ANTIOXIDANTS ASCORBIC ACID AND ALPHA TOCOPHEROL IN RELATION TO OBESITY INDICES AND AGE SPECIFIC BMI AND WAIST HIP RATIO IN GUJARATI AND NONGUJARATI YOUNG GIRLS BEFORE AND AFTER MAIZE DIET

Nita Sahi[1,*], Ashok Kumar verma[2], Randeep Bhullar[3], Viniti Goel[4]

[1]Professor, Deptt of Biochemistry, Pacific University, Udaipur
[2]Professor & Head, Dept of Biochemistry, R.N.T. Medical College, Udaipur
[3]Reader, Dept of Biochemistry, [4]Assistant Professor, Dept of Periodontology,
Luxmi Bai Institute of Dental Sciences & Hospital, Patiala, Punjab.

***Corresponding Author:**
Emal: sahi_mukesh1973@yahoo.co.in

ABSTRACT

Aim: *The present scenario suggests that reduced antioxidant status is the key factor of obesity and hence a management strategy aiming at recommending a healthy eating plan of reduced fat, and increased fiber intake in obesity is envisaged.*

Material and method: *The present study emphasizes on an anthropometric and clinical study with maize diet in 1001 Gujarati and Non Gujarati girls, aged between 18-30 years. Every subject was asked to replace the wheat chapatti by maize chapatti for 30 days. Out of the 1001 girls, 526 girls were Gujaratis and 475 girls were Non Gujaratis. They were further distributed according to age, inhabitance, socio economic status, dietary habits, weight, height, body mass index, waist circumference, hip circumference, waist hip ratio, obesity indices, body fat percent, total body fat, lean body mass, skinfold thickness, family history and blood pressure. In the second part of the study the girls were examined for ascorbic acid and vitamin E, before and after maize diet along with the statistical evaluation of all the parameters.*

Results: *Values of vitamin-C and vitamin E were 1.14 ± 0.19mg/dl and 8.32 ± 2.72mg/l respectively before the diet, which improved to 1.17± 0.19mg/dl and 10.18 ± 2.45mg/l respectively after the maize diet for thirty days. Vitamin C and E showed significant changes (P < 0.001).*

Conclusion: *The effect of maize diet was positive on the levels of vitamin C and vitamin E in all the categories.*

Keywords: *Obesity, antioxidants, ascorbic acid, alpha tocopherol, maize diet.*

INTRODUCTION

The dramatic increase in the prevalence of obesity over the past few decades strongly suggest that preventive strategies will become more important as time goes on. Public health strategies that virtually impose behavior change along with healthy food are more successful in this regard. Long term changes in life style both in diet and in physical activity are required for weight maintenance. [1-3] Cells are protected against oxidative damage by the body's defense system. The defense system includes molecules that interact directly with the free radicals to neutralize them, (ascorbic acid, and alpha tocopherols). A diet high in whole and unrefined foods favorably alters antioxidant defense. Antioxidants have the role of counteracting free radical before free radicals arise in the body from inside and outside sources.[4,5] Behavioral and eating habits modifications to avoid some of effects of obesogenic environment are the cornerstone of long term control of weight.[6] The present study emphasizes on an anthropometric and clinical study with maize diet in 1001 Gujarati and Non Gujarati girls, aged between 18-30 years before and after maize diet. Out of the 1001 girls, 526 girls were Gujaratis and 475 girls were Non Gujaratis. They were further distributed according to age, inhabitance, socio economic status, dietary habits, weight, height, body mass index, waist circumference, hip circumference, waist hip ratio, obesity indices, body fat percent, total body fat, lean body mass, skinfold thickness, family history and blood pressure. In the second part of the study the girls were examined for antioxidants before and after maize diet along with the statistical evaluation of all the parameters.

MATERIAL AND METHOD

The present study encompasses anthropometry study as well as clinical study with Maize diet in 1001 Gujarati as well as Non Gujarati girls aged between 18 to 30 years to study the antioxidants before and after maize diet. Every subject in each group was asked to replace the wheat chapatti by maize chapatti and no change was made in the rest of the dietary ingredients. They were asked to take maize diet for 30 days. The study protocol was approved before the commencement of the study by the Institutional Ethics Committee and all the girls gave their written informed consent. The girls were residing in different institutional hostels. Actually these girls had come from different districts of Gujarat, Rajasthan mainly and also from other States like Maharashtra, Uttar Pradesh, Madhya Pradesh etc.

for study purpose, so they were selected for this study. Interview schedule was developed to collect the general information regarding age, dietary habits, socioeconomic status, suffering from any illness, or taking any treatment, caste, religion etc. The subjects with any clinical or biochemical evidence of liver, kidney or endocrine disease and those on treatment that effect the lipid metabolism were excluded from the study. The categorization of obese subjects were made on the basis of body mass index (BMI) [6,13]or waist hip ratio (WHR)[7,14,15] or standard chart of desirable weight in relation to height, published by Metropolitan Life Insurance Company (Bray,1978) [16] as well as on the basis of skin fold thickness. Normal subjects of identical age group with that of respective obese group acted as control. Measurement of different Anthropometric parameters like age, height, weight was done. The most widely used clinical tool for measurement of obesity is the Body Mass Index (BMI) i.e. wt. in Kg / m^2 height. BMI was accurately calculated using SI units Recently the Ministry's Consensus for the prevention and management of obesity and metabolic syndrome for the country has declared that the country's new diagnostic cut off for the body mass index is 23kg/m^2 as opposed to 25kg/m^2 globally [Health Ministry, Diabetes Foundation of India, All India Institute of Medical Sciences, Indian Council of Medical Research, The National Institute of Nutrition, 11/26/2008]. According to them, a person with a body mass index of 23kg/m^2 will now be considered as overweight and below that as one with normal BMI – unlike the cut off limit of 25kg/m^2 earlier. Those with BMI of 25kg/m^2 will be clinically termed obese as opposed to 30kg/m^2 at the international level, and those with BMI of 32.5kg/m^2 will require bariatric surgery to estimate excess flab. Every subject blood pressure was measured with a standardized protocol with an aneroid sphygmomanometer. Waist Circumference was measured in centimeters at the midpoint between the bottom of the ribs and the top of the iliac crest Women with a waist circumference less than 80cm is considered as normal while with 80 – 87.9 cm were classified as overweight, and women with waist circumference >88 cm were classified as obese (Park 2005). [8] Recently, the Health Ministry has declared that cut off's for waist circumference will now be 90c for Indian men as opposed to 102cm globally, and 80cm for Indian women as opposed to 88cm at the international level.[9] Hip Circumference was measured at the largest posterior extension of the buttocks. Waist Hip Ratio was determined by dividing WC by HC. WHR= WC/HC Women with a WHR less than 0.80 was categorized as normal while between 0.80 – 0.84 were classified as overweight and women with a WHR >0.85 were classified as obese (Park 2005).[10,17] For measurement of Skin fold Thickness[11,17,26] all measurements were taken, with the subject seated on a stool, on the right side of the body. The sites selected were biceps, triceps, subscapular, supra iliac. At these four sites, the skin fold was pinched up firmly between the thumb and forefinger and pulled away slightly from the underlying tissues before applying the calipers for the measurements. The average of the four sites was taken as skin fold thickness in centimeter. The instrument used was the Harpenden skin fold caliper. (British Indicators Ltd. St. Albans, Herts.); which exerts a constant pressure at varying openings of the jaws. Total body fat percent was calculated using the following formula as reported by YMCA formula (Young men Christian association).[12,26] It uses only body weight and waist (at naval) measurements to calculate body fat percentage.[13]

$$\text{Body Fat \%} = \frac{-76.76+4.15 \times \text{Waist} - 0.082 \times \text{Weight} \times 100}{\text{Weight}}$$

Body Fat was calculated by multiplying body weight (kg) with body fat percentage.[14] Lean Body Mass (LBM) was obtained by subtracting the body fat (kg) from total body weight [15.]

The venous blood samples were collected by the Standard techniques. Blood was collected in the morning after minimum of 12 hour of overnight fasting. 5 – 6 ml of blood from anticubetal vein was withdrawn in a perfectly clean dry syringe and was transferred to a clean dry centrifuge tube slowly by the side of the tube after removing needle to avoid haemolysis. All the blood samples were taken in the recumbent position. The blood was allowed to clot at room temperature for 30 minutes. The serum was separated by centrifugation at 3000 revolutions per minute (rpm) for 15 minutes. Samples with signs of haemolysis were discarded. Analytical Grade Chemicals and standards were used. The serum was preserved in refrigerator at 4^0 C. MDA was estimated in serum. The present study encompasses clinical study with Maize diet in 1001 Gujarati as well as Non Gujarati girls aged between 18 to 30 years before and after maize diet. Normal subjects of identical age group with that of respective obese group acted as control. The subjects were divided into two groups:

Control group: Possessing normal body weight with healthy body mass index between 18 -25 kg/m^2.

Study Group: Possessing overweight/obesity having body mass index between 25 to 30 kg/$m^{2.}$ Every subject in each group was asked to replace the wheat chapatti by maize chapatti and no change was made in the rest of the ingredients. They were asked to take the maize diet for thirty days. Physical and biochemical parameters were determined before and after consumption of the maize diet for thirty days, and, the effect of maize fibers was studied. The

difference in the parameters was evaluated when the two communities Gujarati and Non Gujarati were compared with each other before as well as after the maize diet. The study evaluated antioxidant enzymes in obesity and the effect of maize diet on the parameters. The parameters selected were ascorbic acid and alpha tocopherol along with the statistical analysis with t and p values.

Ascorbic acid was done according to method of Natelson,1971 [2]

Procedure:

To 0.4 ml of serum, 1.6 ml of 10 % trichloroacetic acid was added, mixed and centrifuged at 2000 rpm. To 1.0 ml of supernatant, 0.4 ml of dinitrophenyl hydrazine reagent was added, stoppered and incubated at 37°C for 3 hours. After 3 hours it was chilled in ice bath and 1.6 ml of cold 65 % H_2SO_4 was added. After 30 min the sample was read at 520 nm. Blank composed of 1.0 ml of trichloroacetic acid was treated as for serum /plasma filtrate and standard comprised 0.4 ml of 1 mg/100 ml ascorbic acid was also treated in similar fashion.

Calculation:

$$\frac{\text{Absorbance of unknown}}{\text{Absorbance of standard}} \times 1 = \text{mg Ascorbic acid/100 ml}$$

Normal Range = 0.8-1.5 mg/dl.

Alpha-tocopherol was done according to method of Baker and Frank, 1968[3]

This method is based on the reduction of the ferric chloride ions by tocopherols after xylene extraction of the blood samples. The ferrous ions react with alpha, alpha biopyridyl to give a red color which is measured in a 520 nm.

Procedure:

Into 3 stoppered centrifuged tubes measure 1.5 ml serum, 1.5 ml standard and 1.5 ml water (Blank) respectively. To Test and Blank add 1.5 ml ethanol and to the standard 1.5 ml water. Then add 1.5 ml xylene to all tubes, stopper mix well, and centrifuge. Transfer 1 ml of the xylene layers into other stoppered tubes taking care not to include any ethanol or protein. Add 1 ml alpha-alpha dipyridyl reagent to each tube, stopper and mix. Pipette 1.5 ml of the mixture into colorimeter cuvettes and read the extinction of the Teat and the Standard against the Blank at 460 nm. Then in turn beginning with the Blank add 0.33 ml ferric chloride solution, mix and after exactly 1.5 min read test and standard against the blank at 520 nm.

Calculation:

Serum tocopherol (mg/dl) =

$$\frac{\text{(Reading of unknown at 520nm-Reading of at 460nm 0.29)}}{\text{(Reading of standard at 520 nm)}} \times 1$$

Normal range: 0.8-1.5 mg/dl.

STATISTICAL ANALYSIS:

1. $$\text{Mean } (X) = \frac{\text{Sum of observations}}{\text{Total No. of cases}}$$

2. $$\text{Standard Deviation (S.D)} = \sqrt{\frac{\text{Sum +square of term-(mean)}^2}{n-1}}$$
   n=number of obs.

3. Students "t" test:
   $$T = \frac{\text{Mean 1-Mean 2}}{\text{SED}}$$

4. $$\text{SED} = \sqrt{((SE1 \times SE1) + (SE2 \times SE2)}$$

By using t values, P value was determined and if P value was more than 0.05 it is not significant, if it is less than 0.05 then it is significant, P value : a=<0.05; b=<0.01; c=<0.001. Statistical analysis was done with the help of SPSS software.

RESULTS

For antioxidant vitamins E and C, the acceptable ranges are 6-19 mg/l and 0.8- 1.5 mg/dl. Values of vitamin-C and vitamin E were 1.14 ± 0.19mg/dl and 8.32 ± 2.72mg/l respectively before the diet, which improved to 1.17± 0.19mg/dl and 10.18 ± 2.45mg/l respectively after the maize diet for thirty days. Vitamin C and E showed significant changes ($P < 0.001$). Results were almost similar for Gujarati and NonGujarati girls except that vitamin C for Gujarati, NonGujarati was P<0.05. The subjects were divided into categories like age, inhabitance, socioeconomic status, diet, weight, body mass index, waist circumference, hip circumference, waist hip ratio, obesity indices, total body fat percentage, body fat, lean body mass, skinfold thickness, family history and blood pressure and the parameters were studied along with statistical evaluation in all the categories. Nearly both the parameters showed significant changes when comparison was done before and after maize diet (P<0.001). Gujarati girls showed significant changes (P<0.05) but NonGujarati girls of same age range showed no significant change in vitamin-C, before and after the diet. When

comparison was between Gujarati and NonGujarati of this age group, statistically significant changes were observed only for vitamin C (P<0.05), after the maize diet Almost similar results were obtained for age > 20 years for total girls. However, more significant parameters as compared to the group age < 20 years were obtained when comparison was between Gujarati and Nongujaratis. The parameters which were significant before as well as after the maize diet were vitamin-C (P<0.01), vitamin E (P<0.05). Vitamin C was statistically significant for only urban girls (P<0.001). Keeping in view that obesity is a disorder mainly of affluent class; the girls were divided as lower, middle and upper socio economic class. The antioxidant for lower socio economic class was better as compared to that in upper socioeconomic class. Vitamin C, vitamin E, for lower category was 1.15± 0.19 mg/dl, 8.32, ± 2.53mg/l, v/s upper class 1.13± 0.1919 mg/dl, 8.31± 2.69mg/l. Vitamin C was significant for middle and upper socioeconomic class (P<0.05) but insignificant for lower class. As diet plays an imp role in development of obesity, subjects were divided as vegetarian and non-vegetarian. The effect of maize was almost similar for both the groups. Both parameters were highly significant in both the groups. When the antioxidant activity of lower weight category was compared which that of higher weight category the mean values of vitamin C, vitamin E, were in acceptable range for lower weight group (1.4 ± 0.11mg/dl v/s 0.92 ± 0.10mg/dl; 12.92 ± 3.1mg/l v/s 6.01 ± 0.67mg/l). Both the parameters for all three weight ranges were highly significant when matched for maize diet. As far as effect of maize diet is concerned all four groups of body mass index showed almost similar results in form of highly significant parameters (P < 0.001). There were three categories as waist circumference < 80 cm, waist circumference 80-87.9 cm and waist circumference > 88 cm and two categories of hip Circumference as < 36" and > 36". Effect of maize was significant for nearly all parameters (P < 0.001). In both the groups of hip circumference both parameters were statistically significant when matched for maize diet except vitamin C which was significant (P < 0.001) for hip circumference < 36" but insignificant for the other group. There were three groups of waist hip ratio as underweight (< 0.8), normal (0.80-0.84) and obese (>0.84). Vitamin C and vitamin E in all groups altered significantly after maize (P <0.001). Dietary effect of maize was highly significant in all categories of obesity indices (P < 0.001). Comparison of Gujarati and NonGujarati exhibited statistically significant value (P < 0.05) for them in body mass index > 25 kg/m^{2}along with vitamin E (P < 0.05).

Table 1(a): Antioxidants, Vitamin C and vitamin E in total, Gujarati and NonGujarati girls in relation to weight, BMI, W/H, and obesity indices before and after maize diet

Parameters	Total Girls (n=1001)				Gujrati Girls (n=526/1001)				Non Gujrati Girls (n=475/1001)			
	Before		After		Before		After		Before		After	
	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD
Vitamin C (mg/dl)	1.14	0.19	1.17	0.19	1.14	0.2	1.17	0.2	1.13	0.2	1.16	0.2
Vitamin E (mg/l)	8.32	2.72	10.18	2.45	8.42	2.82	10.21	2.61	8.21	2.67	10.09	2.44
WT66-80KG	Total Girls (n=251/1001)				Gujrati Girls (n= 114/251)				Non Gujrati Girls (n=137/251)			
	Before		After		Before		After		Before		After	
	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD
Vitamin C (mg/dl)	0.92	0.1	0.96	0.1	0.91	0.1	0.95	0.1	0.93	0.11	0.97	0.1
Vitamin E (mg/l)	6.01	0.67	8.01	0.67	5.96	0.66	7.96	0.66	6.05	0.67	8.05	0.67
BMI30-34.99	Total Girls (n=24/1001)				Gujrati Girls (n=10/24)				Non Gujrati Girls(n= 14/24)			
	Before		After		Before		After		Before		After	
	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD
Vitamin C (mg/dl)	0.74	0.07	0.77	0.06	0.74	0.03	0.78	0.03	0.73	0.08	0.77	0.08
Vitamin E (mg/l)	5.4	0.06	7.4	0.06	5.4	0.05	7.4	0.05	5.4	0.07	7.4	0.07
W/H>0.84	Total Girls (n=468/1001)				Gujrati Girls (n=240/468)				Non Gujrati Girls (n=228/468)			
	Before		After		Before		After		Before		After	
	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD
Vitamin C (mg/dl)	0.97	0.11	1.01	0.1	0.97	0.1	1	0.1	0.97	0.11	1.01	0.11
Vitamin E (mg/l)	6.35	0.91	8.35	0.89	6.35	0.97	8.35	0.93	6.35	0.84	8.35	0.84
high WC,WHR and BMI >25	Total Girls (n=240/1001)				Gujrati Girls (n=131/240)				Non Gujrati Girls (n=109/240)			
	Before		After		Before		After		Before		After	
	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD	Mean	±SD
Vitamin C (mg/dl)	0.91	0.09	0.95	0.09	0.91	0.09	0.95	0.09	0.9	0.09	0.94	0.09
Vitamin E (mg/l)	5.88	0.47	7.88	0.47	5.93	0.55	7.93	0.55	5.81	0.35	7.81	0.35

TABLE 1(b): Antioxidants, Vitamin C and vitamin E in total, Gujarati and NonGujarati girls in relation to weight, BMI, W/H, and obesity indices with t and P values before and after maize diet
BMI: Body mass index, W/H: Waist hip ratio

Parameters	Total Girls B vs A (n=1001)	Gujrati Girls B vs A (n=526)	Non-Gujrati Girls B vs A (n=475)	Guj vs NonGuj Girls BMD (526vs475)	Guj vs Non-Guj. Girls AMD (526vs475)
Vitamin C (mg/dl)	3.79[c]	2.42[a]	2.34[a]	0.79	0.79
Vitamin E (mg/l)	16.08[c]	10.69[c]	11.33[c]	1.21	0.75
WT66-80KG	**Total Girls B vs A (n=251/1001)**	**Guj Girls B vs A (n=114/251)**	**Non Guj Girls B vs A (n=137/251)**	**Guj vs Non Guj BMD (114vs137)**	**Guj vs Non Guj AMD (114vs137)**
Vitamin C (mg/dl)	4.12[c]	2.93[b]	2.94[b]	-1.51	-1.58
Vitamin E (mg/l)	33.6[c]	23.05[c]	24.59[c]	-1.07	-1.07
BMI30-34.99	**Total Girls B vs A (n=24/1001)**	**Guj. Girls B vs A (n=10/24)**	**Non Guj. Girls B vs A (n=14/24)**	**Guj vs Non Guj BMD (10vs14)**	**Guj vs Non Guj AMD (10vs14)**
Vitamin C (mg/dl)	2.11[a]	3.24[b]	1.28	0.43	0.43
Vitamin E (mg/l)	115.08[c]	87.89[c]	80.1[c]	0	0
W/H>0.84	**Total Girls B vs A (n=468/1001)**	**Guj Girls B vs A (n=240/468)**	**Non Guj. Girls B vs A (n=228/468)**	**Guj vs Non Guj BMD (240vs228)**	**Guj vs Non Guj AMD (240vs228)**
Vitamin C (mg/dl)	5.13[c]	3.81[c]	3.46[c]	0	-1.03
Vitamin E (mg/l)	34.01[c]	22.96[c]	25.48[c]	0	0
high WC,WHR and BMI >25	**Total Girls B vs A (n=240/1001)**	**Guj Girls B vs A (n=131/240)**	**Non Guj Girls B vs A (n=109/240)**	**Guj vs Non Guj BMD (131vs109)**	**Guj vs Non Guj AMD (131vs109)**
Vitamin C (mg/dl)	4.65[c]	3.42[c]	3.17[b]	0.86	0.86
Vitamin E (mg/l)	46.32[c]	29.38[c]	42.66[c]	2.05[a]	2.05[a]

TABLE 2(a) and 2(b): Antioxidants, Vitamin C and vitamin E in total, Gujarati and non Gujarati girls in relation to BFP, TBF, LBM, SFT, family history and blood pressure with t and P values before and after maize diet. BFP: Body fat percent, TBF: Total body fat, LBM: Lean body mass, SFT: Skin fold thickness.

BF% > 33%	Total Girls (n=201/1001)				Gujrati Girls (n= 105/201)				Non Gujrati Girls (n= 96/201)			
	Before		**After**		**Before**		**After**		**Before**		**After**	
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	0.92	0.13	0.96	0.12	0.92	0.12	0.96	0.12	0.92	0.13	0.96	0.13
Vitamin E (mg/l)	6.13	1.22	8.11	1.12	6.13	1.21	8.12	1.13	6.13	1.22	8.11	1.11
BF > 20Kg	**Total Girls (n= 270/1001)**				**Gujrati Girls (n=132/270)**				**Non Gujrati Girls (n=138/270)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	

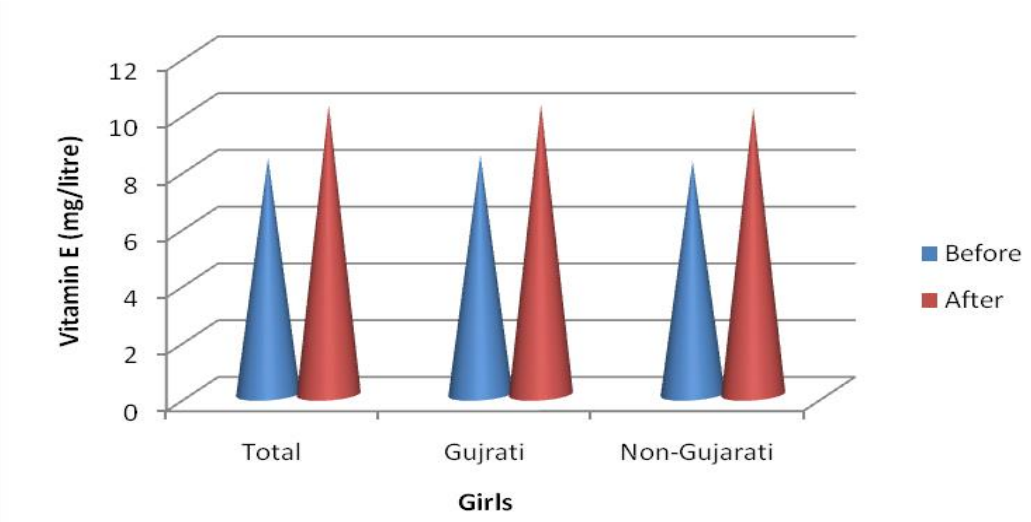
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	0.91	0.08	0.95	0.08	0.9	0.08	0.94	0.08	0.91	0.09	0.95	0.09
Vitamin E (mg/l)	5.83	0.36	7.83	0.36	5.82	0.37	7.82	0.37	5.85	0.36	7.85	0.36
LBM > 42Kg	**Total Girls (n=558/1001)**				**Gujrati Girls (n= 293/558)**				**Non Gujrati Girls (n= 265/558)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	1.06	0.17	1.09	0.17	1.07	0.18	1.1	0.18	1.04	0.17	1.08	0.16
Vitamin E (mg/l)	7.23	1.74	9.17	1.61	7.38	1.87	9.31	1.71	7.06	1.57	9.02	1.48
SFT > 51 mm	**Total Girls (n=691/1001)**				**Gujrati Girls (n= 348/691)**				**Non Gujrati Girls (n= 343/691)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	1.07	0.17	1.1	0.17	1.07	0.17	1.1	0.17	1.06	0.17	1.1	0.17
Vitamin E (mg/l)	7.43	2.06	9.36	1.88	7.44	2.02	9.38	1.84	7.41	2.1	9.35	1.92
family history of hypertension	**Total Girls (n=100/1001)**				**Gujrati Girls (n=29/100)**				**Non Gujrati Girls (n= 71/100)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	0.99	0.15	1.02	0.15	0.95	0.15	0.99	0.15	1.01	0.15	1.04	0.14
Vitamin E (mg/l)	6.61	1.21	8.6	1.17	6.35	1.14	8.35	1.14	6.72	1.22	8.7	1.17
family history of CAD	**Total Girls (n=68/1001)**				**Gujrati Girls (n= 22/68)**				**Non Gujrati Girls (n=46/68)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**± SD**	**Mean**	**±SD**	**Mean**	**± SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	0.92	0.12	0.96	0.11	0.88	0.08	0.92	0.08	0.94	0.13	0.98	0.12
Vitamin E (mg/l)	6.03	0.82	8.03	0.82	5.75	0.31	7.75	0.31	6.17	0.94	8.17	0.94
family history of diabetes	**Total Girls (n= 51/1001)**				**Gujrati Girls (n= 28/51)**				**Non Gujrati Girls (n=23/51)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	0.99	0.16	1.03	0.15	0.97	0.12	1.01	0.12	1.01	0.19	1.05	0.18
Vitamin E (mg/l)	6.65	1.58	8.63	1.49	6.38	1.05	8.38	1.05	6.98	1.99	1.05	0.18
SBP range of 121-139 mmHg	**Total Girls (n=535/1001)**				**Gujrati Girls (n=273/535)**				**Non Gujrati Girls (n=262/535)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	

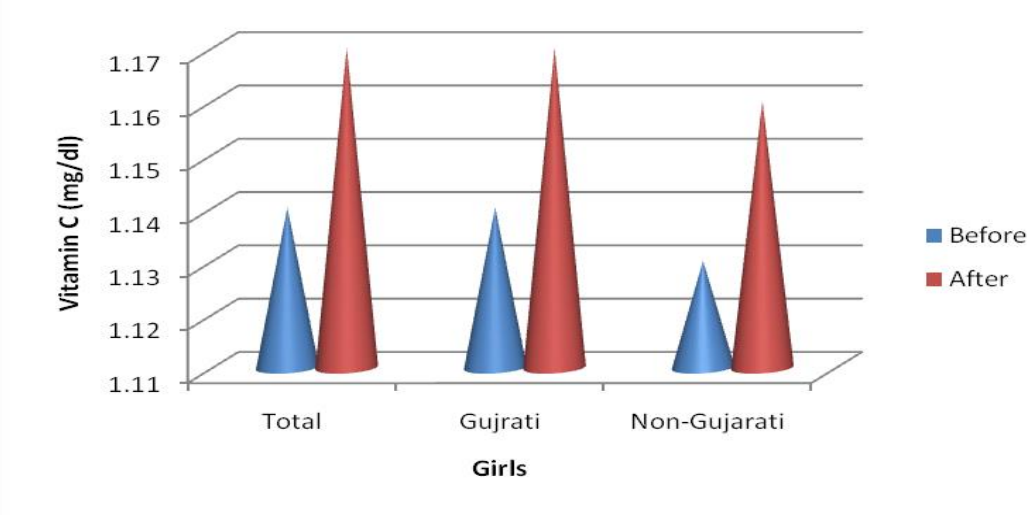
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	1.02	0.15	1.05	0.15	1.01	0.14	1.05	0.14	1.03	0.16	1.06	0.15
Vitamin E (mg/l)	6.85	1.51	8.83	1.44	6.77	1.39	8.76	1.34	6.94	1.63	8.91	1.54
DBP range of 81-90 mmHg	**Total Girls (n= 458/1001)**				**Gujrati Girls (n=237/458)**				**Non Gujrati Girls (n=221/458)**			
	Before		**After**		**Before**		**After**		**Before**		**After**	
	Mean	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**	**Mean**	**±SD**
Vitamin C (mg/dl)	1.01	0.15	1.04	0.14	1.01	0.15	1.05	0.14	1	0.15	1.04	0.14
Vitamin E (mg/l)	6.77	1.52	8.75	1.44	6.86	1.67	8.84	1.59	6.67	1.33	8.66	1.27

TABLE 2(b)

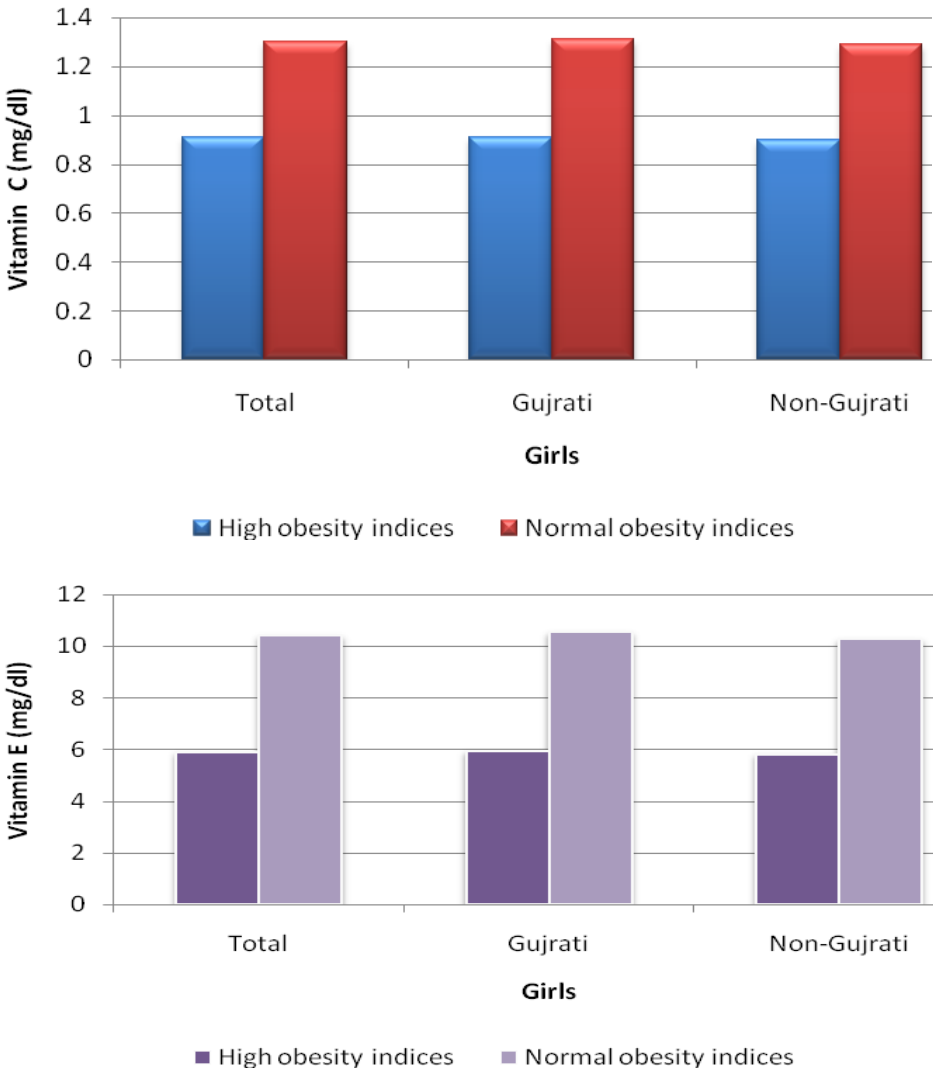
BF% > 33%	**Total Girls B vs A (n=201/1001)**	**Guj Girls B vs A (n=105/201)**	**Non Guj Girls B vs A (n=96/201)**	**Guj vs Non Guj BMD (105vs96)**	**Guj vs Non Guj AMD (105vs96)**
Vitamin C (mg/dl)	3^{b}	2.3^{a}	1.95	0	0
Vitamin E (mg/l)	17.03^{c}	12.31^{c}	11.78^{c}	0	0.06
BF > 20Kg	**Total Girls B vs A (n=270/1001)**	**Guj Girls B vs A (n=132/270)**	**Non Guj Girls B vs A (n=138/270)**	**Guj vs Non Guj BMD (132vs138)**	**Guj vs Non Guj AMD (132vs138)**
Vitamin C (mg/dl)	5.47^{c}	4.09^{c}	3.71^{c}	-0.97	-0.97
Vitamin E (mg/l)	63.98^{c}	43.88^{c}	46.76^{c}	-0.67	-0.67
LBM > 42Kg	**Total Girls B vs A (n=558/1001)**	**Guj Girls B vs A (n=293/558)**	**Non Guj Girls B vs A (n=265/558)**	**Guj vs Non Guj BMD (293vs265)**	**Guj vs Non Guj AMD (293vs265)**
Vitamin C (mg/dl)	3.32^{c}	2.3^{a}	2.42^{a}	2.02^{a}	1.39
Vitamin E (mg/l)	19.36^{c}	13.02^{c}	14.81^{c}	2.20^{a}	2.15^{a}
SFT > 51 mm	**Total Girls B vs A (n=691/1001)**	**Guj Girls B vs A (n=348/691)**	**Non Guj Girls B vs A (n=343/691)**	**Guj vs Non Guj BMD (348vs343)**	**Guj vs Non Guj AMD (348vs343)**
Vitamin C (mg/dl)	3.64^{c}	2.58^{a}	2.56^{a}	0.77	0
Vitamin E (mg/l)	18.24^{c}	13.22^{c}	12.58^{c}	0.19	0.21
family history of hypertension	**Total Girls B vs A (n=100/1001)**	**Guj. Girls B vs A (n=29/100)**	**Non Guj. Girls B vs A (n=71/100)**	**Gu vs Non Guj BMD (29vs71)**	**Guj vs Non Guj AMD (29vs71)**
Vitamin C (mg/dl)	1.67	0.93	1.42	-1.82	-1.54

Vitamin E (mg/l)	11.82[c]	6.7[c]	9.9[c]	-1.44	-1.38
family history of CAD	**Total Girls B vs A (n=68/1001)**	**Guj. Girls B vs A (n=22/68)**	**Non Guj. Girls B vs A (n=46/68)**	**Guj vs Non Guj BMD (22vs46)**	**Guj vs Non Guj AMD (22vs46)**
Vitamin C (mg/dl)	1.88	1.53	1.42	-2.34[a]	-2.44[a]
Vitamin E (mg/l)	14.21[c]	21.06[c]	10.15[c]	-2.74[b]	-2.74[b]
family history of diabetes	**Total Girls B vs A (n=51/1001)**	**Guj. Girls B vs A (n=28/51)**	**Non Guj. Girls B vs A (n=23/51)**	**Guj vs Non Guj BMD (28vs23)**	**Guj vs Non Guj AMD (28vs23)**
Vitamin C (mg/dl)	1.18	1.13	0.65	-0.88	-0.91
Vitamin E (mg/l)	6.52[c]	7.15[c]	3.45[c]	-1.3	36.3[c]
SBP range of 121-139 mmHg	**Total Girls B vs A (n=535/1001)**	**Guj Girls B vs A (n=273/535)**	**Non Guj Girls B vs A (n=262/535)**	**Guj vs Non Guj BMD (273vs262)**	**Guj vs Non Guj AMD (273vs262)**
Vitamin C (mg/dl)	3.83[c]	2.92[b]	2.53[a]	-1.54	-0.8
Vitamin E (mg/l)	21.96[c]	17.08c	14.27[c]	-1.3	-1.2
DBP range of 81-90 mmHg	**Total Girls B vs A (n=458/1001)**	**Guj Girls B vs A (n=237/458)**	**Non Guj Girls B vs A (n=221/458)**	**Guj vs Non Guj BMD (237vs221)**	**Guj vs Non Guj AMD (237vs221)**
Vitamin C (mg/dl)	3.63[c]	2.59[b]	2.55[a]	0.71	0.76
Vitamin E (mg/l)	20.23[c]	13.25[c]	16.01[c]	1.35	1.34





Graph Showing Vitamin C and Vitamin E before and after maize diet



Graphs showing level of Vitamin C and Vitamin E in Total, Gujarati and Non Gujarati girls in high and normal obesity indices

DISCUSSION

According to Aronne LJ (2002)[7], in the recent past, no other research area has generated more cajoling enthusiasm than "Antioxidants" in the modern medicine. Antioxidants come into picture because a sound antioxidant status should logically serve as a guard against obesity and overweight states.[17] Keeping this in view we have examined nutrient antioxidants, vitamin C and vitamin E.

Our results were in agreement with those of Moor de Burges,[9] who found decreased antioxidants in obese adults and those of Kuno et al [10] who found decreased levels in girls;. Our study results clearly demonstrate that obesity, in absence of other confounding factors like smoking, hypertension, diabetes mellitus, hyperlipidemia, is an independent risk factor for lipid peroxidation and depletion of cytoprotective enzymes.[18-22]

The effect of maize diet was positive on all the levels of antioxidant enzymes in all the categories. Vitamin C and vitamin E were significantly increased in all the categories of BMI (1.26 mg/dl v/s 1.29mg/dl; 9.42mg/l v/s 11.23mg/l). Effect of maize diet showed diverse effects on activities of vitamin C and vitamin E levels. They were significant for age category but insignificant for rural category. This was in accordance to study done by Olusi (2000).[8] Vitamin C and vitamin E showed significant changes (1.14mg/dl; 8.32mg/l) v/s (1.17mg/dl; 10.18mg/l) respectively after maize diet. These results were like those of Lalita Kaul[11] who used high fiber diet rich in antioxidants in treatment of obesity. There was significant weight loss. It should not be forgotten that health promoting vitamins, minerals and antioxidants are also present in dietary fiber which may be responsible for above stated beneficial effects of maize noted in clinical trials before.[23-26] The level of vitamin C and vitamin E before as well as after the diet were higher for the girls who were less than 20 years (1.15±0.19 mg/dl and 1.18±0.18 mg/dl; 8.43±2.73 mg/l and 10.30±2.47 mg/l) as compared to the girls who were more than 20 years of age (1.12±0.2 mg/dl and 1.15±0.2 mg/dl; 8.20±2.71 mg/l and 10.05±2.43 mg/l). Their levels were higher for lower socioeconomic class (1.15±0.19 mg/dl; 8.32±2.53), for rural and vegetarian girls (1.14±0.19 mg/dl and 8.36±2.55 mg/l), (1.14±0.19 mg/dl and 8.34±2.67 mg/l). Level of these vitamins was remarkably low for higher weight and body mass index categories (0.92±0.1 mg/dl and 6.01±0.67 mg/l), (0.74±0.07 mg/dl and 5.40±0.06). Waist circumference of more than 88 cm had even low levels of these enzymes (0.85±0.08 mg/dl and 5.63±0.21 mg/l). Waist hip ratio of more than 0.84 had vitamin C level as 0.97±0.11 mg/dl and vitamin E level as 6.35±0.91 mg/l. Last but not the least, the levels of these antioxidants in category of higher obesity indices were much lower than that in normal obesity indices (0.91±0.09 mg/dl v/s 1.30±0.12 mg/dl), (5.88±0.47 mg/l v/s 10.40±2.64 mg/l).

SUMMARY AND CONCLUSION

All the parameters, nutrient antioxidants like vitamin C and vitamin E were well within acceptable ranges as per ATP III guidelines in total, Gujarati and Non Gujarati girls**.** Vitamin C and vitamin E were inversely related with age (1.15 mg/dl v/s 1.12mg/dl; 8.43mg/l v/s 8.20mg/l)**.** Non vegetarian category showed low antioxidant profile when compared with the vegetarian category. Antioxidant level decreased with weight but increased with height; Antioxidant profile was least for girls with family history of CAD. Ascorbic acid level decreased with increasing waist circumference (0.96mg/dl v/s 0.85mg/dl). Maximum value of vitamin C was in the category of WC <80cm (1.21mg/dl). Alpha tocopherol also followed the same trend. It was lowest for WC >88cm, acceptable for WC 80-87.9 (6.15mg/lit) and maximum for WC <80cm. Effect of maize diet was quite evident and almost equal ($P<0.001$) on all parameters in total, Gujarati and Non Gujarati girls.. The nutrient antioxidants showed significant higher values after the consumption of the diet ($P<0.001$). Vitamin C, vitamin E were significantly increased after the maize diet was consumed for thirty days ($P<0.001$). For Gujarati girls, vitamin C levels showed significant changes ($P<0.05$), When matched for age, vitamin E, ascorbic acid was significantly increased for both the age groups. When the two groups were compared with each other, only vitamin C was statistically significant ($P<0.001$). Effect of maize was seen for all socio economic classes. Vitamin C did not increase much for middle class. Highly significant changes were observed for almost all parameters in rural and urban categories for antioxidants when matched for diet. Vitamin C was increased only for the Gujarati urban girls ($P<0.001$). For the vegetarian group of girls most of the parameters improved ($P<0.001$), For the non-vegetarian category, vitamin C did not increase for Gujaratis and Nongujaratis, vitamin E increased only for Non Gujaratis ($P<0.001$). The effect of diet was highly significant for all three categories of BMI. Antioxidant status was high for high BMI group after diet. Antioxidants which were quite low for the high WHR category, were markedly increased after the diet ($P<0.001$). Almost all parameters were significantly changed in the comparative analysis in family history of hypertension and coronary artery disease before as well as after the diet. Hence it was concluded that oxidative stress in obese was in excess while defensive antioxidant levels were depleted and that an association exists between lower antioxidants and high oxidative stress and increased risk of obesity. Free radicals play an important role in

pathophysiology of obesity and they are destroyed by antioxidant defenses. DF modulate glucose response, prevents against carbohydrate induced hypercholesterolemia and hypertriglyceridemia, has sustained energy release-part of energy released in small intestine as glucose (source of energy for brain),part is released in large intestine as SCFA (source in muscles and fat tissue), increases glucose uptake in cells, stimulate action of insulin, slows absorption of carbohydrates by causing delay in gastric emptying, increases glucagon secretion, stimulate gluconeogenesis, divert acetyl Co A to form glucose not cholesterol, protect against atherosclerosis, favorably alters lipid metabolism, decreases TG, LDL, BP, increases HDL, antioxidants, protect against chronic diseases, affect properties like volume, bulk, viscosity, in intestinal lumen, which alters metabolic path of hepatic cholesterol and lipoprotein metabolism and lowers LDL-C-increases fat oxidation, decreases fat storage, fat cell size and its synthesis, change sequence in which body burns food-brings fat on top list, restore normal intestinal functions.[22]

REFERANCES:

1. Benzie IFF and Strain JJ (1999): Ferric reducing/antioxidant power (FRAP); methods in enzymology (oxidative stress), 299:15-27.
2. Natelson S (1971): Techniques of clinical chemistry, third edition; 288,751,162.
3. Baker H and Frank O (1968): In clinical vitaminology, methods and interpretation. Interscience publishers; john villey and sons, Inc. New York. 172.
4. Wollians JA, Wrener G, Anderson PH, McMurrey CH (1983): Research in vertinary science. 34:253-256.
5. Bergman N, Van Citters GW, Mittelman SD, Dea MK, Hamilton-Wessler M, Kim SP, Ellmerer M (2001) : Central role of the adipocyte in the metabolic syndrome. J.Investig.Med. 49:119-126.
6. Sinha AK (1972): Colorimetric assay of catalase. Annal Biochem47:389.
7. Aronne LJ (2002): Classification of obesity and assessment of obesity related health risks. Obesity Research; 10: 105S-115S.
8. Olusi SO (2002): Obesity is an independent risk factor for plasma lipid peroxidation and depletion of erythrocyte cytoprotective enzymes in humans. Obesity; 26: 1159-1164.
9. Moor Montani JP, Moor D Burges, Kuns, Decri, Strauss (2004): Ectopic fat storage in heart, blood vessels and kidneys in the pathogenesis of cardiovascular diseases. *Int J Obes Relat Metab Disord*; 28 (suppl 4): S58–S65.
10. Kuno T, Hozumi M, Morinobu T, Murata T, and Mingci Z, Tamai H (1998): Antioxidant vitamin levels in plasma of obese girls. Free Radic Res; 28: 81-6.
11. Lalita Kaul and Joseph Nidiry (1993): High fiber diet in the treatment of obesity and hypercholesterolemia. Journal of National Medical Association; 85:231-232.
12. Zannad F, Gille B, Grentzinger A et al. Weight reduction. *Am Heart J. (2002)*; 144: 508–515.
13. Arcaro G, Zamboni M, Rossi L, Turcato E, Covi G, Armellini F, Bosello O, Lechi A. Body fat distribution. *Int J Obes Relat Metab Disord. (1999)*; 23: 936–942.
14. Ardern CI, Janssen I, Ross R, Katzmarzyk PT. Development of waist circumferences thresholds within BMI categories. Obes Res. (2004); 12: 1094-103.
15. Barzilai N and Gupta G. Pathophysiology of fat distribution. Ann N Y Acad Sci. (1999); 892: 58-72.
16. Bray GA. Nutrient balance and obesity. Med Clin North Am. (1989); 73.
17. Park YW, Zhu S, Palaniappan L, Heshka S, Carnethon MR, Heymsfield SB. The metabolic syndrome. *Arch Intern Med. (2003)*; 163: 427–436.
18. Pradeep Kumar, Vikas K, Kosambia JK. Prevalence of hypertension among the employs of south Gujarat. Indian Journal of Community Medicine. (2002); 27:19-22.
19. Perry AC, Applegate EB, Allison ML, Miller PC, Signorile JF. Relation between anthropometric measurements of fat distribution in overweight and obese females. Am J Clin Nutr. (1997); 66: 829-36.
20. Pi-Sunyer FX, Aronne LJ, Heshmati HM. Weight measurements. JAMA. (2006); 295: 761-75.
21. Simin Liu et al. Relation between changes in intakes of dietary fiber and grain products and changes in weight and development of obesity among middle aged women. Am J Clin Nutr. (2003); 78: 920-7.
22. Slavin J. Dietary fiber and body weight. Nutrition. (2009); 21: 411-418.
23. Voors AW, Webber LS, Frerichs RR, Berenson GS. Body height and body mass. *Am J Epidemiol. (1997)*; 106: 101–108.
24. Sharma JP, Rastogi V, Niaz MA et al. Socioeconomic status and obesity. Eur Heart J (1997); 18: 588-95.
25. Durnin JV and Rahaman MM. The assessment of the amount of fat in the human body from measurements of skin fold thickness. Br J Nutr. (1967); 21: 681.
26. Nora E, Straznicky, Christopher J, Vicki E, Wilkliams J. Obesity. (2008); 34: 580.