

RESEARCH HIGHLIGHTS

1. Assessment of current nutritional status of below five year children and performance of ABM project in the districts of Madhya Pradesh

The study was undertaken to assess the changes in the nutritional status as well as IYCF indicators among <5 year children after launching of Atal Bal Arogya Evam Poshan Mission (ABM) implemented during the past 5 years (2011 v/s 2016). It was a community based cross-sectional study carried out in all the districts (51) of Madhya Pradesh by adopting systematic random sampling procedure. A total of 37924 <5 year children from 30585 households were covered. About 79% fathers and 68% mothers were literate as against 71% and 48% covered in previous survey. Proportion of HHs living in pucca house has increased from 10% to 24% during the same periods. Use of sanitary latrine was 33% as against 12% in 2011. Antenatal check ups has increased from 78% in 2011 to 98% in 2015 and registration in first trimester has increased to 76% from 28% in last survey. Consumption of ≥ 90 IFA tablets during pregnancy has improved from 20% in 2011 to 59% in 2015. There was change in prevalence of low birth weight (19% vs 16%). The initiation of breastfeeding within 1 hr of birth has increased to 58% from 26% in 2011. About 93% children were exclusively breastfeeding in current survey as against 71% in 2011 among 0-5 mo. children, while exclusive breastfeeding up to 6 months has increased from 43% to 64% among 6-11 mo. children. The ICDS supplementation has also improved among 6 to 59 months children (81-84% in 2015 vs 68-72% in 2011). The prevalence of underweight, stunting and wasting has declined to 41%, 43% to 50% in 45 districts while 5 districts showed increase in prevalence.

2. Introduction of vitamin C rich fruit in supplementary nutrition programme for improving micronutrient status, gut health, growth and development: A randomized trial among ICDS pre-school beneficiaries

Iron deficiency anemia (IDA) is a significant public health issue in India affecting nearly all the vulnerable segments of the population. IDA is caused due to low consumption of iron-rich foods combined with its poor bioavailability. To date, interventions aimed at inclusion of vitamin C rich fruits in the regular diet showed to improve iron absorption, but the effect on iron status is inconclusive. Considering the ongoing national program for preschoolers in India, a cluster randomized controlled trial (RCT) was designed to test the hypothesis that inclusion of vitamin C rich fruit in a regular meal would improve iron absorption and lead to better child iron and micronutrient status, cognitive development and gut health while reducing morbidity. A 3-arm, open-labeled, assessor-blinded, cluster randomized control trial among 2-5yr old ICDS beneficiaries was adopted (CTRI/2014/09/004983) in sixteen villages of Alair sub-district, Nalgonda, Telangana State. Villages were randomized to experimental guava group (GG), passive control banana group (BG) and active control cucumber group (CG). Participants (n=407) received 25g of fruit for 140 (137-144) days along with the supplementary meal catered through Supplementary Nutrition Programme (SNP). Cognitive development, using Mullen's scale, anthropometrics, blood and stool samples were assessed both at baseline and endline. Morbidity was monitored bi-monthly using caregiver's recall for last 15 days. A linear mixed model accounting for adjusting clustering and confounder effect was applied. Demography, socio-economic status and child's dietary diversity at home were comparable across the groups. The inclusion of guava improved iron: vitamin-C molar ratio from 1:1 to 1:12. Group difference between endline and baseline measures were significantly higher in GG for Hb (0.8g/dL; p=0.002), SF (5.5 μ g/L; p<0.001), vitamin-C (0.7mg/dL, p=0.047) and lower for sTfR (-0.6mg/L; p<0.001) over the controls. Though, there was no significant group difference in cognitive development and growth, morbidity on acute lower respiratory infection (ALRI) was significantly lower in GG (42%) compared to BG (62%, p=0.01) and CG (55%, p=0.08). These results suggest that diversifying meals of preschoolers with guava significantly improves iron status and can be adopted as a strategy to address ID among children.

3. Monitoring of nutritional status and catch up growth of boarders of Andhra Pradesh Social Welfare Residential Educational Institutions (APSWREI)

Good nutrition during growing phase is an investment that has many long-term benefits. Andhra Pradesh Social Welfare Residential Educational Institutions (APSWREI) which has been providing residential facilities that includes nutrition at no cost for children from deprived settings provides an opportunity to assess the benefits of good nutrition during adolescent phase. This study was conducted in APSWREI in the state of Andhra Pradesh. Seventeen APSWREI schools were randomly chosen from seven districts. Quantitative and qualitative methods were used in data collection. A total of 7325 children were surveyed and 48.4% of them were boys. The median age of the children was 14 years (8-19 years Range). The overall thinness was 15.8% and was higher among boys (22.5%) compared to girls (9.5%). The overall stunting was 28.5% and was similar in boys (28.3%) and girls (28.6%). Severe thinness and severe stunting were less than or about 5% for both boys and girls. Milk was the commonest food skipped by APSWREIS due to rising costs. The monotonous nature of the diet was a hindrance for regular food consumption among the students. Based on the findings, a new model menu to APSWREIS and inflation adjusted food rates have been recommended.

4. Effect of Yoga Nidra on blood pressure and mental health status of hypertensive subjects - A pilot study

Yoga Nidra is a “systematic” method of inducing complete physical, mental and emotional relaxation. This study was conducted to assess the effect of “Yoga Nidra” on blood pressure and mental health among hypertensive subjects. Both known and newly diagnosed subjects (n=74) with HTN aged between 35-70 years were included in this study. Subjects with critical illness and any psychological ailments were excluded. The Experimental group (n=32) included subjects who showed interest in Yoga Nidra to practice daily (45 min/day) for 3 months. Others were included in control group (n=42) and were asked to continue with their regular diet and physical activity along with medications prescribed by their Physicians. After adjusting baseline data, highly significant reduction of Systolic Blood Pressure (mean 16 mm/hg) and Diastolic Blood Pressure (mean 8.6 mm/hg) was observed among experimental group (p<0.001**). A significant reduction of mean Hs-CRP (p<0.001**) was observed among experimental group. Similarly a significant reduction of Depression, Anxiety and Stress were observed among experimental group. In this pilot study, a significant reduction of blood pressure and level of Stress, depression and anxiety were significantly reduced among Yoga Nidra group as compared with control group were observed. There were no side effects observed during this study.

5. Modeling developmental origins of health and disease in the mouse embryonic stem cells/ mESCs and BMMSCs - with Folate deficiency – Cellular, Molecular / Epigenetics

Nutritional perturbations *in utero* have shown to have a direct bearing in the health of offspring being complemented with several altered metabolic indices which co-precipitate as NCDs adults. Using adult bone marrow derived stem cells (BM-MSCs) which possess multipotent trilineage differentiation functions (osteoblast, adipocyte and chondrocyte), have been able to show for the first time *in vitro*, for an altered cellular milieu of BM-MSCs with 70 % folate deficiency akin to the published data using model system. These include upregulation of pro-inflammatory cytokines, altered: leptin to adiponectin ratio, epigenetic changes and increased commitment towards visceral adipogenesis. The findings advocate for the promises of stem cells as an *in vitro* model system to recapitulate the developmental changes and as an alternative to minimize the usage of large number of animals required for nutritional based studies for better understanding of nutrient and gene interactions.

6. Exploring the beneficial effects of endothelial cells generated from human derived mesenchymal stem cells in the management of lymphoedema - *In vivo* approach

The field of lymphoangiogenesis has made rapid and exciting developments over the last few years more so than any other field in vascular biology. Lymphoedema of the limbs and other parts of the body due to lymphatic blockage secondary to parasitic infection due to wuchereria bancrofti or due to

mastectomy. The importance and therapeutic value of stem cells in lymphangiogenesis are poorly understood due to the lack of specific lymphatic molecular markers and the unavailability of optimal experimental models. *In vitro*, characterization of lymphoangiogenic markers which were more predominantly expressed in omental adipose tissue as compared with subcutaneous depots were demonstrated. The upregulation of VWF and Cd31, specific for lymphoangiogenesis in omental tissues have been depicted in the study. *In vivo* experiments were could not carried out to transplant the omental lymphoendothelial cells in rat model system due to technical difficulties of inducing lymphoedema in rats.

7. Modulation of adipose tissue inflammation by dietary n-3 polyunsaturated fatty acids - Potential role in metabolic syndrome

It is well established that the increase in visceral adiposity is the central component of metabolic syndrome. Increased adiposity leads to a chronic low grade inflammation in the adipose tissue, resulting in increased production of pro-inflammatory cytokines and decreased production of anti-inflammatory cytokines which in turn decrease insulin sensitivity. An animal experiment was carried out to investigate the impact of substitution of dietary n-6 PUFA (linoleic acid) with n-3 PUFA (α -linolenic acid present in vegetable oil or long chain n-3 PUFA present in fish oil) on adipose tissue inflammation in fructose induced model of metabolic syndrome and to understand the molecular mechanism by which n-3 PUFA increases insulin sensitivity. The results showed that rats fed high fructose diet increased visceral adiposity, dyslipidemia and insulin resistance compared with starch fed control rats. Fructose feeding also induced adipose tissue inflammation, infiltration of macrophages and increased oxidative stress. Substitution of linoleic acid with α -linolenic acid (n-6:n-3 ratio of 2) or LC n-3 PUFA (n-6:n-3 ratio of 5) reduced visceral adiposity, increased insulin sensitivity and corrected dyslipidemia. The improvement in insulin sensitivity by n-3 PUFA supplementation was associated with marked reduction in adipose tissue inflammation, decreased macrophage infiltration and decreased oxidative stress. The results of the present study provide insights into the therapeutic implication of n-3 PUFA supplementation in obesity related metabolic sequelae.

8. Assessment of nutritional and morbidity status and utilization of health care facilities in the elderly population aged 60 years and above

The prevalence of chronic energy deficiency (CED) was low (9.6%) and overweight, obesity (43.3%) and central obesity (67.6%) was high among urban elderly population. According to the Mini Nutritional Assessment (MNA) 62.3% of urban elderly are at the risk of malnutrition. The mean consumption of all the food groups, (except cereals & millets and Fats & oils) is below the RDI. Similarly, the mean intakes of all nutrients (except total fat) are below the RDA (Dietary Guidelines, 2010). The overall prevalence of anemia, diabetes and hypertension in urban elderly was 46.44%, 32.4% and 74.6% respectively. The overall self-reported prevalence of general morbidities like poor vision, joint pains, forgetfulness, diminished hearing and chewing problems in urban elderly subjects are 73.3%, 62.6%, 28.3%, 27.1% and 24.8% respectively. Based on Barthel ADL Index, the overall prevalence of functional disability in urban subjects is 23.3%. In overall, majority (63.7%) of urban elderly utilized private hospital services, 14.2% utilized public hospital services. Similarly, majority (80%) of urban elderly utilized private doctor services, 17% utilized medical doctor (government doctor) services.

9. Impact of hyperglycaemia on invasion properties of first trimester trophoblastic cells: Implication on preeclampsia

During early gestation, a hypoxic condition is critically maintained by optimal glucose metabolism and transporter activities. Glucose is readily available energy nutrient required for placentation. However, limited data are available on glucose uptake and its transporters during first trimester placentation processes. To this end, effects of glucose and the roles of glucose transporters (GLUTs) were investigated during hypoxia on trophoblast migration and placental angiogenesis processes using early gestation-derived trophoblast cells, HTR8/SVneo, and first trimester human placental explant tissues. The study suggests that putative roles of GLUT1 in the glucose uptake and tube formation of the first trimester placental trophoblast cells, HTR8/SVneo. Increased glucose uptake and GLUT1 activities favor HIF1 α

activation in first trimester trophoblast cells that may stimulate glycolytic and lipid metabolic activities with stimulation of angiogenesis due to increased tube formation and expression of several pro-angiogenic mediators. such as VEGF, MMP9, GLUT1, and FABP4. Increased lactate production in response to glucose could induce VEGF pathway of placental angiogenesis in first trimester trophoblast cells. This work reported that GLUT1 plays an important role in both basal and glucose-stimulated glucose uptakes, glucose-stimulated tube formation, and insulin-stimulated glucose uptake of the first trimester HTR8/SVneo cells. GLUT1 protein expression and glucose transporter activity are decreased in pre-eclampsia, which indicates that optimum GLUT1 function may be required to prevent development of IUGR. However, further work is required to underpin the mechanisms of GLUT1 action in first trimester placental trophoblast cells.

10. Prevalence of vitamin deficiencies in the apparently healthy urban adult population: Assessed by sub-clinical status and dietary intakes

In this exploratory study, sub-clinical vitamin status of apparently healthy adults is evaluated. A total of 270 apparently healthy urban adults aged 30-70 years from Hyderabad city, forms the study subjects. Blood levels of vitamins (A, B1, B2, B6, total and active B12, D & folate) and homocysteine were assessed. Anthropometric parameters were measured, and dietary intake was obtained by food frequency questionnaire and probability of adequacy (PA) was calculated by estimated average intake. Among the study population, the overall prevalence of deficiency of vitamin B2 was strikingly high (50%) followed by vitamins: B6 (46%), active B12 (46%), total B12 (37%), folate (32%), D (29%), B1 (11%) and A (6%). Hyperhomocysteinaemia (HHcys) was widely prevalent (52%) in the study subjects. In conclusion, the study demonstrated that a high prevalence of multiple sub-clinical vitamin deficiencies, dietary inadequacies along with HHcys among apparently healthy adults which are possible risk factors for disease burden.

11. Factors associated with adequacy of micronutrient intakes among the urban adult population of Hyderabad city

Though the Indian urban population is assumed to have better access to diversified foods, data on micronutrient adequacies are meager in urban adults. This study assessed the adequacy of micronutrient intakes, dietary patterns and the associated factors among the apparently healthy urban adults. The community-based cross-sectional study involved 300 urban adults (distributed into age groups: 21-40, 41-60 and >60 years) residing in Hyderabad city. Dietary intakes were assessed by a three-day 24-h dietary recall and calculated the probability of adequacy (PA) using estimated average requirement. The PA of folate, B12, and zinc (1-11%) were noticeably low. The mean probability of adequacy (MPA) across the ten micronutrients was 38% and was comparable among the age groups and genders. Cereals & millets contributed to thiamine, niacin, zinc, and iron, green leafy vegetables and fruits for vitamins A, C, folate, and iron, animal foods for B12, milk & milk products for calcium, vitamin A, riboflavin and B12. The overall prevalence of anemia, iron deficiency (ID) and iron deficiency anemia (IDA), were 30%, 23% and 14.3%, respectively. The unadjusted and adjusted (age and gender) logistic regression models revealed that the risk of micronutrient inadequacy is associated with higher odds of lower educational status, IDA, and folate deficiency. These results indicate a higher micronutrient inadequacy among the healthy urban adults.

12. Circulating levels of Hsp27 in microvascular complications of diabetes: Prospects as a biomarker of diabetic nephropathy

Heat shock protein 27 (Hsp27) is a small heat shock protein known to protect the cells from apoptosis under stress. In the present study, the plasma Hsp27 levels in type 2 diabetes subjects without and with microvascular complications- diabetic retinopathy (DRe), diabetic nephropathy (DNe), and diabetic neuropathy (DNe) were estimated to understand if it could serve as a marker for these complications. This is a hospital-based case-control study with 754 subjects including 247 controls, 195 subjects with diabetes, 123 with DRe, 80 with DNe and 109 with DNe. Plasma Hsp27 levels were measured by ELISA. The mean plasma Hsp27 was higher in the DNe group (631.5±355.2) compared to

the control (496.55±308.54), diabetes (523.41±371.01), DRe (494.60±391.48) and DNu (455.21±319.74) groups with a p-value of 0.018. Receiver operating characteristic (ROC) curve analysis of Hsp27 in DNe group showed an area under the curve (AUC) of 0.617. Spearman correlation analysis shows a positive correlation of plasma Hsp27 with serum creatinine (p=0.053, r-value 0.083). Gender, age and BMI did not affect the plasma Hsp27 levels. The plasma Hsp27 levels in the DNe group are higher compared to the control and other complications, thereby it could be explored to be used as a potential biomarker of DNe.

13. Implication of altered ubiquitin-proteasome system and ER stress in the muscle atrophy of diabetic rats

Skeletal muscle is adversely affected in type-1 diabetes, and excessively stimulated ubiquitin-proteasome system (UPS) was found to be a leading cause of muscle wasting or atrophy. This study investigated the role of UPS and ER stress in the muscle atrophy of chronic diabetes rat model. Diabetes was induced with streptozotocin (STZ) in male rats and were sacrificed after 2nd and 4th months. In another experiment, 2-months post-STZ-injection diabetic rats were treated with MG132, a proteasome inhibitor, for the next 2-months. The muscle fiber cross-sectional area was diminished in diabetic rats. The expression of UPS components: E1, MURF1, TRIM72, UCHL1, UCHL5, ubiquitinated proteins, and proteasome activity were elevated in the diabetic rats indicating activated UPS. Altered expression of ER-associated degradation (ERAD) components and increased ER stress markers were detected in 4-months diabetic rats. Proteasome inhibition by MG132 alleviated alterations in the UPS and ER stress in diabetic rat muscle. Increased UPS activity and ER stress were implicated in the muscle atrophy of diabetic rats and proteasome inhibition exhibited beneficiary outcome.

14. Development and validation of a comprehensive index for assessing food safety at household level

Food safety is an essential pillar along with nutrition and food security to ensure peoples' health and its sustainable development. In India, majority of foods are prepared at household level. Stored cooked foods (>2 hours) and uncooked foods are susceptible to microbiological contamination due to unsafe handling practices. The study aimed to develop and validate a household food safety index (HFSI) that can predict household food safety status. In this cross-sectional study, for development of HFSI, primary food prepares (N=400) were selected @200 each from rural (Ranga Reddy district) and urban (Hyderabad) homes of Telangana. An 87-item pre-tested questionnaire covering knowledge (43), practices (36) and enabling assets (8) was administered on subjects. Weightages were assigned for responses and maximum possible score was 205. Simultaneously, at consumption point @400 each of stored cooked-food, drinking water samples and hand rinses were collected from participants for microbial analysis. Each of the 87 variables were associated with high-risk food-borne pathogen (*Salmonella* spp.) risk value (1.55logCFU/g) in order to identify the key risk variables that would form the HFSI. Eleven out of 87 parameters were significantly associated (p<0.05) with pathogen risk in foods. The final HFSI therefore was a 11-item questionnaire. The optimal cut-off value for validated 11-item HFSI score estimated 9 and it was found to have a sensitivity (77%), specificity (74%) and AUC-0.808 which are acceptable. Rural households showed significantly lesser HFSI score than the urban. The HFSI was then administered on 200 rural and urban (@100each) subjects for assessing the food safety status. To address the identified critical food safety issues at home, key messages were developed and implemented as an educational intervention on 120 households from Rural, Urban and Slum (@40each). There is a significant (p<0.05) improvement in HFSI score after educational intervention. HFSI is simple tool to rapidly assess food safety status at household level. Identified critical issues of food safety can be addressed through focused food safety education.