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## Original Article

# Diet and exercise intervention, with special reference to micronutrients, reduces cardiometabolic risk in overweight children

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### ABSTRACT

**Objective:** To study the effect of lifestyle intervention in the presence of multivitamin-zinc supplementation in improving the cardiometabolic status of overweight children. **Materials and Methods:** Data were evaluated in 74 overweight children ( $11.3 \pm 2.9$  years) randomly assigned to three groups of intervention for 4 months as follows: group A: diet-exercise counseling with multivitamin-zinc supplementation; group B: diet-exercise counseling; and group C: placebo. Anthropometric, biochemical, carotid arterial and lifestyle parameters were assessed. **Results:** Lifestyle counseling resulted in significant reduction in inactivity, energy and fat intakes and increase in micronutrient density of diets and physical activity in groups A and B in comparison to group C. Percent decline in body fat was more in group A than in groups B and C. Percent change in triglycerides ( $-13.7\%$ ) was significantly higher in group A than in groups B ( $-5.9\%$ ) and C ( $5.7\%$ ). Pulse wave velocity and elasticity modulus reduced and arterial compliance improved significantly in group A than in group B. **Conclusion:** Multivitamin-zinc supplementation with lifestyle intervention has a positive effect of on the cardiometabolic status of overweight children.

**Key words:** Children, India, lipids, micronutrients, stiffness

## INTRODUCTION

Developing countries are facing the double burden of nutritional transition, i.e. malnutrition and micronutrient deficiencies as well as a rapidly growing epidemic of childhood obesity.<sup>[1]</sup> Obesity is highly correlated with a constellation of disorders including dyslipidemia, insulin resistance, and hypertension, which are hallmarks of the metabolic syndrome (MS) and risk factors for cardiovascular disease.<sup>[2]</sup> Studies in children indicate that the process of atherosclerosis starts at an early age and is linked to obesity.<sup>[3]</sup> Prevalence of overweight and obesity in children and

adolescents in India is reported to be high,<sup>[4]</sup> and may lead to increasing risk of atherosclerosis and cardiovascular disease (CVD) in their adulthood.<sup>[5]</sup>

Obesity and MS are considered as oxidative stress related disorders. Oxidative stress plays a critical role in the pathogenesis of hypertension and atherosclerosis by directly affecting vascular wall cells.<sup>[6]</sup> Increased oxidative stress in accumulated fat is an important pathogenic mechanism of obesity-associated MS.<sup>[7]</sup> Therefore, dietary antioxidants may have a positive impact in reducing the oxidative stress, and thus alleviating metabolic complications.

Studies have reported high prevalence of micronutrient deficiencies in children and adolescents.<sup>[8,9]</sup> High consumption of fast foods, soft drinks and sweets, and less intake fruits and leafy vegetables<sup>[10]</sup> in adolescents are associated with obesity and may also lead to micronutrient deficiency.<sup>[11]</sup> Micronutrient deficiencies are linked to a higher risk of overweight and obesity and other debilitating

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diseases,<sup>[12]</sup> which can also have long-term consequences that include the risk of hypertension in adolescents.<sup>[13]</sup> Studies have found lower zinc status in obese children.<sup>[14]</sup> Thus, micronutrient deficiency coupled with obesity may increase the cardiometabolic risk in youth. On the other hand, antioxidant micronutrients, vitamin C, E,  $\beta$ -carotene and zinc, have been found to have a role in hypertension and MS.<sup>[15,16]</sup> Studies have yielded intriguing results suggesting the protective role of zinc against atherosclerosis.<sup>[17]</sup>

Most interventions for pediatric obesity have focused on lifestyle approaches to address the main components of energy balance. A diet and exercise module is the first line of intervention that can be used to alleviate obesity.<sup>[18]</sup> Short-term intervention trials are effective in treating the obese; however, success has not been uniform and the effectiveness tends to decline overtime.<sup>[19]</sup>

An integrated approach in the management of obesity can be devised by focusing on dietary antioxidants which are now known to have positive role in reducing the risk of adverse metabolic effects of obesity such as CVD. Unlike the effect of macronutrients,<sup>[20]</sup> micronutrients have received limited attention and sparsely so in pediatrics. Few studies in adults have reported the benefit of zinc supplementation in protecting the integrity of blood vessel cell lining.<sup>[21]</sup> Some evidences have shown short-term supplementation of antioxidants (vitamin C and folic acid) can partially or almost completely offset the alterations in arterial function in children.<sup>[22]</sup> However, the effect of dietary and nutrient intervention on arterial stiffness has been scarcely reported.<sup>[23]</sup>

We thus hypothesized that lifestyle intervention together with multivitamin-zinc supplementation would have a beneficial effect on cardiometabolic status in overweight or obese children. Thus, a pilot trial was planned to evaluate the effect of i) diet and exercise counseling alone and ii) diet and exercise counseling along with multivitamin-zinc supplementation on metabolic and atherosclerotic parameters in overweight/obese children.

## MATERIALS AND METHODS

### Study participants

Hundred overweight/obese children and adolescents in the age range of 6–17 years were enrolled from three private pediatric clinics/hospitals in Pune city, India, on a voluntary basis. Clinical examination of all children was performed by a pediatrician to assess their health status. Selection of the participants was based upon the exclusion criterion that children with any endocrine disorders other than nutritional obesity were excluded from the study. Of the

100 children, 90 (45 boys,  $11.3 \pm 2.6$  years; 45 girls,  $11.3 \pm 3.1$  years) were fulfilling the criteria of selection and were enrolled for a pilot intervention trial of 4-month duration.

Assent and written informed consent were obtained from their parents before commencement of the study. The research protocol was approved by the Ethics Committee of Hirabai Cowasji Jehangir Medical Research Institute (HCJMRI), Pune, India.

### Study design

A total of 90 overweight/obese children of mean age  $11.3 \pm 2.9$  years were randomly allocated in three groups of 30 each, using stratified randomization for age and gender: i) group A: diet and exercise counseling along with daily tablet containing multivitamins-zinc; ii) group B: diet and exercise counseling only; iii) group C received placebo. All the measurements were recorded at the baseline as well as at the end of 4 months [Figure 1]. However, the effect of intervention on atherosclerotic parameters was assessed on a subgroup only in A and B groups.

### Anthropometric measures

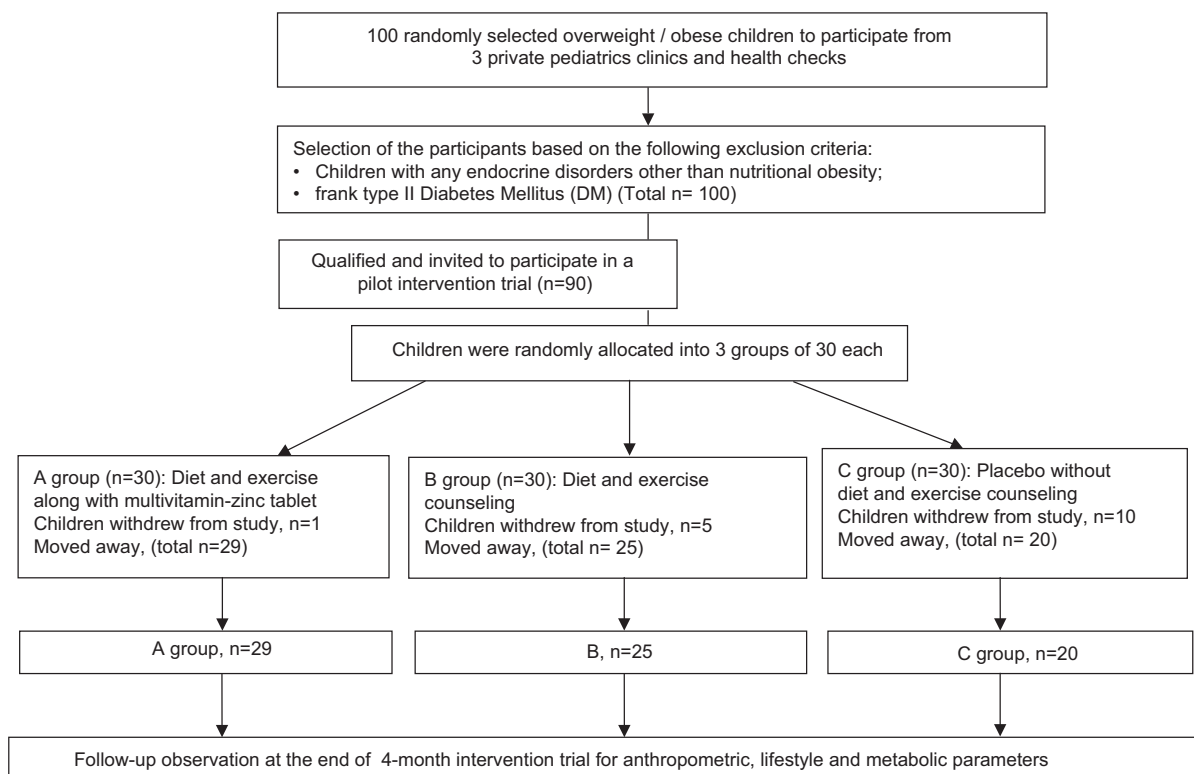
Height was measured to the nearest 0.1 cm (Leicester height meter, Child growth foundation, London, UK; range 60–207 cm). Weight was measured on an electronic digital scale to the nearest 0.1 kg (Libra Industries, Mumbai, India). Waist and hip circumferences were measured with a non-stretchable tape to the nearest 0.1 cm. Waist to hip ratio (WHR) was computed. Body mass index (BMI) was computed using the following formula:  $BMI = \text{Weight (kg)}/\text{Height (m)}^2$ . As per the Indian reference BMI cutoffs,<sup>[24]</sup> children were classified as overweight and obese. Overweight was defined as BMI  $>85^{\text{th}}$  percentile of the reference values and obesity as BMI  $>95^{\text{th}}$  percentile.

### Percent body fat by dual energy X-ray absorptiometry

Dual energy X-ray absorptiometry (DXA) measurements were performed using Lunar DPX-PRO total body pencil beam densitometer (GE Healthcare, Madison, WI, USA) using a medium mode scan (software encore 2005 version 9.30.044). The precision of repeat measurements of body fat percent in adolescents was observed to be 1.1%.<sup>[25]</sup>

### Biochemistry

A venous blood sample (8 ml) was collected at 9:00 a.m. from each child after an overnight fast of not more than 12 hours using vacutainers (BD, Franklin lakes, NJ, USA). Lipid profile of serum samples was estimated on a Dade Dimension RXL Max (Siemens Health Care Diagnostics, Deerfield, IL, USA) with enzymatic procedures for the measurement of cholesterol, triglycerides and high density lipoprotein (HDL). The low density lipoprotein (LDL)



**Figure 1:** Intervention profile. Grouping was as follows: group A: diet and exercise module with multivitamin-zinc supplementation; group B: diet and exercise module; group C: placebo

cholesterol level was calculated by using the formula  $[LDL = TC - (HDL + TG/5)]$ . Hemoglobin (Hb) was estimated using the automated cell counter (Beckman Coulter, Coulter Corporation, Miami, FL, USA). Blood glucose concentrations were analyzed enzymatically and serum insulin was measured by micro-particle enzyme immunoassay kit.

Serum levels of zinc were measured as per the method described in NIN manual.<sup>[26]</sup> Serum zinc was estimated using atomic absorption spectrometer (Perkin Elmer, Model 3110, Norwalk, CT, USA).

### Carotid arterial measurements

To study the effect of micronutrient supplementation along with lifestyle intervention on arterial parameters in overweight/obese children, carotid arterial measurements were carried out on a sub-sample from group A and B.

Measurements were performed using a Prosound  $\alpha$  10 equipment (Model SSD- $\alpha$  10; No: M00542; 2007; Aloka Co. Ltd., Tokyo, Japan) to track the vessel wall motion and automatically construct the diameter change curve in real time by a radiologist. The four physiological parameters, stiffness ( $\beta$ ), elastic modulus (Ep), arterial compliance (AC) and pulse wave velocity (PWV) of the right common

carotid artery along with intima media thickness (CIMT), were analyzed in the present study. Blood pressure was measured with a mercury-column sphygmomanometer. Average of three measurements was used as the final reading.

### Diet

Daily food intakes were evaluated through administering a standardized Food Frequency questionnaire (FFQ) by interview method.<sup>[27]</sup> Healthy foods consist of whole grains, milk, yogurt, sprouts, green leafy vegetables and fruits. Unhealthy foods include fast foods, sweets, and animal foods (chicken, fish and red meat). The number of servings and frequency of consumption were recorded for each food item. Weights of the portion sizes were calculated on the basis of average weight of each food item for that portion size representing the sample. Test-retest reliability of the questionnaire was calculated (intra-class correlation coefficient,  $r = 0.93$ ,  $P < 0.05$ ). Nutrient intakes were estimated by applying nutritive values from the database of Indian cooked foods.<sup>[28]</sup> For consumption of raw foods like fruits and salads, nutritive value tables of National Institute of Nutrition, India, were applied.<sup>[29]</sup>

### Physical activity

Physical activity was assessed by using QAPACE school

children Questionnaire,<sup>[30]</sup> which was adapted for Indian children and adolescents' lifestyle. Time spent by the participant in sports activities (yoga, walking, jogging, swimming, dancing, school sports/games and playing outdoor games) was considered as moderate activity. For inactivity, the total amount of time spent watching television, chatting and other sedentary behavior was calculated. As per the recent USDA guidelines, for health benefits, physical activity should be moderate or vigorous and add up to at least 30 minutes a day.<sup>[31]</sup>

Test–retest reliability was calculated (intra-class correlation coefficient,  $r = 0.94$ ,  $P < 0.01$  for inactivity;  $r = 0.99$ ,  $P < 0.01$  for moderate activity).

Since the groups were coded as A, B, and C, investigators were blinded for group allocation and were not aware of the type of intervention provided while performing all measurements and data analyses.

### Diet and exercise module

#### Diet

The purpose of the diet intervention was to offer food exchange for fast foods and fatty foods with micronutrient rich recipes,<sup>[32]</sup> so as to reduce fat intake and increase micronutrient intake in the diet. Thus, instead of placing the children on a strict hypo-caloric diet, considering their age and daily Recommended Dietary Allowance (RDA), a well-balanced diet in the form of charts was given to all children by trained pediatric nutritionist. Individualized innovative diet counseling was provided at the baseline and periodical monthly follow-up, which consisted of counseling the parents and attractive presentations for children.

#### Activity

In the physical activity counseling session, children were explained the importance of exercise. They were promoted to use staircase instead of lifts, encouraged to do gardening, playing outdoor games, etc. Attractive charts containing 45 minutes of physical activity program were distributed amongst children. Parents were also counseled with the help of presentations. Exercise intervention regime consisted of minimum 45 minutes of activity which included the following:

1. *Morning regime*: 10 minutes of warm up (strengthening and stretching activities to keep the muscles firm), followed by 20 minutes of vigorous activities like brisk walking, dancing, aerobics, cycling or gym-based activities, etc.
2. *Evening regime*: 20–25 minutes of playing outdoor games or any other sports.

Children were asked to restrict watching television and

playing video games so as to reduce their sedentary behavior during the intervention program.

### Micronutrient supplementation

Considering the RDA for all the vitamins and minerals as well as the toxic levels in children and adolescents, a tablet (Becosule-Z; Pfizer limited) containing zinc in the form of zinc sulfate monohydrate USP 41.4 mg equivalent to 15 mg of elemental zinc and multivitamins, B-complex forte with vitamin C, was prescribed by the pediatrician. Placebo group received a tablet which was of the same size and color as that of Becosule-Z.

Children were asked to take one tablet daily 6 days/week. In order to increase the compliance and to identify the possible side effects, all children were followed up through a monthly telephone call to their parents during the trial.

### Statistical analyses

Analyses were performed using SPSS software for Windows (version 11.0, 2001, SPSS Inc., Chicago, IL, USA). All the variables were tested for normality by the Kolmogorov–Smirnov test before any statistical comparisons were made. One-way analysis of variance (ANOVA) with *post hoc* Dunnet's test was used to test differences in means of anthropometric metabolic and dietary parameters in all the intervention groups at baseline. Paired *t*-tests were used to assess the effect of intervention. Differences in percent change for anthropometric and metabolic and dietary parameters over baseline between the three groups were tested using one-way ANOVA with *post hoc* Dunnet's test.

## RESULTS

The effects of intervention were assessed using data on 74 overweight/obese children who completed the study [Figure 1] and for whom follow-up observations were recorded. The baseline data on age, BMI, diet, activity and lipid profile of 16 children who did not complete the study were not significantly different than that of the study cohort of 74 children (data not shown).

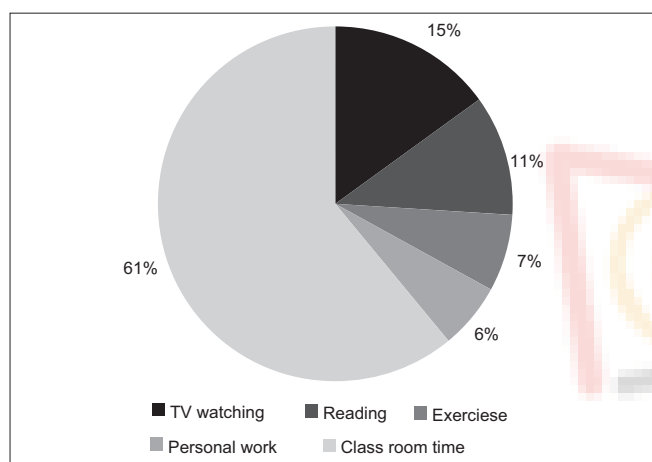
### Baseline characteristics

Mean BMI, waist and hip circumference, body fat percentage (BF%), blood lipid levels, fasting glucose and insulin were similar at the baseline in all the three groups ( $P > 0.1$ ) [Table 1] indicating comparable anthropometric and metabolic status at the start of the study. Figure 2 represents average percent time spent in daily activities by the study children. Other than classroom hours, a major share of 31% time was spent in TV viewing and reading, indicating sedentary behavior or inactivity in children. Hence, lifestyle intervention was warranted in these children.

**Table 1: Anthropometric and metabolic parameters in all the three groups**

Parameters	Group A (29)		Group B (25)		Group C (20)	
	Baseline	Post-intervention	Baseline	Post-intervention	Baseline	Post-intervention
BMI (kg/m <sup>2</sup> )	25.6 ± 0.6	24.8 ± 0.6*	24.8 ± 0.9	23.7 ± 0.8 <sup>†</sup>	27.4 ± 0.9	27.5 ± 0.9
BMI-Z score	1.7 ± 0.08	1.5 ± 0.09*	1.6 ± 0.1	1.3 ± 0.1*	1.9 ± 0.1	1.8 ± 0.1
Waist (cm)	84.9 ± 1.8	82.2 ± 2.0*	83.0 ± 2.4	80.8 ± 2.6	84.7 ± 2.5	84.6 ± 3.0
Hip (cm)	91.8 ± 2.0	90.0 ± 1.7	94.0 ± 2.7	91.8 ± 2.7	98.7 ± 3.5	96.0 ± 2.9
BF (%)	45.8 ± 0.9	43.7 ± 1.0*	43.9 ± 1.5	42.4 ± 1.6	48.3 ± 1.4	48.2 ± 1.8
LBM (%)	50.0 ± 2.8	54.9 ± 1.1 <sup>†</sup>	54.2 ± 1.6	56.0 ± 1.7 <sup>†</sup>	49.7 ± 2.0	49.2 ± 2.5
BMC (g)	1658.3 ± 101.6	1788 ± 109*	1588.6 ± 142	1639 ± 150*	1602 ± 136.2	1738 ± 225
Cholesterol (mg/dl)	181.2 ± 8.0	155.2 ± 5.0*	173.6 ± 7.6	155.3 ± 4.9*	197.2 ± 8.7	188.3 ± 8.8
TG (mg/dl)	124.3 ± 7.3	107.8 ± 9.6*	101.2 ± 7.1	95.9 ± 10.2	117.7 ± 9.7	124.4 ± 11.4
HDL (mg/dl)	50.1 ± 1.6	47.3 ± 1.8	51.5 ± 2.4	48.5 ± 2.2	47.3 ± 2.0	39.3 ± 2.6*
LDL (mg/dl)	106.5 ± 8.0	86.8 ± 5.6*	98.7 ± 6.2	90.7 ± 4.8	119.1 ± 11.6	100.3 ± 6.8
BSL (mg/dl)	90.5 ± 2.0	82.4 ± 0.9*	83.5 ± 1.6	81.1 ± 2.3	85.2 ± 1.7	89.4 ± 1.7*
Insulin (μIU/ml)	22.6 ± 5.7	18.4 ± 1.2	17.4 ± 3.0	15.6 ± 1.2	30.8 ± 9.4	35.2 ± 9.8*
Serum zinc deficiency (%)	36.7	14.3	43.2	18.2	25	22

Group A: Multivitamin-zinc with diet exercise counseling, group B: Diet and exercise counseling, group C: Placebo without diet and exercise counseling, All the values are expressed as mean ± SE, BMI: Body mass index, BF: Body fat, LBM: Lean body mass, BMC: Bone mineral content, TG: Triglyceride, HDL: High density lipoprotein cholesterol, LDL: Low density lipoprotein cholesterol, BSL: Blood sugar level, Mean values of all the parameters at the baseline were similar in the three groups ( $P > 0.1$ ), \*Indicates  $P < 0.05$ ; <sup>†</sup>indicates  $P = 0.06$  when compared with baseline values



**Figure 2:** Average time spent in all the daily activities at the baseline

Average intake of calories, proteins and fats were similar in all the three groups ( $P > 0.1$ ) [Table 2].

Mean micronutrient density of diets in children was similar in all the three groups at the baseline ( $P > 0.1$ ), except for calcium intake which was higher in group A ( $P < 0.05$ ) [Table 2].

All the atherosclerotic parameters in children of groups A and B are described in Table 3. There were no significant differences in mean CIMT, stiffness parameters and blood pressure amongst groups A and B at baseline ( $P > 0.1$ ).

### Effect of intervention

Mean BMI significantly decreased in group A ( $P < 0.05$ ) and marginally decreased in group B ( $P = 0.06$ ), whereas it was observed to be the same in group C. Additionally, to account for age differences, BMI Z-scores were

computed.<sup>[33]</sup> In both A and B groups, significant decline in BMI Z-scores was noted ( $P < 0.05$ ), indicating improvement in overweight/obese status in both A and B groups; no such change was seen in group C. Waist circumference and BF% decreased in both A and B groups; however, the mean change was significant only in group A ( $P < 0.05$ ). Moreover, waist circumference and BF% both remained the same in group C ( $P > 0.1$ ) [Table 1].

Together with a reduction in BF%, improvement was noted in lean body mass percentage (LBM%) in groups A and B, reflecting the effect of physical activity. However, the differences were marginally significant ( $P = 0.07$ ). LBM% remained the same in group C. Bone mineral content (BMC) significantly improved in groups A and B ( $P < 0.05$ ), whereas no significant change was seen in group C ( $P > 0.1$ ) [Table 1].

Furthermore, to assess the effect of intervention on body measurements, percent change in BMI, BF% and waist circumference were compared in the three groups over baseline. Percent change in BMI was of the order of  $-2.8\%$  and  $-3.7\%$  in groups A and B, respectively, which was significantly higher in comparison to group C ( $0\%$ ). Decrease in BF% was more in group A ( $-4.5\%$ ) than groups B ( $-3.4\%$ ) and C ( $0\%$ ). Percent reduction in waist circumference was higher in groups A ( $-3.3\%$ ) and B ( $-2.6\%$ ) than group C ( $0\%$ ).

Amongst the biochemical parameters, mean total cholesterol decreased significantly after intervention in groups A and B ( $P < 0.05$ ), whereas no change was observed in group C ( $P > 0.1$ ). Significant decrease was seen in triglycerides in group A ( $P < 0.05$ ); however, in group B, the decrease could

**Table 2: Mean nutritive intakes of children in all the three groups**

Parameters	Group A (29)		Group B (25)		Group C (20)	
	Baseline	Post-intervention	Baseline	Post-intervention	Baseline	Post-intervention
Dietary intake per day						
Energy (kcal)	1668 ± 105	1372 ± 104*	1603 ± 92	1381 ± 81*	1711 ± 195	1704 ± 129
Fats (g)	67.2 ± 5.3	53.6 ± 5.4*	60.8 ± 4.3	51.7 ± 3.4*	56.3 ± 8.4	66.4 ± 7.2
Protein (g)	46.1 ± 3.1	39.2 ± 3.1	39.6 ± 2.2	38.7 ± 3.0	42.8 ± 5.1	47.6 ± 4.8
Mean micronutrient density of diet in children						
Zinc (mg)	2.9 ± 0.09	3.2 ± 0.1*	2.9 ± 0.08	3.3 ± 0.1*	3.1 ± 0.2	3.2 ± 0.1
Iron (mg)	4.8 ± 0.2	4.3 ± 0.4	5.2 ± 0.2	4.9 ± 0.5	5.4 ± 0.4	7.8 ± 1.5
Copper (mg)	0.07 ± 0.002	0.7 ± 0.02*	0.07 ± 0.001	0.8 ± 0.01*	0.08 ± 0.001	0.8 ± 0.02*
Calcium (mg)	512.6 ± 46.7	602 ± 43.7*	364 ± 35	465 ± 56*	259 ± 56	459 ± 91*
β-carotene (µg)	1053 ± 48	1022 ± 71	1107 ± 93	952 ± 69	911 ± 79	966 ± 103
Thiamine (mg)	0.40 ± 0.02	0.50 ± 0.03	0.40 ± 0.03	0.43 ± 0.02	0.4 ± 0.02	0.41 ± 0.03
Riboflavin (mg)	0.40 ± 0.02	0.50 ± 0.02*	0.30 ± 0.02	0.40 ± 0.03*	0.30 ± 0.03	0.40 ± 0.05*
Niacin (mg)	5.2 ± 0.3	5.4 ± 0.3	5.7 ± 0.2	6.2 ± 0.3*	6.2 ± 0.5	5.9 ± 0.5
Folic acid (µg)	50.7 ± 1.4	54.6 ± 2.1	52.7 ± 1.5	55.4 ± 1.6*	53.2 ± 1.9	54.4 ± 1.9
Ascorbic acid (mg)	16.5 ± 0.8	12.8 ± 0.6	17.0 ± 1.1	14.8 ± 0.9	18.3 ± 1.9	16.1 ± 1.7

Group A: Multivitamin-zinc with diet exercise counseling, group B: Diet and exercise counseling, group C: Placebo without diet and exercise counseling, All the values are expressed as mean ± SE, \*Indicates  $P < 0.05$  when compared with baseline values

**Table 3: Atherosclerotic parameters in intervention groups**

Parameters	Group A (n = 14)		Group B (n = 8)	
	Baseline	Post-intervention	Baseline	Post-intervention
Intima media thickness (mm)	0.37 ± 0.01	0.38 ± 0.01	0.38 ± 0.01	0.40 ± 0.01
Stiffness index (β)	3.5 ± 0.3	3.1 ± 0.2	3.3 ± 0.4	4.1 ± 0.4
Elasticity modulus (kPa)	43.7 ± 3.3	36.4 ± 2.7*	42.9 ± 6.6	54.4 ± 5.9
Pulse wave velocity (m/s)	4.1 ± 0.1	3.7 ± 0.1*	4.0 ± 0.3	4.4 ± 0.3
Arterial compliance (mm <sup>2</sup> /kPa)	1.2 ± 0.07	1.4 ± 0.09*	1.4 ± 0.1	1.1 ± 0.1
Systolic blood pressure (mmHg)	115.0 ± 3.3	106.7 ± 2.3*	119.3 ± 5.0	124.2 ± 2.0
Diastolic blood pressure (mmHg)	75.0 ± 2.6	71.5 ± 2.0	77.9 ± 2.1	77.1 ± 1.8

Group A: Multivitamin-zinc with diet exercise counseling, group B: Diet and exercise counseling, group C: Placebo without diet and exercise counseling, All the values are expressed as mean ± SE, \*Indicates  $P < 0.05$  when compared with baseline values

not achieve level of significance, and group C showed no change [Table 1]. Mean levels of HDL cholesterol remained almost the same in groups A and B ( $P > 0.1$ ), but showed a significant decline in group C ( $P < 0.05$ ). Significant reduction was observed in the levels of LDL cholesterol and fasting blood sugar in group A only, whereas fasting blood sugar increased significantly in group C at the end of the study period. Though there was decrease in LDL cholesterol and blood sugar levels in group B, the differences were not significant. Percent children with serum zinc deficiency reduced significantly in both A and B groups ( $P < 0.05$ ). Moreover, it almost remained the same in group C [Table 1]. The reduction in zinc deficiency is attributed to supplementation and diet counseling in group A and diet counseling alone to some extent in group B.

Additionally, mean percent change in lipids over baseline was computed in order to find significant differences between the three groups. Percent change in triglyceride (-13.7%) was significantly higher in group A than in groups B (-5.9%) and C (5.7%) ( $P < 0.05$ ). Percent decrease

in total cholesterol was significantly higher in groups A (-14.3%) and B (-10.5%) than in group C (-4.5%) ( $P < 0.05$ ).

Change in activity profile after intervention in all the three groups is presented in Figure 3. Percent improvement in exercise was noted to be 82% in group A and 46% in group B, which was significantly higher than in group C (-4%) ( $P < 0.05$ ). On the other hand, TV viewing significantly reduced in groups A (-17.5%) and B (-31.4%) ( $P < 0.05$ ) and increased in group C (26%) [Figure 3].

As a result of dietary counseling, average percent consumption of healthy foods increased in children. Consumption of whole grains increased to 5% in group A and to 12% in group B, whereas it decreased to 25% in group C. Intake of green leafy vegetables increased significantly in group A (25%) ( $P < 0.05$ ), whereas there was no change in group B (-5%), and the intake decreased in group C (-50%). Similarly, increase in intake of milk and yogurt was noticed in both B and C groups (50%), whereas

it remained almost the same in group A (2.8%). Moreover, the differences were not significant ( $P > 0.1$ ). Possibly due to differences in the availability of seasonal fruits, intake of fruits decreased in all the three groups. Consumption of sprouts increased significantly in group A (84%) than B (25%) and C (-46%) ( $P < 0.05$ ) [Figure 4].

Significant percent reduction in consumption of unhealthy foods was observed in both A and B groups. Average intake of fast food decreased significantly in groups A (-54.8%) and B (-59%) in comparison to an increase (16%) in group C. Percent consumption of animal foods also decreased in all groups (-53%, -34% and -20% in groups A, B and C, respectively). The differences were significant in group A in comparison with group C ( $P < 0.05$ ). Average intake of sweet items decreased in all the three groups [Figure 5].

Mean intake of energy and fat significantly decreased in both A and B groups ( $P < 0.05$ ), but not in group C [Table 2]. A significant improvement in mean dietary

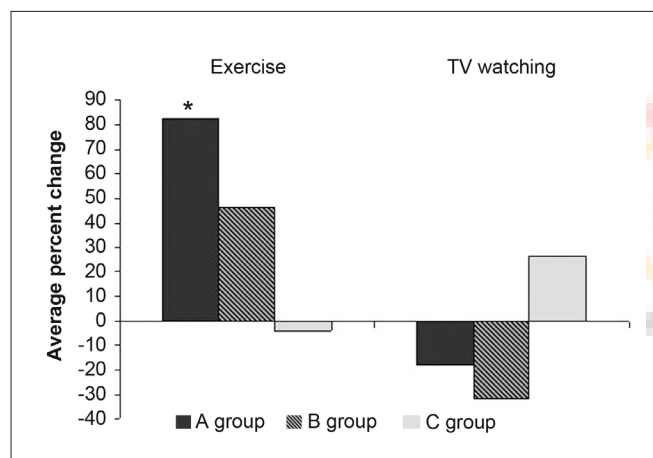
micronutrient density for zinc, copper, calcium and riboflavin was also noted in both A and B groups ( $P < 0.05$ ). Though the intake of niacin and folic acid increased in groups A and B, the differences were significant only in group B ( $P < 0.05$ ) [Table 2]. Hence, overall, the nutrient intakes improved in both A and B groups, indicating the positive effect of diet counseling in the same.

CIMT and  $\beta$  remained the same in both the groups after the intervention ( $P > 0.1$ ). Ep and PWV significantly reduced in group A ( $P < 0.05$ ). However, no such change was observed in group B. AC also significantly improved in group A in comparison to group B ( $P < 0.05$ ). Systolic blood pressure showed a significant reduction in group A ( $P < 0.05$ ) in comparison to group B [Table 3].

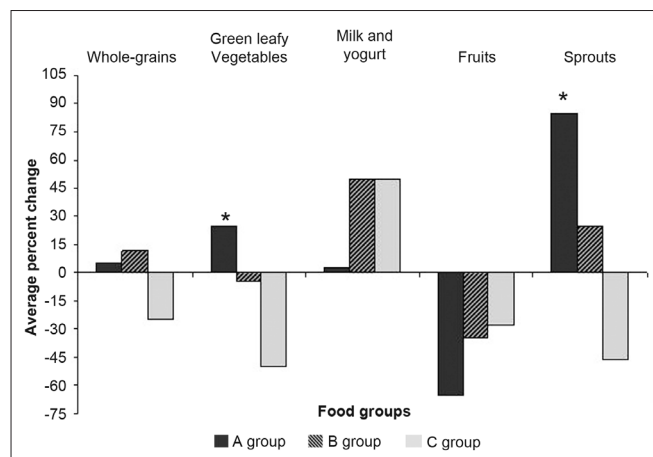
In the interim, favorable changes were observed in terms of diet, activity, anthropometry and lipid levels in both groups A and B in comparison to the placebo, demonstrating the effect of diet and exercise counseling. Moreover, additional reductions in arterial measurements in group A emphasize the importance of micronutrient supplementation in cardiometabolic health.

## DISCUSSION

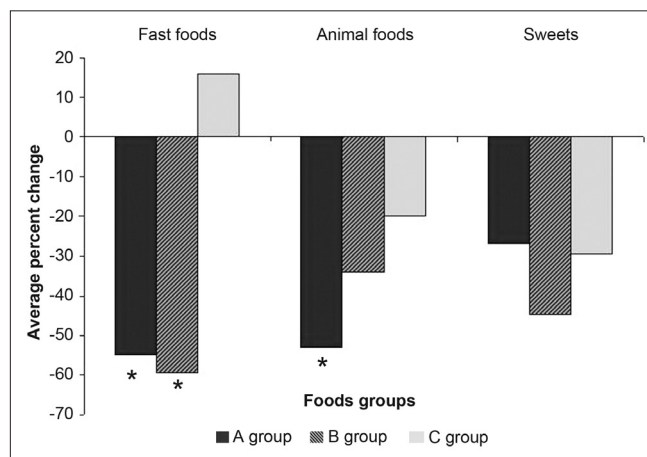
Our study demonstrated a significant improvement in BMI, waist circumference and BF% with a percent change of the order of 3–4% after lifestyle intervention as also with multivitamin-zinc supplementation for 4 months. Moreover, favorable changes were observed in arterial stiffness with multivitamin-zinc supplementation together with lifestyle than lifestyle alone. As CIMT and stiffness are known to be associated with risk of future CVD events,<sup>[34]</sup> the present study findings suggest that such supplementation may have clinical benefits in the long term.



**Figure 3:** Change in activity status after intervention (differences are explained as median percent change; \*indicates  $P < 0.05$ )



**Figure 4:** Change in the consumption of healthy foods after intervention (differences are explained as median percent change; \*indicates  $P < 0.05$ )



**Figure 5:** Change in the consumption of unhealthy foods after intervention (differences are explained as median percent change; \*indicates  $P < 0.05$ )



A similar reduction in BMI has been reported by Chen *et al.*,<sup>[35]</sup> in a short-term lifestyle intervention trial. Moreover, pronounced improvement in physical activity and television viewing was noted in our subjects, which is consistent with other interventional studies.<sup>[36,37]</sup> Also, improvement in LBM along with a reduction in BF% is a remarkable finding emphasizing the positive effect of lifestyle intervention on adiposity in both A and B groups. Reduction in waist circumference and BF% is in accordance with the finding reported by other studies.<sup>[38]</sup>

Mean concentrations of serum triglycerides, LDL and total cholesterol decreased significantly post-intervention in the present study, and our findings are in concordance with those of the studies in children administered a short-term diet and exercise regimen.<sup>[35,39]</sup>

Increased consumption of healthy foods and decreased consumption of unhealthy foods was evident in both A and B groups. Decreased fruits intake in all the three groups is possibly because of the seasonal variations and availability of fruits. Also, after counseling there was decrease in consumption of sweet fruits like mangoes, banana, sapota and figs.

Significant reduction in mean energy and fat intakes was observed in both the intervention groups, which could be likely responsible for decrease in their lipid levels. Besides, multivitamin-zinc supplementation might also have mediated the lowering of lipids in the supplementation group, in addition to lifestyle counseling.<sup>[17]</sup> Similar results were reported by Hashempour *et al.*,<sup>[40]</sup> documenting the effect of zinc supplementation on insulin resistance in pre-pubertal obese children.

Average micronutrient density of diets showed significant increase post-intervention, indicating the positive effect of dietary counseling in both A and B groups. On the other hand, amongst the atherosclerotic parameters, significant reduction in the stiffness parameters along with improvement in arterial compliance was evident only in multivitamin-zinc supplementation group, highlighting the potential benefit of multivitamin-zinc together with lifestyle change on arterial stiffness in overweight/obese children. Obesity exacerbates BP responses, whereas diet and exercise restore it. Thus, in the present study, systolic bloods pressure decreased significantly as a result of intervention. The findings are in concordance with those of other studies.<sup>[41]</sup>

The fact that atherosclerosis is a reversible step at the early ages with improvements in cardiovascular risk factors and

substantial weight reduction has been well established.<sup>[42]</sup> In an intervention trial, Mayer *et al.*,<sup>[43]</sup> have documented that regular exercise over 6 months restores endothelial function and improves CIMT, associated with an improved cardiovascular risk profile in obese children. Furthermore, Woo<sup>[44]</sup> observed some statistically significant improvement in CIMT after 1-year intervention, although the difference was small. Moreover, in the present pilot study, no such change in CIMT has been observed.

Most studies have demonstrated an association between multivitamin supplementation and change in metabolic profile in adults.<sup>[45]</sup> However, the effect of antioxidant micronutrient supplementation has been scarcely reported on arterial function in youth.<sup>[22]</sup> As antioxidant vitamins such as C, E and  $\beta$ -carotene are thought to play a role in atherosclerosis, supplementation of these nutrients has been explored as a means of reducing cardiovascular morbidity.<sup>[46]</sup> Thus, consumption of these micronutrients as nutritional supplements could be effective in improving cardiometabolic status, indeed in the presence of lifestyle changes, in children and adolescents as well.

One of the limitations of our study is the rate of drop-outs as also the absence of follow-up data on arterial measures for the control group. Thus, further intervention trials on larger samples are warranted to confirm these findings of this pilot trial.

Hence, along with lifestyle intervention, micronutrient supplementation may help additionally in improving the stiffness of the artery in overweight/obese children. In summary, our results demonstrate the efficacy of multivitamin-zinc supplementation in conjugation with lifestyle regime in young overweight individuals on attaining better health.

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