

## HIGHLIGHTS

Nutrition related issues pertaining to different geographical regions of the country were probed in detail this year. Uttar Pradesh was surveyed this year to assess the dietary intakes and nutritional status of rural community. North-Eastern India was also the focus of research studies as certain aspects of endemic goitre were investigated and also certain medicinal plants were analyzed for their antioxidant activities. A series of studies on food fortification and trace minerals were carried out as part of basic studies. Also, this year, some new studies were conducted in the areas of cancer-xenobiotics, food safety and extension and training. In the realm of clinical studies, the correlation between women's work and bone health was worked out. Here are the highlights of the research studies carried out during the year.

### 1. COMMUNITY STUDIES

#### *1.1. Nutrition profile of Indians – A districtlevel survey in Uttar Pradesh*

Assessment of nutrition profile of the rural community in 63 districts of Uttar Pradesh revealed that the average intake at the State level of cereals & millets and pulses was satisfactory, while that of milk, sugar & jaggery was less than the recommended dietary allowances. The mean intake of all the nutrients except energy, protein and calcium was less than the RDA. Only about 64% of the households were consuming adequate amount of both energy and protein. In general, about 52% preschool children were undernourished (weight for age < Median – 2SD). The prevalence was >50% in 34 of the 63 districts surveyed. About 42% of adult males and 34% of females had chronic energy deficiency (BMI < 18.5).

### 2. CLINICAL STUDIES

#### *2.1. Women's work and bone health*

It is known that bone-loading postures, adopted during exercise, are recommended for

bone density improvement. However the stimulation of bone turnover during prolonged bone loading postures in the absence of calcium intakes might prove to be deleterious for bone health.

Earlier studies showed that despite low calcium intakes standing or standing with load bearing/bending postures are better for hip densities when compared to the sitting postures at work. However, in the case of the spine, standing and bending postures significantly reduce bone densities at the spine, but sitting postures do not worsen the spine bone densities. Using the WHO classification for diagnosis of osteoporosis, the onset of bone changes around 40 years of age in the low socio-economic group were seen in spine (-2.27), hip (-1.7), forearm (-1.53) in this order. All the women from the low socio-economic (LSE) group have increased osteoclastic activity (indicated by acid phosphatase), but those with spinal osteoporotic changes are worse off on all the biochemical indicators of bone loss.

#### *2.2. Effect of tamarind on mobilization of fluoride in fluorotic subjects*

Tamarind ingestion through diet appears to be helpful in mobilizing deposited fluoride from bone, by enhancing urinary excretion of fluoride after urinary fluoride was stabilized by giving fluoride free water.

### 3. FOOD FORTIFICATION – TECHNOLOGY DEVELOPMENT

Food fortification is one of the important strategies accepted in the National Nutrition Policy to achieve the goals of controlling anemia in the country. Institute has made considerable progress towards realizing this goal by developing new technology.

3.1. Encapsulated source of iron was found to keep iodine in a stable form in double fortified ordinary common salt while the NIN formulation, required high quality salt for providing acceptable level of iodine.

3.2 A laboratory based technology for fortification of whole-wheat flour 'atta' with iron at 25 and 50 mg/Kg and folic acid at 220 mg/Kg was developed. The two indigenous sources of iron that met the criteria as a chemical source of iron were anhydrous ferrous sulfate and indigenous H-reduced iron powder. The percent bioavailability in adult male human volunteers was three times higher with anhydrous ferrous sulfate compared to H-reduced iron powder.

#### 4. NUTRIENT COMPOSITION OF NEW VARIETIES OF RICE

Eleven varieties of rice obtained from the Directorate of Rice Research, Hyderabad, were analyzed for proximate composition. The protein content in general was higher than 8 g as against 6.8g found earlier (Nutritive Value of Indian Foods). In one of the varieties, it was about 12g. Consumption of such high protein rice could increase the protein intake of individuals by 8 to 9 g/day. This perhaps explains the gradual improvement in PEM status in the country, despite little changes in Indian dietaries intake.

#### 5. NUTRITION AND CHRONIC DISEASES

5.1 Studies conducted on WNIN female rats have shown a correlation between maternal micronutrient status and predisposition of the offspring to insulin resistance in later life.

5.2 In a yet another study on rats, it was found that restriction of both dietary micronutrients as well as calories had adverse effects on fasting glucose & insulin and (Homeostasis model of assessment insulin resistance) HOMA IR index as compared to group with calorie restriction alone. It was also found that antioxidant status (Ferric reducing antioxidant potential - FRAP) was better and oxidative stress (Thio barbituric acid reactive substances - TBARS) low in rats of 'calorie restricted' group as compared to calorie and micronutrient restricted group. The results of this study

confirmed the role of micronutrients in modulating oxidative stress and hyperinsulinemia.

5.3 Postponement of cataractogenesis by diet restriction is in part due to improved antioxidant status and / or enhanced protein editing. The effect of longterm restriction of diet/food, protein and vitamin on the aggregation of crystallins and  $\alpha$ -crystallin chaperone activity in rat lens *vis a vis* cataractogenesis was, hence, assessed. Food and protein restriction lowered the susceptibility of  $\alpha$  - and  $\gamma$ -crystallins towards aggregation, while vitamin restriction tended to increase the aggregation of crystallins. Interestingly, only by vitamin restriction improved chaperone activity of  $\alpha$ -crystallin.

#### 5.4. ARI activity in foods

Diabetes mellitus advances onset and progression of cataract probably by modulating oxidative stress and glycation. Aldose reductase is important in diabetic cataractogenesis. Aldose reductase inhibitors (ARI) are valuable in delaying/preventing it. Screening of extracts of different plant foods indicates significant ARI activity in the extracts of amla, bitter gourd, fruits and Tulsi leaves. Curcumin appears to be the most potent ARI. Curcumin inhibits AR in a non competitive manner. Ongoing studies have shown that extract of black tea - but not green tea - significantly inhibited the glycation (argpyrimidine formation) of purified bovine lens  $\alpha$ - crystallin *in vitro*. Significant anti platelet aggregation activity was also observed in water extracts of eight varieties of green leafy vegetables.

#### 5.5. Dietary fatty acids and insulin resistance

Dietary fatty acids are known to affect the risk of chronic diseases particularly of diabetes and CVD. Adequate intake of essential PUFA namely linoleic (18:2n-6) and  $\alpha$ -linolenic acids (18:3n-3) and their optimal balance (n-6/n-3 ratio ~ 5-10) has been found necessary for

cardiovascular health. Studies were undertaken to investigate the effects of dietary fatty acids on membrane lipid composition and insulin sensitivity in target tissue in experimental rats.

Dietary saturated fatty acids (SFA) and trans fatty acids (TFA) decreased insulin sensitivity in target tissues (skeletal muscle and adipose tissue). The effects of TFA were more marked than SFA. The current evidence indicates that the atherogenic effects of TFA (increased LDL cholesterol and decreased HDL cholesterol) can be prevented by increasing dietary 18:2 n-6. However, the present studies show that increasing dietary 18:2n-6 did not prevent the TFA induced decrease in insulin sensitivity. On the other hand, increasing dietary ALNA and therefore decreasing n-6/n-3 ratio to 2 increased insulin sensitivity in target tissues in diet induced insulin resistant rat model.

## 6. RESISTIN AS A MOLECULAR LINK BETWEEN DIABETES AND OBESITY

NIDDM is a result of combination of genetic and environmental factors. Epidemiological observations suggest a strong patho-physiological association of obesity to NIDDM. However, the molecular link between obesity and diabetes remained to be resolved.

Resistin, a cysteine-rich secretory protein, identified from mouse adipocytes, is down-regulated by anti-diabetic drugs like thiazolidinediones (TZDs). Association of resistin with diabetes and obesity in human system has been controversial as opposed to the murine system. Thus, it appears that mouse and human differ resistins greatly not only in their sequence and structure but in their regulation and thus function. The physiological role of resistin in diabetes and obesity can be better understood with knowledge about its molecular features.

The genomic counterpart of resistin encompassing the transcriptional start and translational stop from mouse and human was amplified and sequenced using ABI automated DNA sequencer. Exon/intron boundaries of mouse

and human were demarcated using DIALIGN algorithm.

1. The mouse sequence has an additional intron of 2279 bp (intron X), which is absent in human resistin. *In silico* analysis revealed a number of transcription factor binding sites.
2. Of particular importance was the presence of PPAR responsive element (PPRE) along with the binding sites for other transcription factor like Ap1, NFkB, C/EBP.
3. The binding of PPAR/RXR complex to PPRE present on intron X was shown by gel shift assays.
4. Functional relevance of this DNA-protein interaction was demonstrated by transient transfection assays using luciferase as a reporter.

The typical characteristics of the recombinant proteins were studied further.

## 7. FOOD SAFETY

### 7.1 Application of Hazard Analysis Critical Control Point (HACCP) to animal products

HACCP is a preventive system to reduce microbial hazards. A study on HACCP was carried out in prawns. The samples were found to be contaminated with pathogenic microorganisms immediately after harvest and also at the market place. However, recipes prepared from prawn were free from pathogenic organisms. While pond water samples were contaminated with microorganisms including *Vibrio* sp., feed and ice samples used for preserving were free from pathogenic organisms. Every step in food chain has to be thoroughly investigated for identifying the source of contamination to ensure food quality and safety. The significance of the present study is that the pond water is the source of contamination. Hence, HACCP need to be practiced during the entire food chain from pond to plate and not only at the export processing units.

## **7.2. Utilization of mouldy sorghum using Bio-technological means for animal consumption**

A study was conducted to assess the feasibility of utilizing lactic acid bacteria, through natural fermentation, to detoxify contaminated sorghum and convert it to a value added animal feed ingredient. The laboratory trial of production of *Lactobacillus* fermentation of mouldy sorghum with addition of locally available resources (e.g. *Casia tora* seeds) was tested at village level by feeding it to cattle. This is a preliminary study, and the acceptability was monitored in terms of feed consumption, which was correlated with milk output.

## **8. NUTRITION AND CANCER**

### **8.1. In vivo model for genotoxicology**

A study was conducted to establish *in vivo* model to assess genotoxicity and evaluate the antimutagenicity of feeding allium through diet. Prior feeding of allium vegetables to rats reduced the DNA damage in tissues and urinary mutagens due to carcinogen treatment.

### **8.2. Studies on ginger**

Non-nutrient components of diet have been shown to inhibit carcinogenic process by enhancing the intracellular levels of xenobiotic metabolism enzymes, which play an important role in detoxification pathway. Spices and their active principles have been shown to possess chemopreventing properties. Study on modulation of xenobiotic metabolism by ginger showed that ginger feeding to rats resulted in stimulation of glutathione-s-transferase enzyme in liver, intestine, lung and kidney suggesting that regular intake of ginger can enhance the detoxification enzymes and thereby afford protection to host against tissue damage induced by exposure to xenobiotics.

### **8.3. Effect of vitamin restriction and supplementation on rat intestinal mucosal cell apoptosis**

50% vitamin restriction increases the oxidative stress, decreasing the anti-apoptotic protein (Bcl-2) expression thereby enhancing the apoptotic rates while vitamin supplementation reverses the changes caused by vitamin restriction. Among the individual vitamins, vitamin E showed maximum decrease in oxidative stress, as well as decrease in apoptotic rates.

## **9. Training activities in RCH**

A total of 186 investigators from different States in the country involved in the district level RCH surveys were trained in 10 batches at Nagpur, Bhubaneswar, Chennai and Hyderabad. Orientation was provided on appropriate techniques using finger prick blood samples for the estimation of haemoglobin levels.

At the request of Food and Nutrition Board, Department of Women and Child Development, Government of India, ICDS functionaries from North Eastern States were given orientation training in organization and establishment of Nutrition Surveillance System.

## **10. NATIONAL CENTRE FOR LABORATORY ANIMAL SCIENCES**

### **10.1 R & D Supporting Service Activities**

The Centre was involved actively in the breeding and supply of microbiologically and genetically defined animals to various government and private institutions. Besides supply of animals, blood and blood products were also supplied to different institutions to meet their specific R & D needs. Additionally the Centre was involved in supplying high quality animal feed to different institutes. All these service activities resulted in generating a total income of Rs.31.75 lakhs. The Centre trained a total of 50 candidates in various aspects of laboratory animals care, husbandry, nutrition, diseases, welfare and ethical aspects of experimentation. The Centre also extended technical consultancy in the field of laboratory animal sciences and technology for more than 20 institutions last year.

## **B. RESEARCH ACTIVITIES**

### **10.2 Effect of satiomem in obese rats**

A preliminary project in collaboration with ITRC, Lucknow on the effect of anorexigenic glycoprotein known as "satiomem" isolated from animal and plant membrane was tested in WNIN/Ob rats. The study confirmed the anorexigenic property of the compound shown earlier in normal rats and mice. However, the food intake reduction in these genetically obese rats was only 20%, compared to 40% seen earlier with normal rats. At the level compound is fed to the rats, no apparent toxic effect was seen.

### **10.3 Molecular analysis of WNIN/Ob rats**

The study on the candidate gene involved in obese rats was in collaboration with IISc., Bangalore. It comprehensively showed that leptin receptor gene was not altered in these rats, as per the evidence on coding regions as well as m-RNA levels of various splice variants of the Ob-R gene.

### **10.4 Genetic typing of obese rats using micro-satellite markers**

A project with funding from DBT was initiated for genetic typing of WNIN/Ob and GR-Ob strains using micro-satellite markers. These markers span all the 22 chromosomes (including sex chromosomes) of the rat and the mutants will be typed along with the parental strain WNIN, related strain WKY and unrelated strain Fischer 344. Out

of 100 markers selected for the study (4 markers/chromosomes), 22 markers were completed and unique markers typical of each strain could be observed.

### **10.5. PCR based DNA fingerprinting in obese rats**

The PCR based DNA fingerprinting using random primers yielded a unique DNA fingerprint profile for homozygous obese rats which is different from its other phenotypes and other standard reference strains. This has been submitted for patenting purposes.

### **10.6. Age associated oxidative stress in obese rats**

Studies on oxidative stress in obese mutant rats showed that lipid peroxidation and protein carboxylation to be high at the age of 3 months in cerebral hemispheres while the other regions of the brain were affected only after 9 months of age.

Some of the research studies initiated during IX five year plan period were continued in the X plan period as they were perceived to be of research significance. New thrust areas were identified and new equipments were procured for the fresh projects during the year. The research endeavours of the scientists and the technicians were the major propellers of the research work.

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