HIGHLIGHTS

Being the premier Institute of the country on Nutrition Research, National Institute of Nutrition geared up its potential to tackle various nutrition-related problems affecting our population groups. Several issues relating to nutrition, environmental health, food and drug toxicity and policies and plans of action were taken up for the study during the year. The impact of toxic chemicals on human health and the environment is becoming an increasingly pressing issue and particularly the presence of high levels of fluoride in the water is a cause for concern. A survey was carried out in 13 villages of five North-western districts of Tamil Nadu to study the fluoride levels in the drinking water and its effects on health. The draft country investment plan was prepared for implementation of nutrition programmes in Orissa and Rajasthan. The nutrition problems of different tribal communities at various stages of development were full of obscurities and very little scientific information on dietary habits and nutritional status was available due to lack of systematic and comprehensive research investigations. As part of this, a rapid survey was carried out on the nutritional status of the Saharia tribal population in Baran district of Rajasthan. In addition to these research activities, a state-of-the-art facility "National Facility for Dry Blood Spot Technology for Vitamin A estimation" was set up at NIN. Obesity is one of the predisposing factors for a range of non-communicable diseases. Epidemiological studies indicate a significant positive correlation between type 2 diabetes and obesity. Therefore, studies were initiated to identify the predisposing factors at molecular level as predictors in degenerative diseases such as diabetes in overweight/obese populations. Health benefits of turmeric and ginger -such as anti-inflammatory, anti-carcinogenic, antiviral- were studied. As part of extension activity, more emphasis was given to use school children under an FAO global initiative "Feeding minds and Fighting Hunger" project and NSS volunteers as change agents in the community to pass health and nutrition messages to the community.

1. COMMUNITY STUDIES

a. Fluorosis in North-Western districts of Tamil Nadu

A rapid survey carried out in 13 villages of five North-Western districts of Tamil Nadu viz., Vellore, Dharmapuri, Krishnagiri, Salem and Erode, revealed that the major source of drinking water was bore wells, which had flouride levels, ranging from 0.5 - 6.6 ppm. The overall prevalence of dental mottling among the total population ranged from a low 17% in the district of Vellore to a high 36% in Dharmapuri. The prevalence was relatively higher among the children of 10-14 years (30% to 67%) in all the districts surveyed. The overall prevalence of skeletal fluorosis in population of all age groups was found to be less than 1%. Creating awareness about fluorosis among the community, discouraging consumption of water derived from high fluoride water sources and supply of drinking water from rivers and tanks were recommended as some measures for the prevention and control of fluorosis.

b. Nutritional status in Saharia Tribes of Rajasthan

A rapid health and nutritional assessment survey carried out among Saharia tribes in Shahbad and Kishanganj blocks of Baran district, revealed that the average intakes (CU/ day) of various foods, except for cereals & millets was less than RDI, while that of protective foods like pulses, green leafy vegetables and milk & milk products was grossly inadequate (less than 40% of the RDI). Though the average intake of protein, calcium, iron, thiamine and niacin was comparable with RDA, the intakes of other nutrients were below the RDA. The extent of deficit was maximum with respect to Vitamin A (62 %), followed by vitamin C (43 %), total fat (35 %), folic acid (22 %), riboflavin (14%) and energy (9%). The prevalence of clinical forms of vitamin A deficiency such as conjunctival xerosis (17.6%) and Bitot spots (8.3%) among preschool children was observed to be much higher than that reported for the State of Rajasthan (0.3%). The overall prevalence of underweight (weight for age <Median -2SD) was 72%, while that of severe grade (weight for age < Median -3SD) was 24%. The overall prevalence of stunting was 68% and that of wasting was 13%. The prevalence of chronic energy deficiency (CED) among adults was higher (56%) as compared to that reported for the State (45%).

Verbal autopsy of the deaths reported during the previous six months revealed that none of them were attributable to starvation. The study highlighted the need for strengthening of health and nutrition programmes such as RCH, ICDS and MDM.

2. CLINICAL AND PHYSIOLOGICAL STUDIES

A study on 'Body composition measurement by Dual Energy X-ray Absorptiometry (DEXA) in women from a slum of Hyderabad revealed that Indian women from low-income group have high levels of body fat % at comparatively lower body mass index (BMI) levels than other ethnic groups. Even women in the adequate BMI category (18.5 - 23) had unacceptable high levels of body fat. Increase in weight and BMI was associated with increase in body fat % whereas increase in height was associated with increase in lean mass but not fat %.

Another study on pregnancy induced hypertension showed that pregnant women with pre-eclamptic toxaemia had decreased levels of antioxidants (vitamin C, E and β-carotene) as compared to the control group. Vitamin C was further decreased significantly in women with severe pre - eclampsia compared to mild to moderate groups.

3. BASIC STUDIES

A state-of-the-art facility 'National Facility for Dry Blood Spot Technology for vitamin A estimation" was established with the financial support from MI and MOST, New Delhi. This facility is expected to help achieve the goal of elimination of vitamin A deficiency in the country by 2010 AD, by providing data on extent of sub-clinical Vit. A deficiency in the

community and by capacity building of other institutes in the methodology of assessment of the problem.

On the request from the DWCD, the draft country investment plan entitled "Country investments plan (CIP) for micronutrient fortification of food supplements of ICDS and mid day meal in the States of Orissa and Rajasthan" has been prepared and submitted to DWCD, Govt. of India. Instead of Andhra Pradesh and Rajasthan originally considered and approved earlier the proposal envisages implementation of the CIP in the States of Orissa and Rajasthan for supplementation of micronutrients - iron, zinc, iodine, vitamin A, thiamin, riboflavin, folic acid and ascorbic acid at RDA level to the beneficiaries. The plan requires an investment of Rs. 43.8 cores for a period of 3 years.

Type 2 diabetes and obesity-related metabolic disorders are rapidly emerging as major health risk factors in the Indian population. Epidemiological observations suggest a strong correlation between type 2 diabetes and obesity, however, the mechanisms by which increased adiposity causes insulin resistance is poorly understood. Resistin, a cysteine rich polypeptide, specifically secreted by the adipocytes in mouse has been implicated to be the link between type 2 diabetes and obesity. However, in humans, its role appears to be highly controversial. Therefore, the major focus has been to understand the role of resistin in human physiology. Earlier, the human resistin purified from *E.coli* was cloned, expressed and characterized. Recombinant human resistin was found to be unusually stable protein which undergoes concentration dependant structural transition. It has also been found that the protein exists in dimeric form. Since type 2 diabetes and obesity are considered as inflammatory states, a study was done to examine whether resistin plays a role in eliciting inflammatory conditions. It was demonstrated that resistin induces secretion of TNF $\propto$ and IL-12 from monocytes/macrophages and thus can be considered a pro-inflammatory cytokine itself. The pro-inflammatory action of resistin is mediated by NFkB and was found to involve ERK pathway.

Sprouting, malting, deep and shallow frying, roasting, pressure cooking were some of the processing methods used to study the variation in phenolic content (PC) and antioxidant activity (AOA) of some foods. It was seen that the effect of processing could be different on the AOA and PC of not only foods of different groups but among foods of same groups as well. All the processing methods tested, either had marginal or no effect on AOA of foods. However, sprouting seems to enhance the PC of legumes. Also sprouted legumes had highest AOA amongst all the processed foods tested.

Cataract characterized by the opacification of eye lens, is the main cause of blindness worldwide. Oxidative damage to the lens constituents is an important factor in the etiopathogenesis of cataract. Development of strategies to reduce the incidence of cataract needs emphasis. Exploring and utilizing the beneficial effects of dietary and nutritional components in terms of preventing/ postponing cataractogenesis could be one of the simplest and cost-effective strategies. Studies have been carried out to understand the molecular basis of cataractogenesis and the anticataractogenic role of dietary components.

Turmeric delays diabetic cataract: Turmeric is a widely used spice in Indian cuisine. Curcumin, the active principle of turmeric, has been shown to have numerous health beneficial properties such as anti-inflammatory, anticarcinogenic, antiviral and antiinfectious activities. Studies done here have established a yet another beneficial property of turmeric - delaying of diabetic cataract in rats. The studies suggest that curcumin and turmeric prevented aggregation and insolubilization of lens proteins due to hyperglycemia by minimizing the oxidative and osmotic stress. Furthermore, the results provide a clue, for the first time, that turmeric or curcumin may act downstream to glucose-mediated changes. Hence, turmeric provides a viable food-based, as well as pharmacological approach in the treatment of diabetic complications such as cataract. Therefore, these results imply that ingredients in our dietary sources, such as turmeric, may be explored for their anticataractogenic potential.

Modulation of chaperone activity of α-crystallin under diabetic cataract by curcumin: It has been established that chaperone function of α-crystallin is critical in the maintenance of transparency of the lens vis-à-vis cataract formation. Diabetes is one of the major risk factors in cataract formation. Impaired chaperone function of α-crystallin could be involved in the formation of diabetic cataract. It has been demonstrated that in diabetic cataract, α-crystallin chaperone activity is diminished. Interestingly, dietary curcumin delayed progression and maturation of diabetic cataract by modulating the chaperone activity of α-crystallin. The antioxidant effect of curcumin appears to be the predominant mechanism for the modulatory effect on α-crystallin chaperone activity in diabetes and thereby delaying cataract in rats.

Inhibition of protein glycation by dietary agents: Multiple molecular mechanisms have been proposed to explain the pathogenesis of long-term complications of diabetes, one of them which is considered to be a prominent upstream phenomenon down the line that leads to various deleterious consequences is non-enzymatic glycation. The high glucose in diabetes reacts non-enzymatically with the amino groups of proteins to form advanced glycation end products (AGE). It has been shown that formation of AGE *in vivo* contributes to several pathophysiologies associated with ageing and diabetes mellitus, such as cataract. Hence, the role of antiglycating agents delaying the onset or progression of diabetic complications has gained considerable importance. Preliminary data indicate that dietary agents (code names; MYB1, MYB2, MB1 and MYB3) are found to be the effective inhibitors of protein glycation in vitro, MYB1 being the most potent. Hence, studies are underway to exploit the antiglycating potential of these dietary agents in the management of secondary complications of diabetes.

Effect of methylglyoxal on α-crystallin: Methylglyoxal (MGO), a major dicarbonyl compound, is present in high concentrations in lens compared to plasma or any other tissue and its levels increase several folds during diabetes. Compared to other agents, MGO is very potent in terms of forming advanced glycation end products (AGE). With the given importance of molecular chaperone function of α-crystallin in maintaining transparency, the study investigated the effects of MGO on α-crystallin structure and function. It was shown that

MGO has unfavorable effects on α-crystallin chaperone-like activity, as MGO-modified α-crystallin showed decreased hydrophobicity, altered secondary/tertiary structure and increased oligomeric size. Further, non-enzymatic browning of α-crystallin by MGO leads to decrease in its stability and unfolding that in turn leads to the exposure of buried proteolytic sites causing enhanced proteolytic degradation. ATP could not protect the glycated α-crystallin from proteolytic degradation as observed with native α-crystallin. Results of the present study provide the basis for the role of non-enzymatic glycation on α-crystallin chaperone activity in age-related brunescent and diabetic cataracts.

4. PATHOLOGY

Maternal malnutrition and hyperglycemia during pregnancy as well as foetal undernutrition in long term, in experimental animals, are known to affect the development of foetal pancreas and the structure & function of the islets, particularly the ß-cells.

A study was therefore carried out to look into the status of pancreatic islets with respect to morphological changes in aborted human foetuses, aged 16 to 20 weeks, obtained by MTP, from undernourished mothers (BMI<18) and compare them to those obtained from adequately nourished mothers (BMI>20). There were no significant differences in the number, size or density of the islets as well as the beta and acinar cell counts between the pancreas of aborted foetuses belonging to undernourished and adequately nourished mothers.

Micronutrient status of children during acute respiratory infection and its association with local cytokine (Th1, Th2) response was determined. Also impact of large dose vitamin A on Th1 and Th2 modulation and cytokine response was studied. It was found that vitamin A suppresses Th1 response (IL2) which may be mediated by down regulation of IL12, indicating that vitamin A could be anti-inflammatory.

It was also found that vitamin A might alter the course of immune response in acute respiratory infections like URTI, pneumonia and bronchiolitis and thus influence the outcome of such infections. However, weight-for-age, haemoglobin and zinc did not show any association with Th1 or Th2 cytokines.

5. EXTENSION AND TRAINING

FAO and a group of organizations launched a global nutrition education initiative “Feeding Minds, Fighting Hunger” (FMFH) for school children. A study was conducted between October 2002 and 2004 to evaluate the efficacy of FMFH lesson plans in improving nutrition-related knowledge levels of the schoolchildren in Hyderabad. The schools in experimental and control groups were randomly chosen from the member schools of a collaborating voluntary organization, Confederation of Voluntary Associations (COVA). At baseline, the knowledge levels of VIII and IX class children and teachers in both the groups were measured. A significant improvement in the knowledge levels of the teachers was observed in experimental schools after two workshops, in which FMFH lesson plans were introduced to teachers and topics for development of communication material were identified.

The intervention with communication material such as posters, skit and classroom activities, in the classroom setup through teachers resulted in a significant improvement ($p<0.01$) in the knowledge levels of pupils in experimental group. Significant improvement ($p<0.01$) in nutrition related knowledge was also observed in control group. Effect size indicated by the improvement of the knowledge-levels in experimental schools over control group was medium ($d = 0.40$) indicating the efficacy of FMFH programme in improving nutrition-related knowledge of schoolchildren. No significant decrease ($p>0.05$) in the knowledge levels was observed after two months, indicating retention of the knowledge acquired through the programme.

6. FOOD SAFETY

A study was carriedout to detect transgenic DNA and protein (Cry1Ac) in cotton seed samples obtained from farmers. Presence of Cry1Ac protein was detected in the samples collected. The levels were comparable to that of standard seed samples obtained from popular agencies like MAHYCO.

7. CANCER AND XENOBIOTICS

Antimutagenic property of ginger was observed in *vitro* and *in vivo* experiments. These effects could be due to its antioxidant property. In vivo study in rats showed that ginger intake through diet can result in improved antioxidant status and this could be one of the explanations for its antimutagenic property. A study conducted on people in high risk area for developing upper gastrointestinal cancer indicated that nitrosation potential was higher in these individuals as compared to those from low risk area. In the area of social drug epidemiology, an intervention programme to sensitise the public on rational use of drugs was developed and was used for educating the masses on rational practices in drug usage.

It is a practice to use herbal medicines in treatment of various ailments since time immemorial. The anti-inflammatory and antioxidant properties of herbs used in arthritis are being evaluated for their biological effects using air pouch model in experimental animals.

8. NATIONAL CENTRE FOR LABORATORY ANIMAL SCIENCES

The National Centre for Laboratory Animal Sciences (NCLAS) carries out service as well as basic research activities. The centre caters to the needs of biomedical research community in terms of supply of laboratory animals, laboratory animal feed and also blood and blood products from animals. Compared to last year, there has been 13% decrease in the breeding and 11% decrease in the supply of animals during the current year. However, the income generated as a whole was not affected, since there was a paradigm shift in demand

for more rats than mice compared to last year. With respect to animal feed supply, the supply remained at the same level as last year. But, there was a new demand for experimental feeds, deficient in one nutrient or other, and this new activity generated more than half a lakh rupees income in the current year. The centre continued its supply of blood/sera from various laboratory animals at the same level as last year. Health monitoring of the laboratory animals was carried out with a larger sample size this year. Most of the infections seen were among from the older animals.

The centre has been carrying out basic research work on the obese rat model developed at its experimental facilities for the past few years. DNA finger printing of the obese mutant rats using random primers yielded a fairly constant DNA fingerprint for the GR-Ob strain. The PCR products generated (360 bp, 390 bp, 400 bp, and 600 bp) are cloned in suitable vector and sequenced and were found to have homology with sequences on rat chromosome no.3, 8 and partially with X chromosome. Further, when the clone containing the 360 bp insert was used as a probe, it showed hybridization with WNIN/GR-Ob samples, indicating that the cloned region is a part of the rat genome. With respect to WNIN/Ob, all the attempts to generate an elusive fingerprint remained unsuccessful.

The extramural project (funded by DBT) on genetic typing of the obese mutant rats (WNIN/Ob and GR-Ob) using microsatellite markers concluded this year. Out of the 96 markers screened, 62 markers showed successful amplifications. The cluster analysis of the data generated showed two distinct clusters of rats, the parental WNIN along with mutants forming one cluster and WKY, F-344, forming the second. This clearly shows that the mutants have indeed originated from WNIN and there is no contamination from other strains.

The study also revealed 9 markers which in combination can be used for identifying the three standard rat strains - WNIN, WKY and F-344. Amongst the 9 markers, the primer for leucosianin seems to be very promising as it can distinctly identify all the three strains as well as mutants. A detailed analysis of this marker is now planned.

Baseline data with respect to body composition, physical activity, clinical chemistry and physiology of laboratory animals was initiated and studies on WNIN, SD and F-344 rats were completed. The study showed significant differences between the 3 strains of rats and the study is now extended to other three rat strains (WKY, CFY and Holtzman) as well.

Constant and continuous research pursuits of the Institute not only helped in understanding the newer facets of the nutrition research but also provided appropriate guidelines for National level policy making on Nutrition.