup>2+</sup>-ATPase 1.18  $\mu$ moles Pi/mg/hr in obese rats as against 1.02 and 1.45  $\mu$ moles Pi/mg/hr respectively in lean rats).
2. However, obese male rats of WNIN/GR-Ob strain did not show such a change in the activities of these enzymes.
3. Activity of acetylcholinesterase, yet another membrane-bound enzyme, displayed significantly lower activity in the obese male rats of both WNIN/Ob and WNIN/GR-Ob strains.

Based on these observations, it is planned to study the following aspects:

1. The mitochondrial oxidation of which 80% is coupled to ATP synthesis and 20% is uncoupled by mitochondrial proton leak.
2. To understand the role of brown adipose tissue (BAT) and white adipose tissue in obesity and its regulation by vitamin A.

## **VI. OTHERS**

## 1. Low-cost nutrient supplement for malnourished children - A biotechnological approach

Protein energy malnutrition is one of the commonly seen nutritional disorder in children belonging to poor communities in developing countries like India. A project was undertaken jointly by the 3 Institutes, namely, NIN, Central Food Technological Research Institute (CFTRI) and National Dairy Development Board (NDDB) with the financial support of DBT to develop a low-cost nutrient supplement using a biotechnological approach to combat malnutrition in children.

The main objectives of this study were :

1. To test the composition, nutritive value and biological value of the food preparations developed by NDDB and CFTRI.
2. To test the organoleptic properties of the food preparations.
3. To undertake the evaluation of shelf-life of preparations and to estimate the content of anti-nutritional factor like trypsin inhibitors in the food preparations.
4. To study the clinical evaluation of the food products in school children.

Organoleptic evaluation of the three preparations (*Suruchi meetha*, *Nutrocrispo sweet* and *Nutrocrispo spicy*) developed by CFTRI, was carried out along with comparable snacks from the commercial sector in 58 adult volunteers (both male and female from NIN staff). The products had a minimum protein content of 16% and basically contained defatted soya flour, wheat flour, peanut, sesame seeds, wheat germ, non-fat dry milk and jaggery. Some of the ingredients were treated biotechnologically for better nutrient availability.

The results can be summarized as follows :

1. In the case of *Suruchi meetha*, a burfi like sweet, the overall rating was 3.0 against 3.9 of the market reference. In all the characteristics, this preparation received a significantly lower score than the reference. However, the absolute score was 3.0 indicating satisfactory acceptability by the preparation.
2. The scores for colour, appearance and texture of *nutrocrispo sweet* were comparable to those of a market sample, while those of aroma, taste and after taste were slightly lower, but the difference was statistically significant. Even the overall rating though lesser than the market reference, it was more than satisfactory with a score of 3.1.
3. The results obtained with *nutrocrispo spicy* was rated uniformly poorer as compared to the reference. In the case of aroma, taste, after taste and overall quality, the score was close to 2 and was not acceptable. This product needs improvement before it is consid-

ered for evaluation.

## 2. Beneficial role of Solanaceae family plant leaf powder in poultry feed

The proper utilisation of calcium is under the control of vitamin D<sub>3</sub>, particularly its active metabolite 1,25-dihydroxy cholecalciferol [1,25(OH)<sub>2</sub>D<sub>3</sub>]. The hormonally active form of vitamin D<sub>3</sub> i.e., 1,25(OH)<sub>2</sub>D<sub>3</sub>, essentially serves to maintain serum Ca and P levels by acting on classical target organs namely intestine (increased absorption), bone (mobilisation) and kidney (reabsorption). Apart from these functions, it has recently been found to have many more important functions in the body, like in the differentiation of haematopoietic stem cells, inhibition of growth of cancer cells, immune potentiation, insulin secretion, treatment of resistant rickets, hypoparathyroidism and osteoporosis.

1,25(OH)<sub>2</sub>D<sub>3</sub> is available commercially but it is very expensive as it is chemically synthesized. Hence, an attempt was made to find a cheaper and inexpensive source of the chemical, which has great utility not only in human health, but also in veterinary medicine and animal husbandry.

### *Vitamin D like activity in plants*

The active principle in *Solanaceae* family plants was identified earlier which mimics the biological activity of 1,25(OH)<sub>2</sub>D<sub>3</sub>. It was found that *Cestrum diurnum* (CD) leaves of this region of the world (J. Sci. Food & Agr. 62: 19, 93; Phytochemistry 37: 617, 1994), contain three times higher concentrations of 1,25(OH)<sub>2</sub>D<sub>3</sub> (about 1 mg/kg dry leaves) than that reported from else where, which may be because of geographical reasons.

The efficient use of calcium in chicks may result both in better egg production and in increased egg shell thickness. Indirectly, it may also result in better growth in broilers due to reduction in leg weakness. Hence, studies were carried out on *Cestrum diurnum* leaves to assess its utility in poultry feed.

Preliminary experiments in broilers fed various levels of CD with or without vitamin D in the diet showed that feed efficiency and bone density were better when CD was incorporated at 0.25% in the diet. This was true in the first 2 to 3 weeks of feeding but not till the end of 6 weeks of experimental period. In view of this, the experiment was repeated with the following modifications. Three hundred and twenty, one-day old broiler birds were divided into 8 groups of 40 birds each using randomised block design as given below:

Group	Diet and duration
I	Normal diet (containing vitamin D <sub>3</sub> ) for 6 weeks

- II Normal diet + 0.25% CD for 6 weeks
- III Normal diet + 0.25% CD for 2 weeks and shifted to normal diet for 4 weeks
- IV Normal diet + 0.25% CD for 3 weeks and shifted to normal diet for 3 weeks
- V Vitamin D deficient diet for 6 weeks
- VI Vitamin D deficient diet + 0.25% CD for 6 weeks
- VII Vitamin D deficient diet + 0.25% CD for 2 weeks and shifted to normal diet for 4 weeks
- VIII Vitamin D deficient diet + 0.25% CD for 3 weeks and shifted to normal diet for 3 weeks.

Duration of the experiment was for 6 weeks. Feed and water were given *ad lib*. Daily food intake and weekly body weight were recorded. Five birds in each group were sacrificed at weekly intervals from 2nd week onwards and kidney and blood were collected. Tibia were also collected from these birds.

The following parameters were monitored:

1. Feed efficiency ratio (FER) =  $\frac{\text{Feed intake (kg)}}{\text{Body weight (kg)}}$
2. Serum Ca, P, alkaline phosphatase, cholesterol and triglycerides.
3. Tibia weight and density.
4. Kidney 25-OH-D<sub>3</sub> 1- $\alpha$ -hydroxylase
5. Histopathology of soft tissues.

The results may be summarised as follows:

#### **Kidney 1- $\alpha$ -Hydroxylase (Pmoles 1,25(OH)<sub>2</sub>D<sub>3</sub>formed/mg protein)**

1. The 1- $\alpha$ -hydroxylase activity in the kidney was higher at all the time points from 2nd to 5th week in vitamin D deficient chicks as compared to the corresponding controls. Only at 5th week, the activity in vitamin D deficient chicks (0.056 $\pm$ 0.007 Mean $\pm$ SE) was significantly higher than the controls (0.033 $\pm$ 0.006).
2. As expected, the chicks given CD along with vitamin D deficient diet had similar enzyme levels (0.040 $\pm$ 0.009) as that of control birds and significantly lower than that of the vitamin D deficient chicks.
3. The enzyme values were not significantly different in the other groups.

#### **Feed efficiency ratio (FER)**

1. In general, feed efficiency was found to be better in chicks given CD either with control or vitamin D deficient diets as compared to their respective controls. However, the differences were not statistically significant between control and control + CD.
2. There was a significantly ( $P<0.05$ ) higher FER ( $2.07\pm 0.007$ ) in chicks given CD along with vitamin D deficient diet than the vitamin D deficient chicks ( $2.20\pm 0.043$ ).

### **Bone weight and density**

1. The fat-free dry weights of tibia were significantly ( $P<0.05$ ) higher in all the groups which received CD leaf powder as compared to their respective controls. The increase was found both in control diet and vitamin D deficient diet fed chicks.
2. Bone density ( $0.706\pm 0.007$ ) was significantly higher ( $P<0.05$ ) in chicks which received CD along with vitamin D deficient diet as compared to vitamin D deficient chicks ( $0.600\pm 0.007$ ).
3. The increase in bone density was also observed in CD given chicks ( $0.737\pm 0.009$ ) along with control diet ( $0.706\pm 0.0118$ ), but the difference was not statistically significant.

### **Serum parameters**

1. Serum Ca ( $7.3\pm 0.12$  mg/dl) was significantly low in chicks ( $P<0.001$ ) fed on vitamin D deficient diet as compared to control ( $10.6\pm 0.20$  mg/dl) chicks.
2. CD administration along with vitamin D deficient diet increased the serum levels of calcium significantly ( $P<0.001$ ) to  $10.67\pm 0.401$  mg/dl.
3. No differences in serum P, alkaline phosphatase, cholesterol and triglycerides were observed.

### **Histopathology of tissues**

No lesions of toxicity or calcification were observed in any of the tissues (liver, kidney, breast muscles, aorta, gizzard, proventriculus, lungs and heart) examined.

It may be concluded that broilers given CD leaf powder would result in better feed efficiency and also in improved bone weight and bone density. The improved bone health could be the reason for observing better feed efficiency with CD leaf in the diet.

## **VII. PATHOLOGY**

### **1. Effect of varying carbohydrate diets on glucose intolerance in WNIN/**

## **GR-Ob rats - Histopathology study**

WNIN/GR-Ob rats are obese and show impairment in glucose tolerance on challenge with glucose, even on a stock diet. In view of this, the effect of various carbohydrate sources in diet on fasting glucose levels was studied. Histopathological studies were done to assess organ changes, if any, due to the exposure to various diets.

A total of 48 adult obese rats, aged 105 days, were taken and divided into 4 groups of 12 each. Each group had 6 lean and 6 obese animals of both sexes with equal distribution and they were fed on 20% stock protein-, glucose-, sucrose- and starch-based diets respectively for 60 days, at the end of which all animals were sacrificed and their pancreas, liver and kidney were taken for histopathology.

Fatty changes in liver were seen in almost all obese animals and a few lean animals, while no significant changes were seen in the pancreas and kidneys in all animals, irrespective of the type of diet consumed by them. The composition of the diet had no adverse effect on organ morphology.

## **2. Pre-clinical toxicological evaluation of orally administered chlorella - acute and subacute - Histopathology study in rats**

Chlorella is a heterotrophic mutant green alga, a native of Japan, and is reported to have beneficial health effects. The toxicological evaluation of this compound was sought to be studied and hence fullfledged pre-clinical toxicology experiments were done.

For acute (LD<sub>50</sub>) study, a total of 29 male Fischer rats, aged 10 weeks were divided into 5 groups and four of them were fed chlorella orally at 0.125, 0.250, 0.500 and 2 g/kg dosages respectively while the last group (control) received none at all, for a period of 10 days.

In the sub-acute toxicity study, 60 animals (equal numbers of both sexes) aged between 14 to 16 weeks, were divided into 6 groups and as in the previous experiment, five groups were fed chlorella at 0.125, 0.250, 0.500, 1.000 and 2 g/kg respectively, while the last group was control. The feeding was done in all groups for a period of 15 days, at the end of which half the animals (15 male and 15 female) were sacrificed, while the other half were maintained on stock diet for a further period of 15 days and then sacrificed.

Brain, heart, lungs, liver, spleen, kidneys, stomach, small intestines and respective sex organs of all animals were taken and subjected to histopathology study.

The results are as follows:

The various histopathology changes seen in the organs were found in all groups and

these are common to all conventional colony-bred animals except for non-specific focal myocarditis seen in a few animals (of both acute and subacute studies).

Thus, there were no significant histopathological changes in the various organs which could be attributed to administered chlorella, at various dose levels for varying periods.

### **3. Effect of carcinogen on Fischer rat oesophagus in zinc deficiency and after rehabilitation**

Zinc deficiency is known to increase the risk of oral and oesophageal cancers. Earlier studies on underlying mechanisms suggest that alterations in carcinogen metabolizing enzymes, DNA adducts and antioxidant status in zinc deficiency, increase the risk of cancer. The effect of carcinogen MBN (methylbenzoniolate) in zinc deficiency and subsequent rehabilitation was studied.

Fifty seven weanling, Fischer male rats were divided into 5 groups. The group I was 'control' (n=19) fed on zinc sufficient diet (30 mg/kg), groups II (n=10) and III (n=9) were fed on zinc deficient diet while groups IV (n=10) and V (n=9) were paired with groups II and III respectively. All the groups were fed with carcinogen for 4 weeks and discontinued, after which the II and IV groups were rehabilitated with adequate zinc (while III and V were not rehabilitated) for a further period of 2 weeks. All animals were sacrificed at the end of 6 weeks and their oesophagi were taken for histopathology study.

The various histopathology features were graded on 1 to 10 scale ranging from normal (I) to invasive carcinoma (X).

The results are as follows:

It was seen that focal papillary hyperplasia (feature 5) was significantly different between groups I and V, while papilloma formation (feature 8) was significantly different between group I vs IV, II vs IV and III vs V also. All other histopathology features were seen across the various groups, but were not significantly different.

Thus, it can be inferred from the present study that zinc deficiency induced papilloma formation, and rehabilitation did not have any effect. Also, lower food intake, by itself reduced the extent of papilloma formation as well as papillary hyperplasia.

## **VIII. EXTENSION AND TRAINING**

### **1. Publications**

Popular articles on health and nutrition, contributed by NIN scientists, continued to

be published regularly in four quarterly periodicals viz., Nutrition News, Nutrition (English), Poshan (Hindi) and Poshana (Telugu). These articles have not only been widely quoted in mass media, but were reproduced in other regional periodicals after duly acknowledging the source.

Based on our recent publication "Dietary Guidelines for Indians - A Manual", a special publication "Dietary Guidelines for Vulnerable Groups" was brought out in Telugu, in collaboration with CARE for free distribution to ICDS functionaries in order to help them work more effectively for the improvement of nutritional status, particularly of pregnant and lactating women, infants and children. The book was released during the two-day Workshop on "Nutrition intervention in the next Millennium", jointly organised with CARE on October 5-6, 1999. Translation of the publication "Dietary Guidelines for Indians" into other Indian languages is in progress.

The publications and periodicals of NIN continued to be popular among the public, and from their sale, over Rs.2,70,000/- was generated, which is higher than the previous year's sale. Due to higher sales during the year, stocks of several publications got exhausted and thus they were reprinted. These include: Nutritive Value of Indian Foods, Dietary Guidelines for Indians - A Manual and a booklet (English), Some Therapeutic Diets, Nutrient Requirements and Recommended Dietary Allowances for Indians, Low Cost Nutritious Supplements (English and Telugu) and Dietary Tips for the Elderly.

## **2. Training Programmes**

Iron deficiency anaemia affects a major segment of Indian population, especially women and children. However, due to lack of standardization, it is recognized that considerable variation exists, in the use of simple as well as sophisticated biochemical tests for assessment of anaemia. In view of this, a new training course on "Techniques for assessment of nutritional anaemias" was organised from December 1-10, 1999. Twelve senior teachers, working in the departments of General Medicine, Pathology, Biochemistry, Obstetrics & Gynaecology, Foods and Nutrition from various medical and home science colleges attended the course.

In the wake of the media scenario having undergone a sea change in the recent years, there is a felt need to equip the scientists in biomedical research with the updated knowledge on IEC aspects. Recognizing the importance of need-based health communication, a five-day ICMR sponsored "Workshop on Information, Education and Communication in Biomedical Sciences" was organised from December 20-24, 1999. Seventeen scientists from various ICMR institutes and centres participated in the Workshop and they were trained in the communication skills used in print and electronic media.

A ten-day "Orientation Course on Nutrition for Officials of Food & Nutrition Board" was conducted in two batches by the Field Division. Over fifty officials from different parts of country participated in the programme.

Three WHO Fellows from the Department of Health Services, Kathmandu, Nepal, attended *ad hoc* training courses on maternal child health, conducted in two batches in July and November, 1999.

Besides the above *ad hoc* training courses, three regular annual training courses viz., M.Sc. (Applied Nutrition), Post-Graduate Certificate Course in Nutrition and Annual Training Course in Endocrinological Techniques and their Application were organised. Six non-sponsored private candidates were admitted for the first time in Certificate Course in Nutrition. Their admission was also a source of income generation for the Institute.

### **3. Extension Activities**

#### **3.1 Exhibitions**

As part of extension activities, the division participated in four exhibitions, three held in Hyderabad and the other in New Delhi. Posters, related to health and nutrition, were displayed in the exhibition organised by the local Air-Force Officers' Cooperative Housing

Society at Vayupuri, on July 25, 1999 and the importance of nutrition and balanced diet was explained, particularly to the underprivileged public, who thronged the exhibition.

LIFE, a Hyderabad based non-governmental organization, had organised an exhibition on Food and Hygiene for school children from October 25 to 27, 1999 in Chaitanyapuri, Hyderabad, and ET division participated in the event, and the stall displaying the exhibits was a major attraction.

In connection with the Children's Day, Andhra Pradesh Academy of Sciences conducted "Science exhibition of the Millennium" at Bharathiya Vidya Bhavan, Hyderabad, from November 13-14, 1999. The ET division displayed the exhibits on nutrition in the exhibition and arranged for the sale of NIN publications for the benefit of children, teachers, parents and other general public, who visited the exhibition in large numbers.

As part of the Swadeshi Vignam Mela, a Science and Technology exhibition was organised in New Delhi from February 2 to 6, 2000 and the Institute participated in the event. Besides the sale of NIN publications and display of posters on nutrition, interactive food adulteration detection methods were demonstrated.

### **3.2 Popular Talks**

A lecture was given on "Dietary habits for better health" to the executives, managers of MIDHANI, Hyderabad on June 20, 1999.

Two lectures on "National Nutrition Programmes" and "Health and Nutrition" were delivered by the staff of the ET Division in the training programme, conducted jointly by the Department of Women & Child Development and Food and Nutrition Board on July 20, 1999.

A popular talk on "Balanced Diet" was given to a group of women, at a function, organised by Aga Khan Foundation on August 5, 1999.

A lecture-cum-demonstration on "Weaning foods" was organised for the benefit of the slum dwellers in Sriram Nagar in association with students from College of Social Work, on September 8, 1999.

Popular lectures on "Adolescent health" were given by the scientists of ET division to the school children at Shankarpalli village, at a programme arranged by 'Panchavati', a NGO, on September 26, 1999.

A lecture on "Nutrition and Health" was delivered to the women employees of Income-Tax Department on October 27, 1999.

A popular talk on "Importance of nutrition" was delivered to the pregnant and lactating

women residing in a slum of Secunderabad, on November 17, 1999. Demonstration on cooking of low cost nutrition supplements was also arranged.

A popular lecture on "Youth and Nutrition" was given to NSS volunteers who were engaged in community development projects, on November 19, 1999.

Two talks on "Dietary Guidelines for vulnerable groups" and "National Nutrition Programmes related to RCH" were given by the staff to the Municipal Councillors and other community leaders, at the training camps conducted at the Indian Institute of Health and Family Welfare, Hyderabad in November and December 1999.

### **3.3 TV Films and Radio Talks**

The scientists of ET division, as resource persons, provided the script and other inputs required for the production of the following six tele films, produced by the Educational Media Research Centre (EMRC), Hyderabad : (i) Fighting malnutrition (ii) Nutritional problems and challenges (iii) Bridging the calorie gap (iv) Adding pink tinge to health (v) Drops for life, and (vi) "Hum Nagarik Vigyan Lok ke (Hindi).

In addition, several radio talks on nutrition themes, delivered by the staff were broadcast over All India Radio, Hyderabad. On special events like National Nutrition Week and Breast-feeding week, a series of radio talks were arranged to create awareness among the public on the importance of the event.

### **3.4 Health Camps**

A ten-day "Health and Nutrition Awareness camp" was conducted from October 2-12, 1999 in a slum, in collaboration with NSS wing of the Kasturba Gandhi College, Secunderabad. A series of street-plays and lecture-cum-demonstration programmes were organised for the slum-dwellers.

On the request of NSS Regional Centre, Ministry of Human Resource Development, Govt. of India, ET division conducted a training programme for the district level NSS functionaries at Chamber of Commerce, Vijayawada, from October 1-16, 1999 to educate them on various issues related to "Health and Hygiene". They were also trained in developing posters on health and nutrition for imparting nutrition education to the community.

### **3.5 Special Events**

#### **3.5.1 National Nutrition Week**

In connection with the National Nutrition Week, several extension programmes were undertaken during September 1-7, 1999, which included Inter-school Painting competi-

tion for school children, Orientation Course on Nutrition for 22 voluntary organisations and Nutrition Awareness programme in a local slum. In addition, a popular talk on "Youth for Healthy Society" for NSS volunteers and five radio talks on nutrition-related topics on AIR were arranged.

### *3.5.2 World Food Day*

As part of World Food Day, a Symposium on "Commercialization of traditional foods" was organised jointly with Association of Food Scientists and Technologists (Hyderabad Chapter), on October 16, 1999.

### *3.5.3 National Science Day*

A series of programmes were organised in connection with the National Science Day during the last week of February. A lecture reflecting the theme of NSD "Recreating interest in basic sciences" was organised and an inter-college elocution composition in Hindi was held to commemorate the event.

## **4. Public Relations**

Regular liaison with the mass media - Radio, TV and Newspaper organisations, was maintained throughout the year. Press releases relating to all scientific deliberations taking place on the campus were issued and media coverage of the Institute's activities was ensured.

Apart from attending to the technical correspondence, dietary counselling was provided to the public. In addition, suitable modifications were suggested for improving the diets of industrial workers and residential school children, when concerned organisations approached the Institute.

Several batches of school and college going students and other community health workers from all over India visited the Nutrition Museum. Apart from popular talks on nutrition and activities of the Institute, visitors were shown the tele film on NIN, video films of various aspects of nutrition.

## **IX. FOOD AND DRUG TOXICOLOGY RESEARCH CENTRE**

### **A. FOOD SAFETY**

#### **1. Bioactive substances in less known legumes**

Legumes play an important role in the human nutrition in India. There are a number of lesser known legumes which need to be exploited. In view of the presence of bioactive substances and involvement of these legumes in export/import trade, there is a need for

generation of scientific data to provide new information on biologically active compounds.

The present study was undertaken with the objectives of :

- 1) Carrying out detailed nutritional and toxicological evaluation.
- 2) Studying the nature of bioactive substances.
- 3) Studying the effect of various processing methods to remove or detoxify the active substances, if present.

Plants of *Cassia sps.* grow in a wild state all over India and produce seeds which are occasionally collected and exported. Cassia is reported to be a source of gum. Seeds of different Cassia plants were collected from different locations, seeds sown and plants were raised till flowering stage. Based on floral characteristics, plants were identified. The different species identified include *Cassia tora*, *Cassia sophera*, *Cassia occidentalis* and *Cassia obtusifolia*. However, *Cassia tora* was selected for further indepth studies.

#### **Development of Method for Gum Extraction**

The two major products obtained from the seeds of *Cassia sps.* are gum and germ meal. Existing methods used for gum extraction could not be applied to *Cassia tora* as the seed coat is very hard compared to seeds of other *Cassia sps.* A modified gum extraction procedure has been standardized. Briefly, the method involved boiling the seeds with alkali solution for 7 minutes cooling and adding 0.2 N hydrochloric acid, followed by dehiscing. It is further washed with water and dried in the sunlight. The seeds are milled and passed through coarse sieve. The endosperm splits remain in the sieve and germ passes through the sieve. The endosperm splits are washed, dried and powdered. The powder is kept in water for further extraction of the gum. Finally, the gum is extracted in isopropanol and washed with acetone. The yield of gum is 36% and germ is 31%. An alternative simple method (dry method) also has been standardized. In this method, seeds are roasted, ground and sieved to obtain endosperm splits. Further, gum is extracted as indicated above. However, in the dry method, germ meal cannot be obtained as by-product unlike in wet method.

In order to obtain gum in large quantities, a pilot scale method has also been standardized. A dehuller used for sesame was found suitable for dehiscing *Cassia tora* seed. In this method, 10 kg of the sample can be processed at a time, and gum yield is 30%.

#### **Proximate analysis of various seed components**

The whole seed has 17% crude protein while germ meal has almost 40%. Since germ powder has high protein, it was further analysed for amino acid profile. It is rich in

leucine (603 mg/g N) and threonine (316 mg/g N) while all other amino acids were comparable to pulses.

### **Studies on germ meal**

Earlier studies carried out at the Marathwada Agricultural University, Parbhani, on germ meal of *Cassia tora* had indicated that it was toxic to rats and detoxification procedures used like autoclaving, acid treatment, alcohol extraction could not remove the toxicity of germ meal. Hence a detoxification method with ethanol, hydrochloric acid treatment followed by hot ethanol extraction was used. The protein efficiency ratio (PER) as well as the toxicity, if any, of the processed germ meal was carried out. PER in Wistar rats indicated that it has very low (23%) efficiency compared to casein diet. However, there were no mortalities.

A 90-day short-term toxicity study did not indicate any toxicity in the experimental rats. During the course of the experiment, on one occasion, 4 experimental rats were found excreting blood stained faeces. But this could not be observed subsequently. At the end of 90 day feeding trial, no visible toxic effects were observed in rats. Histopathological studies of small and large intestine had not shown any significant difference. However, 4 experimental rats had mesenteric lymphadenopathy indicating that there was irritation to the intestine.

### **Utilization of gum and germ meal**

Seed gums are used as thickeners. *Cassia tora* gum was used in pineapple jam at concentrations ranging from 0.05, 0.1 and 0.2%. A concentration of 0.05% *Cassia tora* gum was found to be suitable for pineapple jam preparation. The germ meal, after detoxification could be explored for its use as animal feed ingredient.

## **2. Quality of ready-to-eat (RTE) foods prepared in the unorganized sector**

Ready-to-Eat (RTE) foods are being increasingly accepted as a supplement to the traditional diet. The study on quality of RTE foods prepared in the unorganized sector was carried out with the following objectives:

- i) To find out the type and extent of RTE foods sold in the market.
- ii) To check the quality of RTE foods processed in the unorganized sector under different food laws.

iii) To fix quality parameters for selected RTE foods.

A survey was conducted in the twin cities of Hyderabad and Secunderabad to know the type of RTE foods sold. The survey revealed that there are a variety of RTE foods sold such as bakery products, hard boiled sugar confectionaries, savories, non-alcoholic beverages, ice creams and ice candies. The information on implementation of Prevention of Food Adulteration Act was also collected to find out whether any RTE foods were taken up for investigation. The data revealed that during the five year period, out of 67,043 total number of samples analysed, the number of RTE food samples analysed were only 641. Among these 29 samples were found to be adulterated.

A total of 105 samples of *muruku*, *chekodi* and potato chips were analysed for their quality as per Bureau of Indian Standards (BIS) specifications, and were found not to be conforming to BIS specifications. However, there are no Prevention of Food Adulteration (PFA) standards prescribed for RTE foods. Therefore, it is not possible for the food control authorities to enforce regulations in this regard.

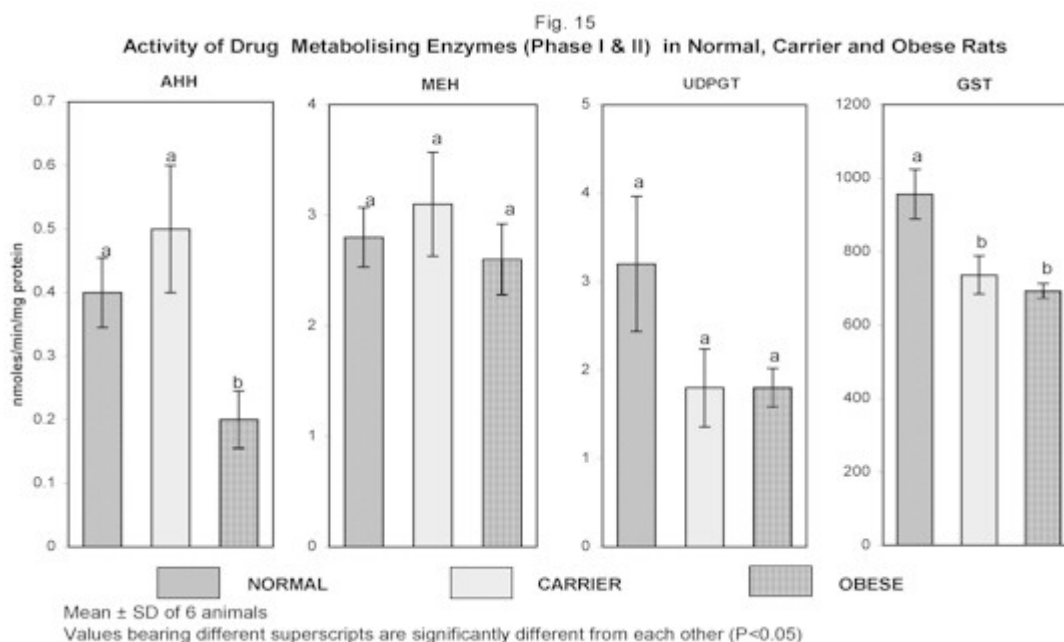
In order to evolve a quality parameter, for the above mentioned RTE foods, a study was conducted to correlate the organoleptic evaluation, with chemical analysis. Among the various chemical parameters such as moisture, acid value, peroxide value, Kries test, p-anisidine value, and thiobarbituric acid tests conducted, only peroxide value had shown a linear correlation with the flavour scores during the initial stages of storage for about 30 days. Such association was not to be found after 90 days of storage, indicating that even peroxide value is not a very good quality indicator for foods stored beyond 30 days. Under the regime of World Trade Organization, all the member countries have to adopt science based standards developed by Codex Alimentarius Commission for the safety of food. However, the specifications prescribed by BIS for *namkeen* cannot be applied for assessing the overall quality of various categories of RTE foods as they are not science based. Therefore, there is a need for further studies to evolve a chemical parameter for each food item for the purpose of testing quality.

As a part of the study, about 100 samples of coloured foods such as sweet meats and hard boiled sugar confectionaries were analysed for colours. About 83.5% of samples contained permitted colours, 15.4% were exceeding the limits prescribed by PFA and 16.4% contained unpermitted colours such as fast red, amaranth, rhodamine, auramine and metanil yellow. A 100 ppm of any permitted colour is allowed according to the PFA Act.

## B. DRUGS/XENOBIOTICS

## 1. Drug metabolism in obese rats

The role of fat in the aetiology of cancer has been a subject of intense investigations. Many epidemiological studies have shown that increased incidence of cancers of breast, endometrium, ovary, pancreas, colon and rectum occur in obese subjects. Higher fat intake is known to increase body weight as well as cancer at certain sites. Since the advent of genetically developed mice and rats, a model is readily available to study mechanisms of carcinogenesis. Such studies were conducted during the course of the year with Wistar/NIN obese rats which have been developed at NIN. To begin with, Phase I and Phase II enzymes of drug metabolism in natural mutant obese rats were analysed (**Fig.15**).



Three groups of male animals, aged between 25-30 weeks, were chosen and designated "obese", "carrier" and "normal (lean control)". After overnight fast, the animals were sacrificed, hepatic microsomes and cytosol prepared and preserved at  $-70^{\circ}\text{C}$  for the assay. The following enzyme estimations were carried out by well established procedures.

Activating Phase : i) Benzo(a)pyrene hydroxylase (BPH)  
ii) Microsomal epoxide hydrolase (MEH)

Conjugating Phase : i) Uridine diphosphoglucuronyl transferase (UDPGT)  
ii) Glutathione-S-transferase (GST)

The results can be summarised as follows:

Of the two activating enzymes studied, benzo(a)pyrene hydroxylase activities were found to be lowest in mutant group, followed by normal and carrier groups (0.22 vs 0.41 vs 0.53 nmoles/min/mg protein). No changes were observed in microsomal epoxide hydrolase activity between groups.

There was a significant reduction in the activity of the conjugating enzymes, GST in both obese and carrier groups of animals compared to controls (693 vs 736 vs 956 nmoles of CDNB conjugated/min/mg protein). UDPGT was, however, not affected. These results demonstrate that the ability to handle xenobiotics is clearly impaired in mutant obese rats as well as in their carriers. Such defect in detoxification mechanisms may lead to increased chemical toxicity and DNA damage. These results suggest that the NIN/Ob/Ob mutant rats could very well serve as a suitable model for studies in xenobiotic toxicity and carcinogenesis with dietary manipulations.

## **2. Hepatoprotective activities of certain medicinal plants from north-eastern region of India**

Medicinal plants are known to be effective in certain diseases and have been in vogue for centuries in many parts of the world. However, they need to be scientifically tested, proved, active principles and dosages identified before introduction into market. Studies were initiated in collaboration with Institute of Advanced Study in Science and Technology (IASST), Guwahati, on some plants, claimed by the local tribals to possess hepatoprotective properties. A pilot study was carried out earlier to identify a suitable model producing liver injury in rats and optimal dosage to be administered. Subsequently, a detailed study was undertaken, which is presently reported.

### **Methodology**

Sixty adult male Wistar rats, weighing 180 - 230 g, were divided into 10 groups of 6 animals each as indicated below :

1. Control
2. Toxin (paracetamol at 2 g/kg b.wt oral route)
3. Toxin + Methanolic Extract of *Leucos levandafolia* (200 mg/kg)
4. Methanolic Extract of Leucos (200 mg/kg) only
5. Toxin + Methanolic Extract of Leucos (400 mg)
6. Methanolic Extract of Leucos (400 mg) only
7. Toxin + Aqueous Extract of Leucos (200 mg)
8. Aqueous Extract of Leucos (200 mg) only
9. Toxin + Methanolic Extract of Costos (200 mg/kg)
10. Methanolic Extract of Costos (200 mg/kg) only

Animals were given stock diet and water *ad lib* during the entire course of the experimentation. For producing liver injury, paracetamol was prepared in 40% sucrose solution and administered at a dose of 2 g/kg body weight by oral route. Methanolic or aqueous extracts of plants at a dose of 200 mg or 400 mg/kg body weight were given to appropriate groups orally, after the administration of the toxin. After 48 hours, rats were sacrificed and blood samples collected for biochemical analyses. Liver specimens were studied for histopathological changes due to paracetamol administration and possible beneficial effects of feeding the various medicinal plant extracts.

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The results can be summarised as follows :

1. Mean liver weight and liver to body weight ratio was highest in the toxin fed group compared to all others.
2. Mean liver SGPT values in the control group was 39.0 U/ml, which was significantly ( $P < 0.05$ ) lower, compared to toxin fed group (76.5 U/ml). The same trend was seen in the values of SGOT also, where the levels in toxin fed group was significantly higher than in the controls (178 vs 118 U/ml).
3. Administration of methanolic or aqueous extracts of *Leucos levandafolia* or *costos*

significantly brought down the toxin-mediated increase in SGOT and SGPT enzymes activity in all groups. The reduction ranged from 25% to 66% in the case of SGPT and 23% to 54% in SGOT enzyme activity.

4. Histopathology studies of the liver were strongly supportive of biochemical findings.

These results suggest that the methanolic or aqueous extracts of some plants from North Eastern Region of India are effective in conferring protection against liver toxicity. In the present study, two such plants have been taken up for screening. Future studies will include more such promising plants as well as identifying their mechanism of action.

#### C. NUTRITION AND CANCER

##### 1. Antioxidant effects of certain medicinal plants from north-eastern India

In recent times, a great deal of interest centres around plant products as remedies for illnesses. They are considered to be less toxic and free from side effects. Many plants and their extracts have been reported to have beneficial effects in a variety of physiological conditions. Last year, in collaboration with IASST, Guwahati, a study was conducted to screen *Leucos levandafolia*, and *Costus* from North Eastern India for their hepatoprotective properties, using a paracetamol-induced liver toxicity model (See previous Section).

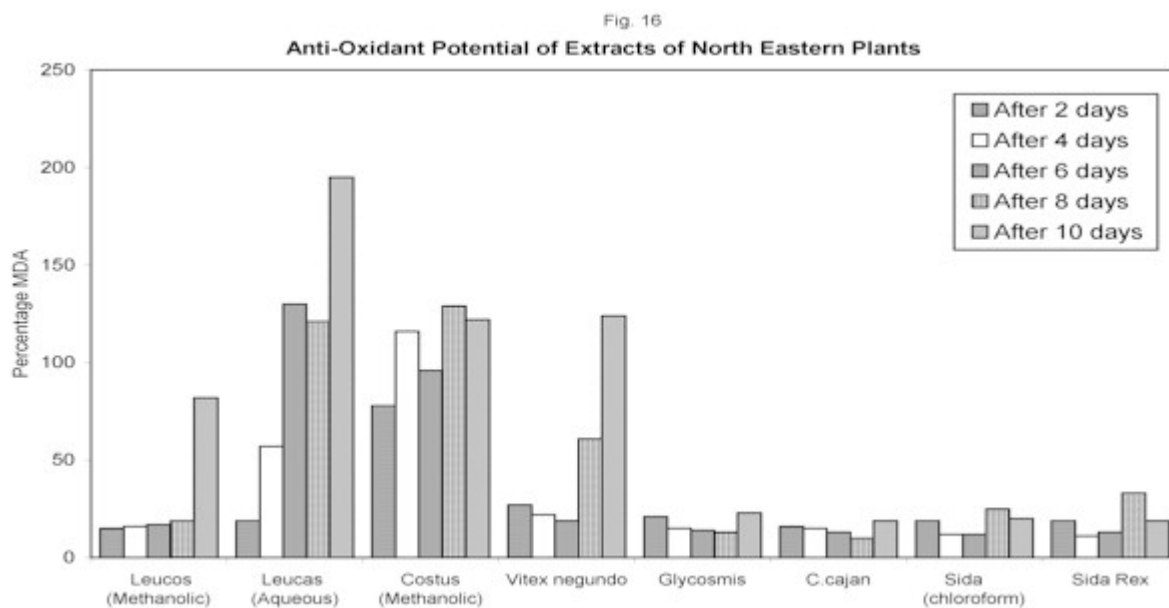
These results demonstrated that (a) there was a significant increase in SGOT and SGPT in paracetamol treated group (b) administration of methanolic or aqueous extract of leucos with the toxin decreased the elevated serum enzymes by 15-30%, and (c) histological studies confirmed the hepatoprotective effect of leucas extract. In order to understand the underlying mechanisms, the present study was undertaken to investigate the antioxidant potential of such plant extracts.

*In vivo* linoleic oxidation assay for the assessment of antioxidant potential of various plant extracts was adopted. It involved the incubation of linoleic acid with plant extracts of established concentrations for different periods of times, estimating the MDA formed and comparing it with control oxidation without plant extracts. The following plant extracts were investigated :

1. Leucos sp. (methanolic and aqueous extracts)
2. Costus sp. (methanolic extract)
3. Vitex negundu “ “
4. Glycosmis sp. “ “
5. C. Cajan “ “

6. *Sida* sp. Chloroform and aqueous extracts

The study was carried out for a period of 10 days. The aliquots were withdrawn at 2, 4, 6, 8 & 10 days to estimate the MDA content (**Fig.16**). The results are represented as nmoles of MDA formed as well as percentage of MDA compared to control incubation (without plant extract which is taken as 100%).



The results can be summarised as follows:

1. The methanolic extracts of leucos, glycosmis, c. cajan, and sida rex as well as chloroform extract of sida were very effective in preventing the oxidation of linoleic acid,

leading to the formation of MDA. The inhibition was between 70-90% of the control. All plant extracts except leucos and vitex demonstrated their antioxidant potential throughout the observation periods.

2. Aqueous extract of leucos and methanolic extract of costos did not show any inhibitory effect on LA oxidation.

These results indicate that many North-East Indian plants are effective as antioxidants, possibly explaining their mode of action in hepatoprotection. These studies are proposed to be followed up with more such promising plants from North-East region and attempts will be made to isolate the active principle responsible for their antioxidant effects.

## 2. Biomarkers of genotoxicity : Antimutagenicity of heated garlic

Phytochemicals of plant origin exhibit good antimutagenic/anticarcinogenic properties. Many antimutagens are considered to be chemopreventive agents and modulate key steps in carcinogenic processes. Our earlier studies demonstrated that garlic feeding through diet, in rats can exert antimutagenic effects when exposed to Benzo(a)Pyrene [B(a)P] (Ann.Rep. 1996). Since garlic is subjected to high temperature during cooking, boiled garlic was tested for its antimutagenicity against Benzo(a)Pyrene using *S.typhimurium* TA 98 as test organism.

The Ames Salmonella test was adopted for detecting mutagenicity. Overnight culture tester strain TA 98 was treated with B(a)P at 0.4, 1 and 2 mg concentrations in the presence and absence of boiled and unboiled garlic added at 1.5, 3 and 7.5 mg levels. The reaction mixture was plated on minimal glucose agar medium plates and the number of revertants were counted after 48 hours of incubation at 37°C.

The experiments were conducted thrice and the means were calculated.

The results can be summarised as follows :

1. B(a)P tested alone showed dose dependent increase in mean number of revertants.
2. Unboiled and boiled garlic at 1.5, 3 and 7.5 mg levels showed significant antimutagenic response against B(a)P at all levels ( $P < 0.05$ ) (**Fig.17**).
3. There was no difference between the boiled and unboiled garlic.

These results indicate that the antimutagenic principle in garlic is not destroyed on

heat treatment during cooking process.

### 3. Evaluation of antioxidant micronutrients and GST-m in normals, smokers and oral cancer patients

The etiological factors associated with the development of oral cancer include, use of tobacco and cigarette/beedi-smoking. Recent epidemiological and experimental evidences suggest that certain types of cancers are most often related to micronutrient deficiencies. This study was undertaken with a view to study the antioxidant micronutrient status in oral cancer patients and compare it with that of normals and smokers. Study consisted of normals who did not have the habit of either smoking or alcohol consumption whereas smokers group had in addition to smoking, alcohol consumption.

The oral cancer patients were recruited from MNJ Cancer Hospital, Hyderabad. The blood samples were drawn from all the subjects and analysed for vitamins A, C and E,  $\beta$ -carotene and MDA in plasma, whereas selenium and GST-m were analysed in erythrocytes and lymphocytes respectively.

The results indicate the following:

1. GST-m, one of the polymorphic forms of GSTs, was significantly low in smokers as well as cancer patients as reported earlier. Since this enzyme plays a key role in the detoxification of polycyclic aromatic hydrocarbons (PAHs) metabolites, the smokers may be at risk in developing cancer whose GST-m is low and is further aggravated by poor antioxidant status.

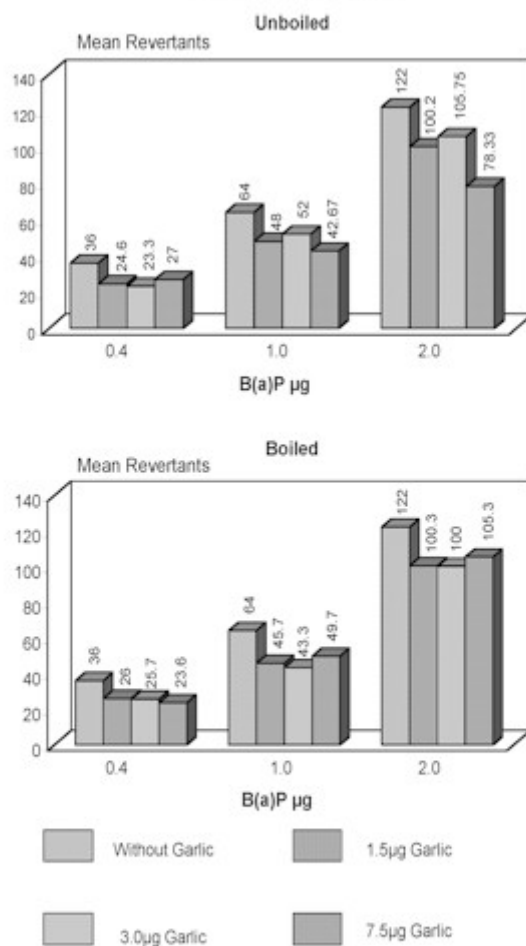
Table - 1

#### Evaluation of Antioxidant Micronutrients and GST-m in Normals, Smokers and Oral Cancer Patients

Groups	Vit A mg %	Vit E mg %	$\beta$ -Carotene mg %	Vit C mg %	MDA nmol/ ml plasma	Red cell Se mg/ml RBC	GST-m pmol/min/ mg
Normals	33.76 $\pm$	0.68 $\pm$	8.06 <sup>a</sup> $\pm$	1.0 $\pm$	3.64 $\pm$	155.4 $\pm$	
(Non-Smk)	2.95	0.10	1.76	0.03	0.08		

4.03 Non-Alc) (8)	5.85						
Smokers 3.70 <sup>b</sup> ± (6) 8.99	38.4±	1.23±	2.72 <sup>b</sup> ±	1.25±	3.42±	145.0±	
	3.06	0.12	0.17	0.11	0.18		
	0.77						
Oral	26.12±	0.47±	1.28 <sup>b</sup> ±	0.90±	3.22±	145.3±	6.84 <sup>b</sup> ±

Fig. 17  
Antimutagenesis of Garlic



Values are Mean Revertants in TA-98

Without garlic group is significantly different from others (P<0.05)

cancers	2.49	0.01	0.15	0.03	0.02
4.06	1.27				
(12)					

Values are : Mean  $\pm$  SE; No. of subjects is indicated in paranthesis;  
Values bearing different superscripts are significantly different from each other (P<0.05).

2. The micronutrient levels i.e., vitamins A, C and E were low in cancer patients compared to the rest of the groups whereas  $\beta$ -carotene was significantly low in smokers/ cancer patients compared to controls (**Table 1**).

3. There is no change in red cell selenium, and MDA levels.

This study provides information for dietary intervention of antioxidant micronutrients for cancer prevention who fall in high risk group.

## **X. NATIONAL CENTRE FOR LABORATORY ANIMAL SCIENCES (NCLAS)**

### **A. SERVICE ACTIVITIES**

#### **1. Breeding and Supply of Animals**

During the year, 45,466 animals of various species were bred (23,868 in the conventional colony and 21,598 in the barrier-maintained colony) and this is 29% lower than the corresponding period last year. Supply of animals also decreased by 23% i.e., 43,106 animals during this year when compared to 56,058 animals supplied during last year. This was due to a delay in obtaining the registration with CPCSEA. During three months (July to September, 1999), the breeding and supply of animals were suspended. The sale value of animals supplied was Rs.24.86 lakhs.

#### **2. Animal Feeds**

During the above period, 9810 kgs of rat feed and 2473 kg of rabbit feed were supplied to both Government and pharmaceutical concerns amounting to Rs.5.39 lakhs. During the same period in the previous year, a total of 12,095 kgs of feed was supplied and there was a marginal increase of 1.5% in the feed supply as compared to the drop in supply of animals during the year. Feed supply was continued even while the animal supply was suspended during the period between July and September 1999.

#### **3. Blood and blood products**

During this year, 1600 ml blood was supplied to 7 different institutions on 34 occasions and the sale value was Rs.21,500/-. Besides this, 170 ml of blood and sera was supplied to the Institute on eight different occasions.

#### **4. Health Monitoring**

Samples were collected from different species - rats - 25; mice - 81; rabbits - 17; guinea pigs - 25; hamsters - 6 and monkeys - 68; totalling 222 samples. These animals were from either isolators, barrier maintained or conventional colonies. Other materials (49) like diet (13), water (13), bedding (17) and personnel (6) were also screened during the period. All the samples were subjected to both microbiological and parasitological screening.

Microbiological screening showed the presence of *E.coli*, *Klebsiella pneumoniae*, *Proteus mirabilis* and *Listeria monocytogenes* in all the colonies. Presence of *Corynebacterium kutscheri* was observed in guinea pig colony during the period. None of the animals showed clinical symptoms of any disease.

Parasitological examination showed the presence of ectoparasite *Mycopetes musculus* and the endoparasites *Aspicularis tetraptera* and *Syphasia obvelata* in mice colony. While the endoparasites did not produce clinical symptoms, the ectoparasite infestation was heavy and caused skin changes in BALB/c mice colony. Steps were taken immediately to eradicate the mite infestation.

As part of the preclinical toxicological evaluation of  $\alpha$ -interferon, 66 monkeys were obtained from the wild and were quarantined. During this period, the animals were screened for tuberculosis, Hepatitis B (by both ELISA and conventional methods) ecto and endo parasites, and faecal samples were examined for Salmonella and Shigella. While organisms like *E.coli*, *Klebsiella pneumoniae* and *Proteus vulgaris* were isolated, Shigella was not present. The animals were treated with antiparasitic drugs and after ensuring that the animals were free of any disease, they were utilised for experimentation.

#### **5. Human Resource Development**

The 32nd Annual Laboratory Animal Technicians' Training Course was conducted for 6 weeks and seven participants successfully completed the course. Adhoc training and orientation to Laboratory Animal Science was provided to students of Medical Laboratory Technician Course, and Post-Graduate students in Nutrition from Acharya NG Ranga Agricultural University, Hyderabad.

#### **6. Technical Consultancy**

Several queries on availability of different strains of animals, husbandry, care and maintenance of animals were answered. Steps were taken to make animal users in the mailing list of the facility aware of the provisions of Breeding of and Experimentation on Animals (Control and Supervision) Rules, 1998, through correspondence. They were provided with the gazette notification and the necessary application for registration. Books on Laboratory Animal Care published by WHO were mailed to all the Institutes.

## INSTRUMENTATION SERVICES

### List of Equipment Installed

- ~~1. Varian Model AA220 Atomic Absorption Spectrophotometer~~
2. Varian Model Cary 100 Double Beam Spectrophotometer
- ~~3. Eric Jaeger Exercise Test Monitoring System consisting of :~~
  - (a) Oxycon Pro Ergo spirometric system
  - (b) Tread Mill Model LE 200 CE and
  - (c) Personal Computer & Printer
4. Halogic Model QDR 4500W Dual Energy X-ray Absorptiometer
5. Em-Scan Inc. Model SA 3000 Body Composition Analyser (TOBEC)
6. Interface Design ADAM upgrade for Beckman DC Plasma Emission Spectrometer
7. Systech Instruments Model Portamatic-2 O<sub>2</sub> and CO<sub>2</sub> Analyser
8. AVIV Biomedical Inc. Model 206D Hematofluorimeter
9. Unicam Model Helios Gamma Spectrophotometer
10. Gerhardt Model Turbotherm TT625 & Model Vapodest 20 - Digestion and Distillation System
11. Med Image Technologies Image Analysis System
12. Usha Informatics Model Coral-240 EPABX System
13. Eppendorf Model 5301 Volume Concentrator
14. Shimadzu Model 10XL Fluorescence Detector for HPLC
15. Sartorius Model BL-600 Balance, Capacity 600 g
16. Conweigh Model CLT-1002 Balance, Capacity 1 kg.
17. Harvard Apparatus Rat Tail Blood Pressure Monitoring System
18. Broviga Horizontal Submarine Electrophoresis Apparatus
19. Genie Vertical Slab Gel Electrophoresis Apparatus
20. Biorad Model Mini Protein 3-Cell Electrophoresis Apparatus
21. Biorad Model Mini Trans Blot Electrophoretic Transfer Cell
- ~~22. Udy Cat. No. 3010-019 Sample Cyclone Mill~~
23. Tritronics UPS System, 5 KVA capacity
- ~~24. Electronics & Controls UPS System, Capacity 3 KVA~~
25. Integrated Instruments & Systems UPS Systems, Capacity 500VA - 5 Nos.
26. Cintex Cat. No. CIC-84 Block Digester
27. Thermal Instruments & Equipments Digital Temperature Indicator for under water weighing system
28. York Scientific Model YSI-472 Tissue Homogenizer
29. Labline Cat. No.600 Slide Warmer
30. Holtain Instruments Skin Fold Calipers - 12 Nos.
31. Kinderman Model ECO 24 Overhead Projectors - 2 Nos.
32. Exhibitor Slide Projector
33. Kodak 500 Ektalite Automatic Slide Projector

Research Scholar/ Staff	Title of the project	Guide
34.	MJ Research Inc., Model PTC-150 Thermal Cyclor	
35.	Heraeus Muffle Furnace M110	
36.	Telephones: 84 Nos	

## II. Training Programmes

Inplant training programmes were organised for a period of six weeks to two batches of candidates deputed by Advanced Training Institute, Ramanthapur, Hyderabad, working under the aegis of Ministry of Labour, Govt. of India. One batch was undergoing a course on Process Instrumentation at A.T.I. and the other on Medical Electronics. The inplant training was a part of these courses. The Ministry of Labour, Govt. of India, considers the training as a nation building activity.

## III. Deputation

Mr. B. Ramulu, Technical Officer, Instrumentation Division was deputed twice as external examiner to Shramika Vidyapeeth, Hyderabad. He was also deputed to attend a seminar on Conversion of Freon 12 Gas into Halocarbon based refrigerants.

## IV. Audio-Visual Arrangements

1. Seminar Clubs
2. Journal Clubs
3. National Science Day Celebrations
4. Meeting of Officers-in-Charge of NNMB State Units
5. Pre-SAC & SAC meetings for the Basic Studies Group
6. National Nutrition Week Celebrations
7. World Food Day Celebrations
8. Seminar organised by Indian Pharmaceutical Association
9. Indian Radical Humanist Association (3-days)
10. Dept. of Science & Technology Meeting (2 days)
11. Brain Storming Session of Research Projects
12. Workshop on Tribal Health & Nutrition ( 2 days)
13. Video Projection & PC for IEC Workshop (5 days)
14. Workshop organised by National Academy of Agricultural Sciences (2 days)
15. The staff of the department also provided audio-visual arrangements for various training programmes of the Institute.

## V. Extension of Repair and Maintenance Services

Dr. R. Subramanian, Asst. Director, Instrumentation Division, was deputed to RMRC, Port Blair to attend to the repairs of Beckman Model 600 Spectrophotometer and ELISA

Research Scholar/ Staff	Title of the project	Guide
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Washer. He was also deputed to the Institute of Pathology, New Delhi to attend to the problems relating to the Direct Current Plasma Emission Spectrophotometer, Spetraspan V Beckman.

#### **VI. Service Calls attended**

- |                                              |   |     |
|----------------------------------------------|---|-----|
| 1. Electronics Equipment                     | : | 379 |
| 2. Electro-Mechanical Equipment              | : | 220 |
| 3. Refrigeration & Airconditioning equipment | : | 150 |
| 4. Winding Jobs                              | : | 64  |

5. Total No. of service calls attended : 813

## LIBRARY AND DOCUMENTATION SERVICES

Library continued to provide information, documentation and reprography support to the Institute's staff as well as to the scientists of other research organisations and universities.

INTERNET based information services have been introduced and various search engines such as PUBMED Com., Biomed Net Com., Infotrieve Com., etc., have been regularly accessed for retrieving data and information required for the scientists. Shef-access search engine was frequently accessed and various biomedical bibliographies with abstracts have been retrieved for the usage of researchers. NIN library data continues to be updated and is also made available on the INTERNET <<http://icmr.nic.in>>. Computer facilities have been further augmented with the addition of two more Pentium machines (P2), and the MEDLINE CD-ROM accessibility is made fully operational through these computers. A Pentium at "Food Toxicology Division" and another at the library counter quadrangle have been installed for scientists' use, for accessing MEDLINE and other library in-house data bases.

Inter-library loan facility, especially sharing the resources of British Library, IICT/CCMB libraries etc., has been further stepped up by making frequent library visits and by getting required books and journals on loan for the use of scientists of the Institute. Under the International Health Literature Exchange (IHLE) scheme of W.H.O., Geneva, over 42 journals have been received on complimentary/exchange basis.

A research study on "Designing and developing an Information System for Nutrition Science (NISNUTS) for India" was completed. The study involved a descriptive methodology of investigation largely confined in collecting the qualitative and purposive data from 120 nutrition scientists and 25 medical Libraries in the country. The study stressed the importance of i) conducting frequent user-education programmes; ii) provision of timely and liberalised photostat facilities, and iii) augmenting the library resources by continuous resource sharing and networking programmes.

The Library services were expanded by the addition of the following:

### 1. New Additions

Books	...	...	...	...	104
Reports	...	...	...	...	502
Reprints	...	...	...	...	473
Theses and Dissertations	...	...	...	...	5
Journals (New Subscriptions)	...	...	...	...	10
Microfiches	...	...	...	...	296

Slides	...	...	...	...	154
CDROMS of MEDLINE	...	...	...	...	28
Current Contents (LS) on Diskettes	...	...	...	...	104

## 2. Other Activities

Journals Bound	...	...	...	...	412
Visitors using the library	...	...	...	...	4868
Circulation of Books, Journals etc.	...	...	...	...	7422
CDROM MEDLINE Abstracts provided	...	...	...	...	303
No.of E-mails sent/received	...	...	...	...	2996
Photocopying (No.of Pages)	...	...	...	...	279636
No.of Annual Reports mailed	...	...	...	...	496
No.of Reprints mailed	...	...	...	...	498
No.of Books/Journals received on Inter-library loan	...	...	...	...	380
No.of Documentation Bulletins compiled	...	...	...	...	5
No.of INTERNET Searches provided	...	...	...	...	55

## 3. Total Library Collections

Books	...	...	...	...	14734
Journals (Bound Vols)	...	...	...	...	25426
Journals Subscribed for 1999	...	...	...	...	200
Journals received on gratis/Exchange	...	...	...	...	280
Microfiches	...	...	...	...	863
Slides	...	...	...	...	277
Reports	...	...	...	...	9495
Reprints	...	...	...	...	306598
Theses & Dissertations	...	...	...	...	329
MEDLINE CDROM Discs (1982-C)	...	...	...	...	92
Current Contents (LS) on Diskettes	...	...	...	...	446

## 4. Bibliographies Compiled

- a) Current Awareness List : List of New Reprints received through Current Contents (Quarterly)
- b) Current Information Services :
  - i) List of NIN Scientific Papers sent for Publication (Quarterly)
  - ii) List of NIN Scientific Papers Published (Quarterly)
  - iii) List of Subscription Journals received (daily chart)
- c) List of NIN Scientific Publications, 1999
- d) A Guide to NIN Library and Information Services, 1999.
- e) A Select List of Nutrition Research Institutes in India, 1999.

## Ph.D. PROGRAMMES

#### A. Ph.D. Awardees

Research Scholar/ Staff	University	Year	Title of the thesis
1. Ayesha Ismail	Osmania	1999	Studies on interaction between vitamin D and carbohydrate metabolism in diabetes
2. Sunita, Y.	Osmania	1999	Studies on blood-brain barrier : Effect of iodine status
3. Sampathachary, K.	Osmania	1999	Design and development of a National Information System in Nutrition, Science for India (NISNUTS) : A Study
4. Sujatha, T.	Sri Padmavati Mahila	1999	Studies on energy expenditure patterns of women in Hyderabad
5. Uma Nayak, M.	Sri Padmavati Mahila	1999	Social marketing strategies for nutrition education
6. Sangeetha	Osmania	1999	Effect of infection on riboflavin metabolism

#### B. Research Scholars Registered for Ph.D.

Research Scholar/ Staff	Title of the project	Guide
1. Abhay Kumar (1988)	Nutritional aspects of selenium	Dr. Kamala Krishnaswamy
2. Gal Reddy, Ch. (1993)	Influence of child labour on socioeconomic and nutritional status	Dr. Abbasayulu, YB.
3. Anil Kumar Dube (1994)	Nutritional education for urban adolescents : Use of social marketing principles in communication	Dr. Mohan Ram, M.
4. Harishankar, N. (1994)	Characterization of obese mutant strain from Wistar/NIN rats	Dr. Giridharan, N.V.
5. Mallikarjuna Rao, K. (1994)	Nutrition profile and household food security among rural population in different	Dr. Hanumantha Rao, D.

regions of Andhra Pradesh

- |                                    |                                                                                                                                      |                                   |
|------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------|
| 6. Pulkit Mathur<br>(1994)         | Risk analysis of food adulterants                                                                                                    | Dr.Ramesh V Bhat                  |
| 7. Ramulu, P.<br>(1994)            | Studies on dietary fiber content of Indian foods and its physiological effects                                                       | Dr.Udayasekhara Rao, P.           |
| 8. Chennaiah, S.<br>(1995)         | Isolation and characterization of calcinogenic principle from <i>Solanum melogena</i> leaves                                         | Dr. Raghuramulu, N.               |
| 9. Hemalatha, S.<br>(1995)         | Biochemical and metabolic studies with sesame lignans                                                                                | Dr. Ghafoorunissa                 |
| 10. Radhika, M.S.<br>(1998)        | Effect of food-based vitamin A supplementation during pregnancy on maternal and child health                                         | Dr.P.Bhaskaram                    |
| 11. Rajendraprasad, M.P.<br>(1998) | Nitrosamines and its relevance to cancer in India                                                                                    | Dr.Kamala Krishnaswamy            |
| 12. Raghu, P.<br>(1999)            | Characterization and significance of TTR <sub>2</sub>                                                                                | Dr.B.Sivakumar                    |
| 13. Nirmala, K.<br>(1999)          | Plant constituents as chemopreventive agents                                                                                         | Dr. Kalpagam Polasa               |
| 14. Sarvanan, N.<br>(2000)         | Effects of dietary alteration of n-6 and n-3 polyunsaturated fatty acids on insulin resistance, structure and function of adipocytes | Dr.Ghafoorunissa                  |
| 15. Sreedhar, B.<br>(2000)         | Iron and zinc interactions at the site of absorption                                                                                 | Dr.K.Madhavan Nair                |
| 16. Jayakumar, S.M.<br>(2000)      | Studies on food intake regulation and obesity in WNIN/Ob and WNIN/GR-Ob rats                                                         | Dr.A.Vajreswari                   |
| 17. Vijayalakshmi, A.<br>(2000)    | Role of nutrition in modification of apoptosis                                                                                       | Dr.M.Raghunath/<br>Dr.B.Sesikeran |

## **AWARDS/HONOURS CONFERRED ON SCIENTISTS**

### **1. Dr.Kamala Krishnaswamy**

Basant Devi Amir Chand Prize of ICMR for the year 1997.

The Bires Chandra Guha Memorial Oration (1999) by Indian National Science Academy, New Delhi.

Knoll (Boots) Oration (1999) by Indian Society of Gastroenterology, Lucknow.

Kamala Puri Sabharwal Memorial Oration (1999) by Lady Irwin College, New Delhi.

Shakuntala Das Gupta Oration (2000) by the Physiological Society of India, Calcutta.

Glaxo Oration (2000) by National Academy of Medical Sciences, New Delhi.

### **2. Dr.P. Bhaskaram**

Fellow, A.P. Akademi of Sciences, Hyderabad.

### **3. K.Madhavan Nair**

BGRC Silver Jubilee Oration Award of ICMR for the year 1996.

### **4. M.S.Radhika**

Young Scientists' Junior Award in the Experimental Nutrition by the Nutrition Society of India, presented at Annual Meeting held at Coimbatore.

### **5. Sujatha, T, Shatrugna, V., Venkataramana, Y. and Nazeema, B.**

Certificate of honour in recognition of their poster presentation on "Time and Energy Expenditure Patterns of Women and Low Socio-Economic Group from India, at the 2nd International Congress "Women Work Health", Rio-de-Janeiro, Brazil, 1999.

**PARTICIPATION OF SCIENTISTS IN  
INTERNATIONAL MEETINGS/CONFERENCES**

**1. Dr. Kamala Krishnaswamy**  
*Director*

Indo-European Symposium on "Micronutrients, Maternal and Child Health" at Goa (April 25, 1999).

WHO Fifth Meeting of "South-East Asia Nutrition-Cum-Action Network" at the Institute of Nutrition, Mahidol University, Thailand (June 1, 1999).

Consultative Meeting on "World Bank Activities in Health Sector in India" at the World

Bank, New Delhi (Aug.9, 1999).

VIII Asian Congress of Nutrition at Seoul, Korea (Aug.29, 1999).

International Conference on “Heart Health in Developing Countries: An agenda for action in the 21st Century”, at New Delhi (Oct.10, 1999).

International Conference on “Biotechnology for sustained productivity in Agriculture”, at Hyderabad (Nov. 1, 1999)

International Seminar on “Anaemia in South Asia” at Kathmandu (Dec. 2, 1999).

WHO-SEARO Inter-Country Workshop to review implementation on “National Plan of Action for Nutrition”, at New Delhi (Dec.8, 1999).

XV Asia Pacific Conference on Cancer at Chennai (Dec.12, 1999).

Indo-US Workshop on “Health and Nutrition of Women, Infants and Children”, at Hyderabad (February 10-12, 2000).

**2. Dr.B.Sivakumar**  
***Deputy Director (Sr.Gr.)***

Indo-European Symposium on “Micronutrients, Maternal and Child Health” at Goa (April 25, 1999).

International Conference on “Biotechnology for sustained productivity in Agriculture” at Hyderabad (Nov.1, 1999).

## SCIENTIFIC PUBLICATIONS

### A. PAPERS PUBLISHED IN SCIENTIFIC JOURNALS

1. Anuradha, P., Yashoda Devi, P., Shiva Prakash, M. : Effect of handwashing agents on bacterial contamination. Indian J. Pediatr. 66(1):7-10, 1999.
2. Ashok, A., Sunita Rao, D., Chennaiah, S., Raghuramulu, N. : Vitamin D<sub>2</sub> is not biologically active for Rora (*Labeo rohita*) as vitamin D<sub>3</sub>. J. Nutr. Sci. Vitaminol. 45(1):21-30, 1999.
3. Bala, T.S.S., Raghunath, M. : Severe gestational hypothyroidism increases BBB nutrient transport in the off spring. Nutritional Neurosci. 2(2):75-83, 1999.
4. Bala, T.S.S., Rupalatha, M., Raghunath, M. : Probable basis of altered BBB nutrient transport in the offspring of severely hypothyroid dams. Nutritional Neurosci. 2(2): 85-91, 1999.
5. Bhanuprakash Reddy, G., Bhat, K.S. : Protection against UVB inactivation (*in vitro*) of rat lens enzymes by natural antioxidants. Mol. Cell. Biochem. 194(1-2):41-45, 1999.
6. Bhaskaram, P. : Nutritional modulation of immunity in response to vaccines (Measles and Tuberculosis as examples). Proc. Nutrition Soc. India. 46: 64-72, 1999.
7. Bhaskaram, P., Balakrishna, N., Goud, B.N., Sukanya, M : Post-vaccination scenario of measles : A retrospective analysis. National Med. J. India. 12(3): 111-112, 1999.
8. Ghafoorunissa : Antiatherogenic potential of oils in Indian subjects consuming cereal based diets. Proc. Nutrition Soc. India. 46: 33-46, 1999.
9. Hemalatha, R., Vijayalakshmi, P., Gyaneshwari, Rama Rao, M.V., Ramani, A. : Multidrug resistant salmonella typhi in Hyderabad. Indian J. Med. Microbiol. 17(1):39-41, 1999.
10. Kamala Krishnaswamy : Turmeric - A source of phytochemicals in Indian diets. Proc. Nutrition Soc. India. 46: 73-76, 1999.
11. Kamala Krishnaswamy, Neelam : Chemoprevention. Indian J. Nutr. Dietet. 36(5):244-254, 1999.
12. Kamala Krishnaswamy., Prasad, M.P.R. : Diet and cancer in India. Internat. J Gastroenterol. 4(3) : 5-8, 1999.

13. Kanjilal, S., Prasad, R.B.N., Kaimal, T.N.B., Ghafoorunissa., Rao, S.H. : Synthesis and estimation of calorific value of a structured lipid-potential reduced calorie fat. *Lipids*. 34(10):1045-1055, 1999.
14. Laxmaiah, A., Sarma, K.V.R., Rao, D.H., Gal Reddy, Ch., Ravindranath, M., Rao, M.V., Vijayaraghavan, K. : Impact of mid-day meal programme on educational and nutritional status of school children in Karnataka. *Indian Pediatr*. 36(12):1221-1228, 1999.
15. Mujeebur Rahman., Viqarunnissa, Visweswara Rao, K.: Weighment method of diet survey - A case study of reference periods by economic status. *Indian J. Nutr. Dietet*. 36(2):505-520, 1999.
16. Murthy, S.N., Janardana Sarma, M.K. : Identification of alpha-aminoacid/L-lysine, alpha-amino oxidase in mouse brain. *Mol. Cell. Biochem*. 197(1):13-23, 1999.
17. Rajeshwar Rao, M., Balakrishna, N., Visweswara Rao, K. : Suitability of CANSCORE for the assessment of the nutritional status of New borns. *Indian J Pediatr*. 66(4):483-492, 1999.
18. Rupalatha, M., Srinivasa Rao, P. : Activities of synaptosomal membrane-bound enzymes in response to dietary alterations in linoleic and alpha-linolenic acid. *J. Clin. Biochem. Nutr*. 26(3):201-211, 1999.
19. Saibaba, A., Sarma, DS., Balakrishna, N., Raghuram, TC: Utilization of IEC by middle level health personnel in the implementation of National Nutrition Programmes. *Indian J Public Health*. 24(2):75-85, 1999.
20. Sangeetha, B., Lakshmi, AV. : Tissue distribution and turnover of (3H) riboflavin during respiratory infection of mice. *Metabolism*. 48(12):1608-1611, 1999
21. Sangeetha, B., Lakshmi, AV., Bamji, MS : Mitochondrial oxidative metabolism during respiratory infection in riboflavin deficient mice. *J Nutritional Biochem*. 10(12):728-732, 1999.
22. Shahnaz Vazir, Kashinath, K. : Influence of the ICDS on psychosocial development of rural children in Southern India. *J. Indian Academy of Applied Psychology*. 25(1-2): 11-24, 1999.
23. Sivakumar, B. : Protein turnover studies using stable Isotope tracer techniques. *Proc. Nutrition Soc. India*. 46: 77-86, 1999.
24. Smitha Gheye., Lakshmi,AV., Krishna, TP., Kamala Krishnaswamy : Fibrinogen and homocysteine levels in coronary artery disease. *Indian Heart Journal*. 51(5): 499-502, 1999

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27. Sunita Rao, D., Raghuramulu, N. : Is vitamin D redundant in an aquatic habitat. *J. Nutr. Sci. Vitaminol.* 45(1):1-8, 1999.
28. Sunita Rao, D., Raghuramulu, N. : Vitamin D<sub>3</sub> and its metabolites have no role in calcium and phosphorus metabolism in *Tilapia mossambica*. *J. Nutr. Sci. Vitaminol.* 45(1):9-19, 1999.
29. Veena Shatrugna, Leela Raman, Uma Kailash, Balakrishna, N., Visweswara Rao, K. : Effect of dose and formation on iron tolerance in pregnancy. *National Med. J. India.* 12(1):18-20, 1999.
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31. Vijayaraghavan, K., Kamala Krishnaswamy : Effect of supplementation with vitamin A or beta-carotene on mortality to pregnancy. *Brit Med. J.* 319(7218):1201-1202, 1999.
32. Visweswara Rao, K., Balakrishna, N., Leela Raman : Nutritional status of new borns and the associated factors. *Man in India.* 79(1-2):101-109, 1999.

#### **B. PAPERS PUBLISHED IN PROCEEDINGS OF WORKSHOPS/CONFERENCES**

1. Babu, S. : Nutritional value of mustard oil. In "Mustard oil crisis : Why, how, and what next". Conference organised on 21st April 1999, by Mustard Research and Promotion Consortium, New Delhi. P.41-43, 1999.
2. Bhaskaram, P. : Immunobiology of mild-moderate micronutrient deficiency. In "Micronutrients, maternal and child health : Indo-European Symposium, 25-27 April 1999, Goa, India, organised by AIIMS, ICMR and Centre for International Health, Norway. P.5-7 & 35, 1999.

3. Bhat, R.V., Babu, S. : Food safety requirements for sustainable agricultural export in the post WTO scenario. In "Souvenir of 4th Agricultural Science Congress on Sustainable Agricultural Export, held on 21-24 February 1999, at Jaipur. P.33-37, 1999.
4. Dinesh, B., Kamala Krishnaswamy : Lead toxicity : Treatment and prevention. In "George A.M., (ed.) : Lead poisoning prevention and treatment : Implementing a national program in developing countries. Proceedings of Int. Conference, February 8-10, 1999, held at Bangalore. Bangalore, The George Foundation, P.301-303, 1999.
5. Ghafoorunissa : Fat requirements of Indians. In "Proceedings of the National seminar on Edible rice bran oil, held on 8th May 1999, at New Delhi. Mumbai, Solvent Extractors Association India. P.42-44, 1999.
6. Kamala Krishnaswamy : Importance of folate in human nutrition. In "Micronutrients maternal and child : Indo-European Symp. 25-27 April 1999, Goa, India, organised by AIIMS, ICMR, Centre for International Health, Norway. P.25, 1999.
7. Kamala Krishnaswamy : Nutrient requirements and assessment of nutritional status. In "Sainani, G.S., ed.: API Textbook of Medicine. 6th ed. Mumbai, Association of Physicians of India, P.160-164, 1999.
8. Kamala Krishnaswamy : Nutrition and risk of metal toxicity. In "George, A.M. (ed.) : Lead poisoning prevention and treatment : Implementing a national program in developing countries. Proceedings of Int. Conference, 8-10 February 1999, held at Bangalore. Bangalore, The George Foundation, P.305-306, 1999.
9. Kamala Krishnaswamy : Obesity in an urban middle class in Delhi. New Delhi, Nutrition Foundation of India, P.1-26, 1999. (NFI Sci.Report. 15).
10. Kamala Krishnaswamy., Brahmam, GNV : Double fortification of common salt with iron and iodine-Indian study. In "National Conference on Micronutrient Fortification of Food, Jaipur, 8-9 February 1999, New Delhi, ILSI-India and Micronutrient Initiative, 89-94, 1999.
11. Kamala Krishnaswamy : Rajammal P. Devadas - A penchant for perfection and excellence. In "Amma, Dr.Rajammal P. Devadas : A beacon of light. A compilation of articles in appreciation. Vol.1, Coimbatore, Daughters of Saradalaya, Avinashilingam University, P.88-89, 1999.
12. Nair, K.M. : Recent findings on iron absorption and implications on iron folate supplementation programme. In "Micronutrients, maternal and child health : Indo-European Symposium, 25-27 April 1999, Goa, organised by AIIMS, ICMR and Centre for International Health, Norway, P.20-22, 29, 1999.

13. Raghuram, T.C. : Rajammal P.Devadas - A saga of service. In "Amma, Dr.Rajammal P. Devadas - A penchant for perfection and excellence. In "Amma, Dr. Rajammal P. Devadas : A beacon of light. A compilation of articles in appreciation. Vol.1, Coimbatore, Daughters of Saradalaya, Avinashilingam University, P.106-110, 1999.
14. Sesikeran, B. : Intestinal mucosal cell apoptosis and its relevance to nutrient absorption and enteral nutrition. In "Souvenir of 5th Annual Conference of Indian Society for Parenteral and Enteral Nutrition (ISPEN), Pune. P.88-92, 1999.
15. Sivakumar,B. : Fortification of appropriate foods with iron and other micronutrients. In "Micronutrients, maternal and child health : Indo-European Symposium, 25-27 April 1999, Goa, India, organised by AIIMS, ICMR and Centre for International Health, Norway. P.30-32, 40, 1999.
16. Vijayaraghavan, K : Vitamin A deficiency - Current status and strategies for control. In " National Conference on Micronutrient Fortification of Food, Jaipur, 8-9 February 1999. New Delhi, ILSI-India and Micronutrient Initiative, 95-103, 1999.

#### **C. PAPERS PUBLISHED AS ABSTRACTS IN SELECTED JOURNALS**

1. Anitha Kumari, S., Sree Ramkumar, N., Singotam, L. : Scanning electron microscope studies on the gills of fishes collected from a polluted fresh water lake. Scanning. 21(2): 157-158, 1999.
2. Singotamu, L. : Hemin as an immuno modulator to combat anaemia due to infestation of malarial parasites. Scanning. 21(2):167-168, 1999.
3. Singotamu, L. : Scanning electron microscope studies of bread prepared by alternate method. Scanning. 21(2):166-167, 1999.
4. Singotamu, L. : Scanning electron microscope studies on milk samples of buffalo, goat, cow, sheep, ass and human. Scanning. 21(2):168-169, 1999.
5. Singotamu, L., Vasanthi, S. : Scanning electron microscope studies on seeds of sesame (Til), mustard, sunflower, groundnut, castor, cotton, coconut and oil palm. Scanning. 21(2):168, 1999.

#### **D. REPORTS FROM NIN/NNMB**

1. Report of NNMB Second Repeat Survey - Rural (1996-97), 1999.

## **E. POPULAR ARTICLES**

### **E1 NUTRITION NEWS**

1. Ayesha Ismail, Raghuramulu, N. : Vitamin D deficiency and insulin homoeostasis. Nutrition News. 20(3):1-4, 1999.
2. Bhat, R.V., Sudershan Rao, V., Kashinath, L. : HACCP approach for improvement in quality of Khoa - A case study. Nutrition News. 20(1):1-4, 1999.
3. Nair, K.M. : Iron absorption and its implications in the control of iron deficiency anemia. Nutrition News. 20(2):1-4, 1999.
4. Uma Nayak, M., Shahnaz Vazir, Vijayaraghavan, K. : Prevention of vitamin A deficiency - Social marketing approach. Nutrition News. 20(4):1-4, 1999.

### **E2 NUTRITION**

1. Anila, E.M. : Trace elements in health and disease, Pt.I. Nutrition. 33(1):23-32, 1999.
2. Anila, E.M. : Trace elements in health and disease, Pt.II. Nutrition. 33(2):26-32, 1999.
3. Anila, E.M., Jayalakshmi, P.M., Behera, U.M., Sivakumar, B., Nair, K.M. : Why food fortification? Nutrition. 33(1):12-22, 1999.
4. Babu, S., Bhat, R.V. : Know your food additives : Acidulants. Nutrition. 33(3):25-32, 1999.
5. Bhooma Mani, N., Buchi Babu Reddy, O., Devender Rao, P., Shanti Kumar Singh, L., Jagadeesan, V. : Your doctor in the kitchen. Nutrition. 33(1):3-11, 1999.
6. Brahmam, GNV : How nutritionally sound are our elderly? Nutrition. 33(4):13-21, 1999.
7. Dube, AK : The angst of ageing. Nutrition. 33(4):22-33, 1999.
8. Hemalatha, R: Ageing and immunity. Nutrition. 33(4):7-12, 1999.
9. Hemalatha, R. : Diet therapy in classical phenylketonuria. Nutrition. 33(2):11-18, 1999.
10. Indira Ravindranath : Fish as food. Nutrition. 33(3): 16-24, 1999.
11. NageswaraRao, R. : Banana - The God's gift to mankind. Nutrition. 33(2):19-25, 1999.

12. Reddy, C.V.K. : Greens for good health. *Nutrition*. 33(3):3-8, 1999.
13. Rita Saxena : How green is your diet. *Nutrition*. 33(3): 9-15, 1999.
14. Sreeramulu, D., Raghuramulu, N : Nutrition and ageing. *Nutrition*. 33(4):3-6, 1999.
15. Suryakumari, M.V.L., Venkataramana, Y. : Science in Sports - An overview. *Nutrition*. 33(2):3-10, 1999.

### **E3 OTHERS**

1. Bhat, RV., Geetha, T : Vegetarianism sensu stricto:the way ahead. *Indian Vegetarian Congress Quarterly*. 38(3):30-36, 1999.
2. Kalpagam Polasa, Kamala Krishnaswamy : Alliums as food for health life. *ICMR Bull*. 29(4-5):43-49, 1999.
3. Kamala Krishnaswamy : Bias blocks female health promotion in India. *Monitor (Spring)*. : P.4, 1999.
4. Neelam, Uday Kumar, P., Kaladhar, M. : Risk of Aluminium toxicity in the Indian context. *ICMR Bull*. 29(8):86-90, 1999.
5. Sivakumar, B. : Protein turnover studies using stable isotopes. *ICMR Bull*.29(3):33-38, 1999.
6. Vijayalakshmi, T., Hemalatha, R. : Those offending foods - 1. *Health Action*. 12(10):25-26, 1999.
7. Vijayalakshmi, T., Hemalatha, R. : Those offending foods - 2 : Types of immunological reactions in food allergy. *Health Action*. 12(11):55-56, 1999.
8. Vijayalakshmi, T., Kamala Krishnaswamy : Obesity. *Health Action*. 12(6):21-22, 1999.
9. Vijayalakshmi, T., Kamala Krishnaswamy : Obesity. *Health Action*. 12(7):21-23, 1999.

### **E4 BOOK REVIEWS**

1. Nair, K.M. : Annual review of nutrition, 1998, ed. by McCormick, D.B., Bier, D.M. and Goodridge, A.G. Palo Alto, Annual Reviews Inc., 1998. \$.65.00 [Reviewed in *Current Science*. 76(5):698-699, 1999].
2. Sesikeran, B. : A historical dictionary of Indian food, ed. by K.T.Achaya. Oxford University Press, Delhi, 1998. Rs.525=00. Reviewed in *Indian J. Med. Res*. 109(5):195,1999.

- Indo-US Workshop on "Health and Nutrition of Women, Infants and Children", at Hyderabad (February 10-12, 2000).
3. **Dr. Ghafoorunissa**  
*Dy. Director (Sr. Gr.)*  
  
International Conference & Exhibition on "Oilseeds and vegetable oil processing", at New Delhi (February 19-21, 1999).  
  
VIII Asian Congress of Nutrition at Seoul, Korea (Aug. 29, 1999).  
  
International Conference on "Heart Health in Developing Countries: An agenda for action in the 21st Century", at New Delhi (Oct. 10, 1999).
  4. **Dr. P. Bhaskaram**  
*Deputy Director (Sr.Gr.)*  
  
Indo-European Symposium on "Micro-nutrients, Maternal and Child Health" at Goa (April 25, 1999).  
  
Indo-US Workshop on "Health and Nutrition of Women, Infants and Children", at Hyderabad (February 10-12, 2000).
  5. **Dr.K.Vijayaraghavan**  
*Deputy Director (Sr.Gr.)*  
  
Indo-US Workshop on "Health and Nutrition of Women, Infants and Children", at Hyderabad (February 10-12, 2000).
  6. **Dr. Ramesh V Bhat**  
*Deputy Director (Sr.Gr.)*  
  
WHO Task Group meeting on Folic Acid, at Geneva (May 10, 1999).  
  
XXXII Session of the Codex Committee on Food Additives and Contaminants, at Beijing, China (March 20, 2000).
  7. **Dr. N. Raghuramulu**  
*Deputy Director (Sr.Gr.)*  
  
First International Conference on "Chemistry & Biology of Vitamin D Analogs" at Providence, Rhode Island, USA (Sept. 26, 1999).
  8. **Dr. L. Singotamu**  
*Deputy Director*  
  
"Scanning 99" at Chicago, Illinois, USA. (April 11, 1999).
  9. **Dr. P. Udayasekhara Rao**  
*Deputy Director*  
  
III International Food Data Conference, organised by FAO, IUNS and UN University at Rome, Italy (July 5, 1999).
  10. **Dr. Veena Shatrugna**  
*Deputy Director*  
  
2nd International Congress on "Women Work Health" at Rio-de-Janeiro, Brazil (Sept. 19-22, 1999).
  11. **Dr.Kalpagam Polasa**  
*Assistant Director*  
  
International Congress on Frontiers in Pharmacology and Therapeutics in 21st Century, New Delhi (Dec. 1-4, 1999).
  12. **Dr.Shahnaz Vazir**  
*Senior Research Officer*  
  
Indo-US Workshop on "Health and Nutrition of Women, Infants and Children", at Hyderabad (February 10-12, 2000).
  13. **Dr. K. Madhavan Nair**

**Senior Research Officer**

Indo-European Symposium on "Micro-nutrients, Maternal and Child Health" at Goa (April 25, 1999).

**14. Dr.M.P.Rajendra Prasad**  
**Senior Research Officer**

International Congress on Frontiers in Pharmacology and Therapeutics in 21st Century, New Delhi (Dec. 1-4, 1999).

**15. Dr.B.Dinesh Kumar**  
**Research Officer**

International Congress on Frontiers in Pharmacology and Therapeutics in 21st Century, New Delhi (Dec. 1-4, 1999).

**16. Dr. T. Sujatha**  
**Technical Assistant**

2nd International Congress on "Women

Work Health" at Rio-de-Janeiro, Brazil (Sept. 19-22, 1999).

**17. Dr. K. Bhaskarachary**  
**Technician**

XII International Symposium on "Carotenoids", organised by International Carotenoid Society" at Cairns, Australia (July 13-18, 1999).

**WORKSHOPS/CONFERENCES/  
SEMINARS/TRAINING  
PROGRAMMES  
HELD AT NIN**

1. A two-day meeting jointly sponsored by NIN/Micronutrient Initiative (Canada) to discuss the issues related to food fortification, especially double fortified salt (May 24-25, 1999).

2. A one day Symposium on "Bone Health" (June 7, 1999).

3. Meetings of the Pre-SAC and Scientific Advisory Committee of NIN/FDTRC/NCLAS (June 26-28, 1999).
4. Meeting of the Steering Committee of NNMB (June 28, 1999).
5. 32nd M.Sc. (Applied Nutrition) Course (15th June 1999 - 15th March 2000).
6. Adhoc Training Programme on Nutrition for Mr.D.R.Adhikari, WHO Fellow, from the Department of Health Services, Child Health Division, Kathmandu, Nepal (July 1-29, 1999).
7. Orientation Training Course on Nutrition for officials of Food and Nutrition Board (July 12-23, 1999).
8. XXVIII Annual Training Course on Endocrinological Techniques and Their Applications (2nd August to 17th September 1999).
9. Meeting of the Steering Committee of the National Nutrition Monitoring Bureau (Aug. 19, 1999).
10. National Workshop on "Tribal Health and Nutrition", sponsored by the World Health Organisation (WHO), New Delhi (Aug.26-27, 1999).
11. Department of Science & Technology Project Advisory Council Meeting on Health Sciences (Sept. 14-15, 1999).
12. Meeting of the Steering Committee of National Nutritional Monitoring Bureau (Sept.16, 1999).
13. Prof. P. K. Devi Memorial Oration, organised in association with Bharatiya Vidya Bhavan, Hyderabad (Sept. 26, 1999).
14. Orientation Training Course for officials of Food and Nutrition Board (Sept.20-Oct.1, 1999).
15. Workshop on "Nutrition intervention in the next Millennium", jointly organised by NIN and CARE, AP (October 5-6, 1999).
16. Short-term training programme in Maternal and Child Health, for Dr.Mohammed Daud and Dr. Dinesh Chapagain, WHO Fellows from Nepal (Nov. 15-26, 1999).
17. Symposium on "Diversification of Agriculture for Human Nutrition", sponsored by National Academy of Agricultural Sciences, Hyderabad (Dec. 16-17, 1999).

18. Training Course on “Techniques for Assessment of Nutritional Anaemias” (Dec. 1-10, 1999).
19. IEC Training Workshop in Biomedical Sciences, sponsored by the ICMR, New Delhi (Dec. 20-24, 1999).
20. XXVII Post-Graduate Certificate Course in Nutrition (Jan.3 - March 15, 2000).
21. INDO-US Workshop on “Health and Nutrition of Women, Infants and Children”, sponsored by ICMR, NIN, NICHD, ODS, CDC and USAID (Feb.10-12, 2000).

### **SERVICES RENDERED TOWARDS INCOME GENERATION FOR THE INSTITUTE**

#### **A. Pathology Services**

The Pathology Department has been providing histopathology and cytology services to various Centrs such as Indian Institute of Chemical Technology, Osmania University in the twin cities. A total of 202 samples were screened and reports prepared. From this activity, an amount of Rs.88,300/- was generated.

#### **B. Salt Testing**

To ensure quality of iodised and iron fortified salt samples, the Institute has been providing technical assistance in analysis of iodine and iron content of salt samples received from various organisations. The iron content of 52 iron fortified salt samples and the iodine content of one iodized salt, which were received from the Govt. of Orissa and Brilliant Salt Industries, Karnataka, were determined and reports sent. By analysing the salt samples, Rs.5,450/- was generated.

### **SCIENTIFIC ADVISORY COMMITTEE**

1. Dr. B.N. Tandon *Chairman*  
Director  
Pushpawati Singhanian Research  
Institute for Liver, Renal & Digestive  
Diseases, New Delhi
2. Dr. N.K. Ganguly  
Director General  
Indian Council of Medical Research  
Ansari Nagar, New Delhi
3. Dr. Asha Oumachigui  
Professor & Head  
Dept. of Obstetrics & Gynaecology  
JIPMER, Pondicherry
4. Dr. Ashok Khar  
Deputy Director  
Centre for Cellular and Molecular  
Biology, Hyderabad
5. Dr. Bhan, MK  
Professor Paediatrics  
All India Institute of Medical Sciences  
Ansari Nagar, New Delhi
6. Dr. Bhisey, AN  
Director (Retd)  
Cancer Research Institute  
Tata Memorial Centre, Mumbai
6. Dr. Gopinathan, KP  
Director  
Monsanto Research Centre  
Indian Institute of Science, Bangalore
7. Dr. Gupta, CM  
Director  
Central Drug Research Institute,  
Lucknow
8. Dr. Gupta, SK  
Staff Scientist VI & Chief  
Gamete Antigen Lab  
National Institute of Immunology  
New Delhi
9. Dr. Handa, SS  
Director (Retd)  
Regional Research Laboratory  
Jammu Tawi
10. Dr. Khanna, SK  
Deputy Director & Head  
Food Toxicology Division  
Industrial Toxicology Research Centre  
Lucknow
11. Dr. Lalji Singh  
Director  
Centre for Cellular & Molecular Biology,

## SCIENTIFIC PUBLICATIONS

### A. PAPERS PUBLISHED IN SCIENTIFIC JOURNALS

1. Anuradha, P., Yashoda Devi, P., Shiva Prakash, M. : Effect of handwashing agents on bacterial contamination. Indian J. Pediatr. 66(1):7-10, 1999.
2. Ashok, A., Sunita Rao, D., Chennaiah, S., Raghuramulu, N. : Vitamin D<sub>2</sub> is not biologically active for Rora (*Labeo rohita*) as vitamin D<sub>3</sub>. J. Nutr. Sci. Vitaminol. 45(1):21-30, 1999.
3. Bala, T.S.S., Raghunath, M. : Severe gestational hypothyroidism increases BBB nutrient transport in the off spring. Nutritional Neurosci. 2(2):75-83, 1999.
4. Bala, T.S.S., Rupalatha, M., Raghunath, M. : Probable basis of altered BBB nutrient transport in the offspring of severely hypothyroid dams. Nutritional Neurosci. 2(2): 85-91, 1999.
5. Bhanuprakash Reddy, G., Bhat, K.S. : Protection against UVB inactivation (*in vitro*) of rat lens enzymes by natural antioxidants. Mol. Cell. Biochem. 194(1-2):41-45, 1999.
6. Bhaskaram, P. : Nutritional modulation of immunity in response to vaccines (Measles and Tuberculosis as examples). Proc. Nutrition Soc. India. 46: 64-72, 1999.
7. Bhaskaram, P., Balakrishna, N., Goud, B.N., Sukanya, M. : Post-vaccination scenario of measles : A retrospective analysis. National Med. J. India. 12(3): 111-112, 1999.
8. Ghafoorunissa : Antiatherogenic potential of oils in Indian subjects consuming cereal based diets. Proc. Nutrition Soc. India. 46: 33-46, 1999.
9. Hemalatha, R., Vijayalakshmi, P., Gyaneshwari, Rama Rao, M.V., Ramani, A. : Multidrug resistant salmonella typhi in Hyderabad. Indian J. Med. Microbiol. 17(1):39-41, 1999.
10. Kamala Krishnaswamy : Turmeric - A source of phytochemicals in Indian diets. Proc. Nutrition Soc. India. 46: 73-76, 1999.
11. Kamala Krishnaswamy, Neelam : Chemoprevention. Indian J. Nutr. Dietet. 36(5):244-254, 1999.
12. Kamala Krishnaswamy., Prasad, M.P.R. : Diet and cancer in India. Internat. J Gastroenterol. 4(3) : 5-8, 1999.

13. Kanjilal, S., Prasad, R.B.N., Kaimal, T.N.B., Ghafoorunissa., Rao, S.H. : Synthesis and estimation of calorific value of a structured lipid-potential reduced calorie fat. *Lipids*. 34(10):1045-1055, 1999.
14. Laxmaiah, A., Sarma, K.V.R., Rao, D.H., Gal Reddy, Ch., Ravindranath, M., Rao, M.V., Vijayaraghavan, K. : Impact of mid-day meal programme on educational and nutritional status of school children in Karnataka. *Indian Pediatr*. 36(12):1221-1228, 1999.
15. Mujeebur Rahman., Viquarunnissa, Visweswara Rao, K.: Weighment method of diet survey - A case study of reference periods by economic status. *Indian J. Nutr. Dietet*. 36(2):505-520, 1999.
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