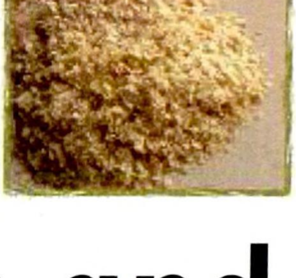

Results and Discussion

IV. RESULTS AND DISCUSSION

The results of the study entitled "Impact of supplementation of wheat germ, bran and grass on diabetic, hyperlipidemic and tuberculosis subjects" is discussed under the following headings:

A. Nutrient content of wheat germ , bran and grass
B. Background details of the subjects
C. Anthropometric measurements of the subjects
D. Food pattern and dietary practices of the subjects
E. Impact evaluation of individual and combined supplementation of wheat germ, bran and grass on diabetic, hyperlipidemic and tuberculosis subjects

A. Nutrient content of wheat germ, bran and grass

The nutrient content of wheat germ, bran and grass as analyzed is given in Table IV.

TABLE IV

NUTRIENT CONTENT (per 100 g) OF WHEAT GERM, BRAN AND GRASS

Nutrients	Wheat germ	Wheat bran	Wheat grass
Moisture (g)	11.1	9.9	12.2
Ash (g)	4.2	5.8	6.8
Carbohydrates (g)	51.8	64.5	3.1
Dietary fiber (g)	13.2	42.8	17.3
Protein (g)	23.1	15.5	20.5
Tryptophan(mg)	317	282	11
Threonine(mg)	968	500	105
Isoleucine(mg)	847	486	88
Leucine(mg)	1571	928	162
Lysine(mg)	1468	600	82
Methionine(mg)	456	24	43
Cystine(mg)	458	371	22
Phrnylalanine(mg)	928	595	108
Tyrosine(mg)	704	436	50
Valine(mg)	1198	726	125
Arginine(mg)	1867	1087	111
Histidine(mg)	643	430	45
Alanine(mg)	1477	765	137
Aspartic acid(mg)	2070	1130	222
Glutamic acid(mg)	3995	2874	242
Glycine(mg)	1424	898	117
Proline(mg)	1231	882	94
Serine(mg)	1102	684	242
Total fat(g)	9.7	4.3	0.0
Saturated fat (g)	1.7	0.6	0.0
Monounsaturated fat (mg)	1.4	0.6	0.0
Polyunsaturated fat (mg)	6.0	2.2	0.0
Vitamin C (mg)	9.2	9.5	14.1
Thiamine(mg)	1.9	0.43	0.08
Riboflavin(mg)	0.5	0.11	0.13
Niacin (mg)	6.8	4.3	0.11
Vitamin B6(mg)	1.3	1.2	0.20
Folic acid(mcg)	281	100	86
Vitamin B12(mcg)	2.3	103	99
Calcium(mg)	39	73	242
Iron(mg)	6.3	10.6	0.61
Magnesium(mg)	239	611	24
Phosphorus(mg)	842	1013	1210
Potassium(mg)	892	1182	147
Sodium(mg)	12	2.0	15.6
Zinc(mg)	12.3	7.3	0.33
Copper(mg)	0.8	1.0	0.2
Manganese(mg)	13.3	11.5	10.2
Selenium (mcg)	79.2	77.6	52.3

The moisture content of wheat germ, bran and grass were 11.1, 9.9 and 12.2g per 100g. The carbohydrate content of wheat bran was 64.5g followed by 51.8g in wheat germ and 3.1g in wheat grass. The dietary fiber content was also found to be highest in wheat bran (42.8g) followed by wheatgrass with 17.3g and wheat germ with 13.2g. Wheat germ contained 23.1g of protein followed by 20.5g in wheat grass and 15.5g in wheat bran. The essential amino to non essential amino acid ratio of wheat germ, bran and grass were 0.52, 0.43 and 0.56 respectively proving a higher ratio in wheat grass. Total fat content of wheat germ was 9.7g followed by 4.3g in wheat bran. It was interesting to note that wheat grass did not contain any fat. Further the saturated, mono and polyunsaturated fat content of wheat germ were 1.7, 1.4 and 6.0g respectively. In wheat bran the saturated and monounsaturated fat were 0.6g and 06g respectively and the polyunsaturated fat was 2.2g revealing less content of saturated fat when compared to wheat germ. Wheat grass had the highest content of vitamin C (14.1mg) followed by wheat bran with 9.5mg and wheat germ with 9.2mg.

The thiamine (1.9mg), riboflavin (0.5mg), niacin(6.8mg), vitamin B_6 (1.3 mg) and folic acid (281mcg) content were found to be highest in wheat germ. Vitamin B_{12} (103mcg) was found to be highest in wheat bran. Calcium (242mg) was found to be highest in wheat grass followed by 73mg in wheat bran and 39mg in wheat germ .Iron (10.6mg) content was maximum in wheat bran, whereas the iron content of wheat germ was 6.3mg and that of wheat grass was 0.61mg.Wheat bran provided highest content of magnesium (611mg) and wheat grass provided 24 mg of magnesium. Wheat bran contained 1182 mg of potassium followed by 892mg in wheat germ and 1210mg in wheat grass. The zinc content of wheat germ was found to be highest (12.3 mg) followed by 7.3mg in wheat bran and only 0.33mg in wheat grass. Copper 1.0mg) was found to be highest in wheat bran. Manganese and selenium were found to be highest in wheat germ with 13.3mg and 79.2mcg respectively.

B. Background details of the subjects

Age, sex, marital status, type of family and income are the factors that influence the socioeconomic profile. Background details and demographic factors play an important role on the pattern of consumption of food and nutrients (Rahman and Rao,2002).The selected subjects in diabetic, hyperlipidemic and tuberculosis group were interviewed and the details of the background information as collected are given in Table V and Figures 3 to 7.

TABLE V
BACKGROUND DETAILS OF THE SUBJECTS

Details	Diabetes mellitus						Hyperlipidemia						Tuberculosis					
	Male (N=60)		Female (N=45)		Total (N=105)		Male (74)		Female (N=31)		Total (N=105)		Male (N=38)		Female (N=22)		Total (N=60)	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
Age (years) & Sex 40-45	23	38.3	14	31.1	37	35.2	14	18.9	14	45.2	28	26.7	7	18.4	5	22.7	12	20.0
46-50	18	30.0	16	35.6	34	32.4	33	44.6	7	22.6	40	38.1	14	36.8	8	36.4	22	36.7
51-55	19	31.7	15	33.3	34	32.4	27	36.5	10	32.3	37	35.2	17	44.7	9	40.9	26	43.3
Type of family Joint	15	25.0	6	13.3	21	20.0	20	27.0	10	32.3	30	28.6	8	21.1	2	9.1	10	16.7
Nuclear	45	75.0	39	86.7	84	80.0	54	73.0	21	67.7	75	71.4	30	79.0	20	90.9	50	83.3
Educational status Illiterate	9	15.0	15	33.3	24	22.9	16	21.6	19	61.3	35	33.3	23	60.5	12	54.6	35	58.3
Primary	24	40.0	18	40.0	42	40.0	22	29.7	6	19.4	28	26.7	6	15.8	3	13.6	9	15.0
High school/Higher secondary	20	33.3	10	22.2	30	28.6	32	43.2	4	12.9	36	34.3	9	23.7	7	31.8	16	26.7
Degree	7	11.7	2	4.4	9	8.6	4	5.4	2	6.5	6	5.7	-	-	-	-	-	-
Occupational status Agriculture	38	63.3	4	8.9	42	40.0	-	-	-	-	-	-	30	79.0	5	22.7	35	58.3
Business	2	3.3	-	-	2	1.9	15	14.9	-	-	11	10.5	-	-	-	-	-	-
Government /Private job	20	33.3	3	6.7	23	21.9	59	79.7	15	48.4	90	85.7	8	21.1	-	-	8	13.3
Housewives	-	-	38	84.4	38	36.2	-	-	16	12.9	4	3.8	-	-	17	77.3	17	28.3
Income status* Low income	14	23.3	15	33.3	29	27.6	10	13.5	1	3.2	11	10.5	30	79.0	15	68.2	45	75.0
Middle income	34	56.7	24	53.3	58	55.2	45	60.8	18	58.1	63	60.0	6	15.8	5	22.7	11	18.3
High income	12	20.0	6	13.3	18	17.1	19	25.7	12	38.7	31	29.5	2	5.3	2	9.1	4	6.7
Duration of the disease <one year	7	11.7	13	28.9	13	12.4	13	17.6	6	19.4	9	8.6	38	100.0	22	100.0	60	100.0
1-3 years	27	45.0	15	33.3	45	42.9	20	27.0	11	35.5	35	33.3	-	-	-	-	-	-
4-6 years	26	43.3	17	37.8	47	44.8	41	55.4	14	45.2	61	58.1	-	-	-	-	-	-
Familial tendency** mother	23	38.3	20	44.0	43	41.0	41	55.4	23	74.2	64	61.0	3	7.9	2	9.1		
Father	46	76.7	39	86.7	85	81.0	44	59.5	5	16.1	96	91.4	2	5.2	-	-	-	-
Grand mother	8	13.3	5	11.1	13	12.4	8	10.8	15	48.4	23	21.9	-	-	-	-	-	-
Grandfather	7	11.7	5	11.1	12	11.4	16	21.6	19	61.3	35	33.3	-	-	1	4.6	1	1.7
Mode of treatment taken Unnai	2	3.3	-	-	2	1.9	1	1.4	2	6.5	3	2.9	-	-	-	-	-	-
Ayurvedha	15	25.0	10	22.2	25	23.8	31	41.9	8	25.8	46	43.8	7	18.4	5	22.7	12	20.0
Homeopathy	9	15.0	10	22.2	19	18.1	7	9.5	5	16.1	12	11.4	6	15.8	7	31.8	13	21.7
Siddha	18	30.0	11	24.4	35	33.3	13	17.6	5	16.1	8	7.6	12	31.6	5	22.7	17	28.3
Allopathy	16	26.7	14	31.1	24	22.9	22	29.7	11	35.5	36	34.3	13	34.2	5	22.7	18	30.0

* Housing Urban Development Corporation (HUDCO), (2004);** Multiple response

According to Rajeev, (1999) age is an important risk factor for diabetes. Of all the diabetic subjects selected 35.2 per cent were in the age group of 40-45 years, 32.4 per cent in the age group of 46-50 years and 32.4 per cent were in the age group of 51-55 years. Herman *et al.*,(1998) reported that the majority of the people with diabetes are in the age group of 45-64 years in the developing countries

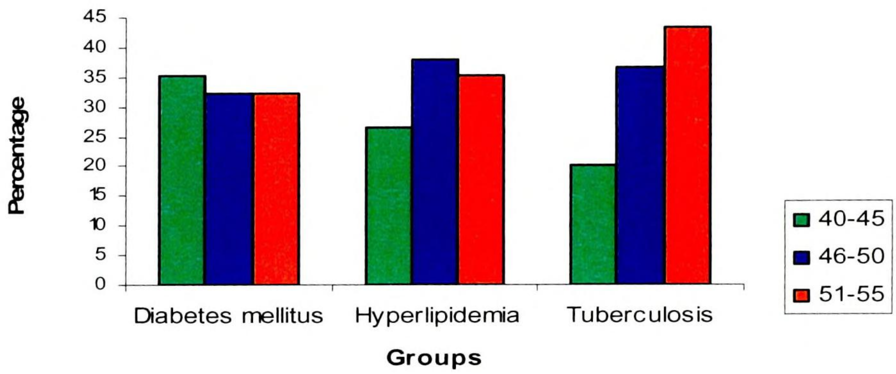

Figure 3
Agewise distribution of the subjects

Age is one of the uncontrollable causes of hyperlipidemia. As age increases the total cholesterol and LDL cholesterol increases (Kathleen, 2000). Of the selected hyperlipidemic subjects 26.7 per cent were in the age group of 40-45 years, 38.1 per cent in the age group of 46-50 years and 35.2 per cent in the age group of 51-55 years. Epidemiological studies in India have revealed that the incidence of myocardial infarction is on the increase, particularly the peak period being 41-55 years. Age is a non-modifiable risk factor for coronary heart disease. The prevalence of cardiovascular diseases increases with a doubling of rates between ages 45-54 years (American Heart Association, 2001).

In the tuberculosis group 20 per cent were in the age group of 40-45 years, 36.7 per cent were in the age group of 46-50 years and 43.3 per cent were in the age group of 51-55 years. Prevalence rate of tuberculosis rose with age. National Sample Survey conducted by ICMR (1998) described increase in prevalence rate of tuberculosis along with increase in age.

In the present study it can be noted that in the diabetic group there were 42.9 per cent women and 57.13 per cent men. Globally diabetes prevalence is similar in men and women but it is higher among men after the age of 50 years (Wild *et al.*, 2004). In the hyperlipidemic group 70.5 per cent per male and 29.6 per cent were female. The data of the present study is in par with the study of Adam, (2001) where the prevalence rate of heart disease was more prone in men than in women. According to Chadha (2005) 75 per cent of all the new pulmonary tuberculosis infection in India, are reported in men. Similarly in the tuberculosis group studied revealed that 63.3 per cent were male and only 36.7 per cent were women among the tuberculosis subjects.

Sabarwal (2005) reports that joint family system is slowly disintegrating with the advent of modern utilization. The data on type of families of the present study revealed that 20 per cent of the diabetics belonged to joint family system and a majority of 80 per cent were in the nuclear type of family system.

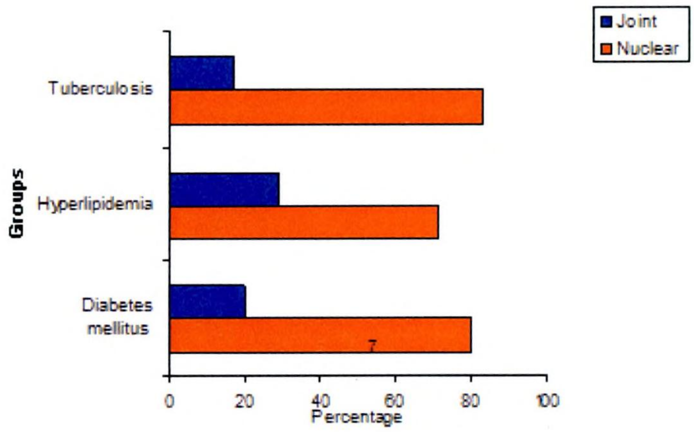

Figure 4
Type of distribution of families

In the hyperlipidemic group 28.6 and 71.4 per cent of subjects belonged to the joint and nuclear family type respectively. Tuberculosis group also showed the same trend where 83.3 per cent of tuberculosis subjects belonged to the nuclear family system and only 16.7 per cent of the tuberculosis subjects lived in the joint family system. This observation is in accordance with the reports published that more than 80 per cent of the families in India are of nuclear family type (Zimmet *et al.*,2001).

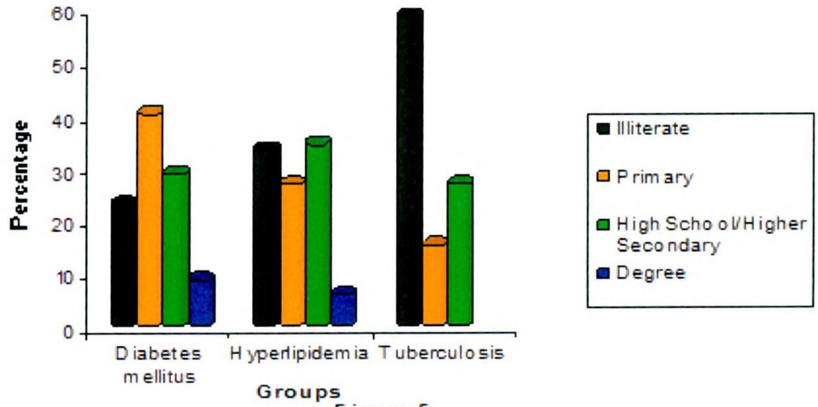

Figure 5
Educational status of the subjects

Among the selected diabetics 40 per cent of the subjects had primary education, 28.6 per cent had completed high school / higher secondary education and only 8.6 per cent were degree holders. In the hyperlipidemic group, 26.7 per cent had completed primary education, 34.3 per cent had completed high school /higher secondary education and only 5.7 per cent were degree holders. Among the tuberculosis subjects 15 per cent had primary school education, 26.7 per cent had high school/higher secondary education and none of them were degree holders. In the diabetic, hyperlipidemic and the tuberculosis groups it was noted that 22.9, 33.3 and 58.3 per cent were illiterates. Illiteracy predisposes the adults to many infections and metabolic diseases and lack of awareness augments spread of the disease (WHO,2007).

In the diabetic group, 40 per cent were agricultural landlords and only 1.9 per cent was doing business. Further 21.9 per cent were either in Government or private jobs. In the hyperlipidemic group 85.7 per cent were government /private job holders and only 10.5 per cent constituted the agriculture group. Advent of modernization has resulted in low physical activity; these conditions are conducive for the onset of metabolic diseases (Vijay, 2005).

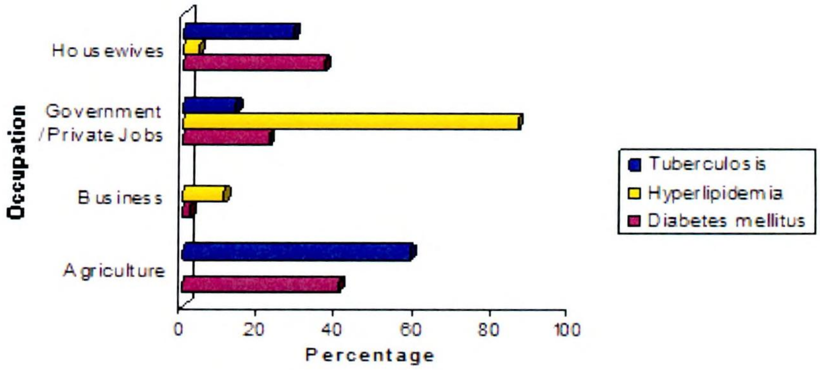

Figure 6
Ooccupational status of the subjects

In the tuberculosis group 58.33 per cent were agriculturists and only 13.3 per cent were either in government or private jobs. This phenomenon of higher prevalence of tuberculosis in agriculture workers and labourers may be ascribed to poor living condition. Higher prevalence rate (61.88/1000) was observed in lower socioeconomic class (Khan ,2006).

As with the regard to income status, subjects in diabetes mellitus, hyperlipidemia and tuberculosis groups were categorized according to the HUDCO (2004) classification. It was found in the diabetic group 27.6 per cent belonged to low income group (Rs.2,101-7,500),17.1 per cent belonged to high income group (Rs.>7500/-) and a majority of 55.2 per cent belonged to the middle income group (Rs.4,501-7,500). According to Dudeja (2001) prevalence of diabetes was lower among those with low income that among the high income group. When income level raises people change their lifestyle pattern and dietary habits leading to diet related disorders.

The present observation is in line with the reported finding. In the hyperlipidemic group 60 per cent of the subjects belonged to middle income group (Rs.4,501-7,500) and 29.5 per cent belonged to the high income group (Rs.>7500/-). Only 10.5 per cent of the subjects belonged to the low income group(Rs.2,101-7,500). In developing countries, high-socioeconomic status is associated with higher risk of cardiovascular diseases and diabetes (Davey , 1997).Indeed cardiovascular diseases are even more numerous in India and China than in all economically developed countries in the world. Chronic diseases are emerging at a faster rate in developing countries (Baker, 1999).

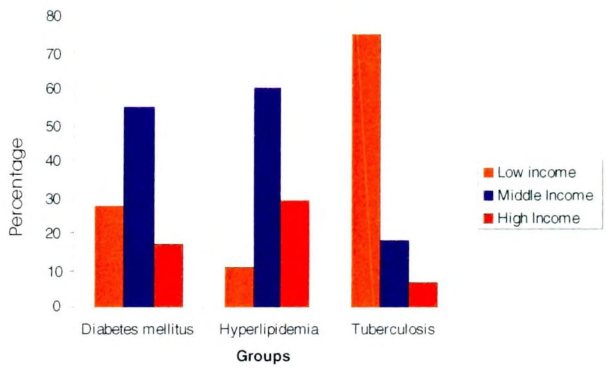

Figure 7
Income status of the subjects

In the tuberculosis group 75 per cent belonged to the low income group, 18.3 per cent belonged to the middle income group and only 6.7 per cent belonged to the high income group. The more prevalence of tuberculosis in low socio-economic class might be due to ignorance, poverty and closed proximity of positive cases in vicinity as well as within the family (Khan.,2006).

Duration of the disease as expressed by the diabetic subjects revealed that 58.6 per cent had the condition over four to six years, 42.9 per cent of the subjects had diagnosed within the past 1-3 years and only 12.4 per cent of them had diagnosed the condition only during the past one year. In the hyperlipidemic group it could be noted that 58.1 per cent had diagnosed the condition before four to six years while 33.3 per cent of the subjects had the condition over one to three years and only 8.6 per cent of the hyperlipidemic subjects had diagnosed the condition only during the past one year. All the 60 tuberculosis subjects had diagnosed the infection within one year.

Of the 62 per cent diabetic population in India having a positive family history of diabetes, 53 per cent have their first degree relatives as diabetics (Ramachnadran, 1992). In accordance with this, among the selected 105 diabetic subjects 41 per cent of the mothers of the selected subjects were diabetic, 81 per cent of the fathers of the selected subjects were diabetic. Further it was also noted that 12.4 and 11.4 per cent of the grand mothers and grand fathers respectively were found to be diabetic. In the hyperlipidemic group 61 and 91.4 per cent of the mothers and fathers respectively of the selected subjects had heart disease, 21.9 and 33.3 per cent of the grand mothers and grand fathers of the selected subjects had heart disease. A family history of premature disease is strong risk factor, even when other risk factors are considered. A family history is considered to be positive when myocardial infarction and sudden death occurs before age of 55 in male first degree relative. Numerous types of hyperlipidemia are inheritable and lead to premature atherosclerosis and coronary heart diseases (Wood, 2001).

Among the selected diabetic subjects 1.9, 23.8, 18.1, 33.3 and 22.9 per cent respectively had taken unnai,allopathic, ayurvedic, homeopathic, siddha and allopathic treatment and in the hyperlipidemic group, 2.9,43.8,11.4,7.6 and 34.3 per cent were on unnai , allopathic, ayurvedic, homeopathic, siddha and allopathic treatment.

Regarding the mode of treatment undertaken by the subjects before getting admitted to the tuberculosis center revealed that 21.7,16.7, and 35 per cent respectively were taking ayurvedic, homeopathic and siddha treatment. Only 20 per cent were undergoing allopathic treatment.

C. Anthropometric measurements of the subjects

The mean anthropometric measurements of the selected subjects are presented in Table VI.

TABLE VI

MEAN ANTHROPOMETRIC MEASURMENTS OF DIABETIC, HYPERLIPIDEMIC AND TUBERCULOSIS SUBJECTS

Groups	Weight (Kg)*		BMI•		Wait to hip ratio♦		MUAC (cm)		Skin fold thickness (cm)	
	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female
Diabetic	69.98	68.55	25.81	26.21	0.94	0.92	-	-	-	-
Hyperlipidemic	71.25	65.99	25.35	26.85	0.95	0.93	-	-	-	-
Tuberculosis	41.23	40.37	17.62	17.92	0.62	0.71	23.22	22.38	1.7	1.6

Standard: * Male: 60Kg; Female: 50Kg (ICMR, 2002)

• Normal: 20-25; grade I: 25-30; grade II :> 30 (Brahmam *et al.*,2005)

♦ 0.8 (Brahmam *et al.*,2005)

The mean weight of the diabetic male and female subjects were 69.98 and 68.55kg respectively. In the hyperlipidemic group the mean weight of the male and female subjects were 71.25 and 65.99 kg and in the tuberculosis group it could be noted that the male and female subjects weighed only 41.23 and 40.37kg respectively. The standard reference weight as recommended by ICMR (2004) was 60 and 50kg for male and female respectively. As compared with the reference standard, it would be revealed that the diabetic and hyperlipidemic male and female subjects were well above the normal standards confirming overweight as the risk factor for diabetes and hyperlipidemia (Hennekens *et al.*,2004). In the tuberculosis group it could be noted that the male and female subjects were below the normal standards proving the muscle wasting in tuberculosis.

The Body Mass Index determined for the diabetic male and female subjects were 24.81 and 26.21 respectively. In case of hyperlipidemic subjects the BMI of males was 25.35 and that of the females was 26.85. In case of tuberculosis subjects BMI for male and female subjects were 17.62 and 17.92 respectively. From the classification of BMI according to Brahmam *et al.*,(2005) it could be noted that the diabetic and hyperlipidemic male and female subjects were in the Grade I obesity (25-30) and the tuberculosis male and female subjects were in mild Chronic Energy Deficiency (CED)-grade I. According to Roulant (2004) in the urban Indian population BMI of >23 showed the risk of diabetes in both genders. Rabkin *et al*.,(1997) revealed that the prevalence of dyslipidemia is related to BMI as LDL-C and triglyceride concentration were higher in those with higher BMI.

The Waist Hip Ratio (WHR) of the male and female diabetic subjects were 0.94 and 0.92 respectively. In the hyperlipidemic group the WHR of the male and female subjects were 0.95 and 0.93 respectively. Further in the tuberculosis group WHR was 0.62 for male and 0.71 for female subjects. The normal standard values for WHR as recommended by Brahmam *et al*.,(2005) is 0.8 and it could be noted from the Table that both the diabetes and the hyperlipidemic group subjects were well above the normal standard values. Jerum (2003) opined that WHR is an indicator of abdominal obesity which indirectly leads to diabetes. Krause (2002) revealed that subjects with abdominal obesity are prone to cardiovascular disease and also pointed out the precautionary measure to avoid cardiovascular disease by avoiding abdominal obesity.

Mid upper arm circumference and skin fold thickness are related to the fatness or weight of an individual and these two measurements were measured on tuberculosis subjects. The MUAC of male and female subjects were 23.22 and 22.38 respectively and that of the skin fold thickness of the male and female subjects were 1.7 and 1.6 cm. Jahon *et al*.,(2002) explain that reduced food intake , nutrient malabsorption and altered protein and lipid metabolism are mechanisms that appear to contribute to loss of lean body mass.

D. Food pattern and dietary practices of the subjects

1.Food and nutrient intake of diabetic subjects

a. Food intake pattern

Table VII and Figure 8 present the data on the food intake by the diabetic subjects.

TABLE VII

FOOD INTAKE PATTERN OF DIABETIC SUBJECTS

Food stuff (g)	Actual intake	Per cent adequacy	Suggested allowance*
Cereals	226	+132.9	170
Pulses	46	-76.7	60
Green leafy vegetables	16	-80	200
Other vegetables	79	-39.5	200
Fruits	48	-48	100
Milk and milk products	416	+138.7	300
Fats and oils	39	+260	10-15

* Raghuram *et al*., (2007)

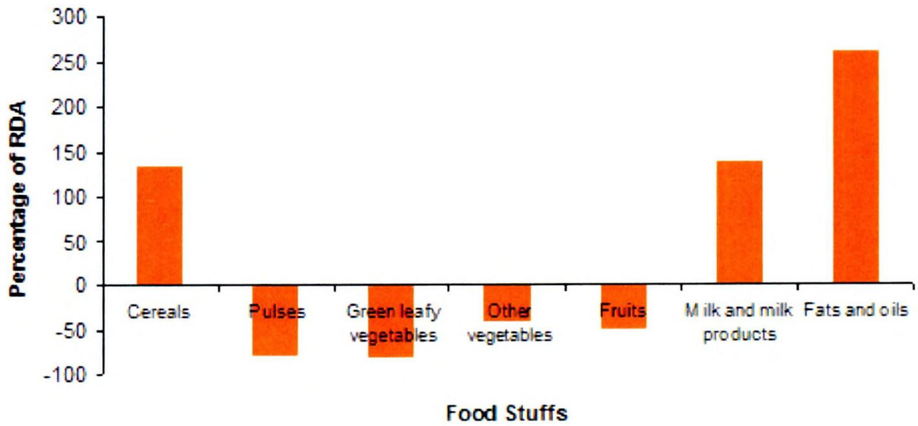

Figure 8
Percentage Adequacy of food intake of diabetic subjects

Consumption of cereals, milk and milk products and fats and oils were found to be excess and the percentage adequacy of these food stuffs included +132.9 per cent,+138.7 per cent and +260 per cent respectively. The consumption of pulses, green leafy vegetables and fruits were found to be in deficit when compared against the suggested allowance given by Raghuram *et al*.,(2007) and the percentage inadequacy of pulses , green leafy vegetables, other vegetables and fruits were 76.7, 80,39.5 and 48 per cent respectively.

b. Nutrient intake

Table VIII presents the data on the nutrient intake and contribution of carbohydrate, protein and fat to per cent energy intake by the diabetic subjects .

TABLE VIII

NUTRIENT INTAKE OF DIABETIC SUBJECTS

Nutrients	Actual intake	Suggested allowance*
Energy (Kcal)	1688	1800
Carbohydrate (g)	251	245.50
Total fat (g)	51	30.2
Protein (g)	56.75	59.5
Dietary fiber (g)	39.37	34

*Raghuram *et al.*, (2007)

The mean energy intake of the diabetic subjects was 1688 Kcal as against the suggested allowance of 1800 Kcal. The carbohydrate and fat content of the diabetic subjects was 251 and 51g respectively. The protein content of the diabetic subjects was 56.8g as against the suggested allowance of 59.5g and the dietary fiber content was 39.4g as against the suggested allowance of 34g. The high energy value of diets of diabetics was mainly due to high intake of fat and oils and milk and milk products.

2. Food and nutrient intake of the hyperlipidemic subjects

a. Food intake pattern

Table IX and Figure 9 present the data on the food intake by the hyperlipidemic subjects .

TABLE IX
INTAKE PATTERN OF HYPERLIPIDEMIC SUBJECTS

Food stuff (g)	Actual intake	Per cent adequacy	Suggested allowance*
Cereals	250	+178.57	140
Pulses	59	+147.50	40
Green leafy vegetables	20	-13.33	150
Other vegetables	45	-45.00	100
Fruits	25	-25.00	100
Milk and milk products	450	+225.00	200
Fats and oils	45	+450	10

* Ghafoorunissa and Krishnasamy (2007)

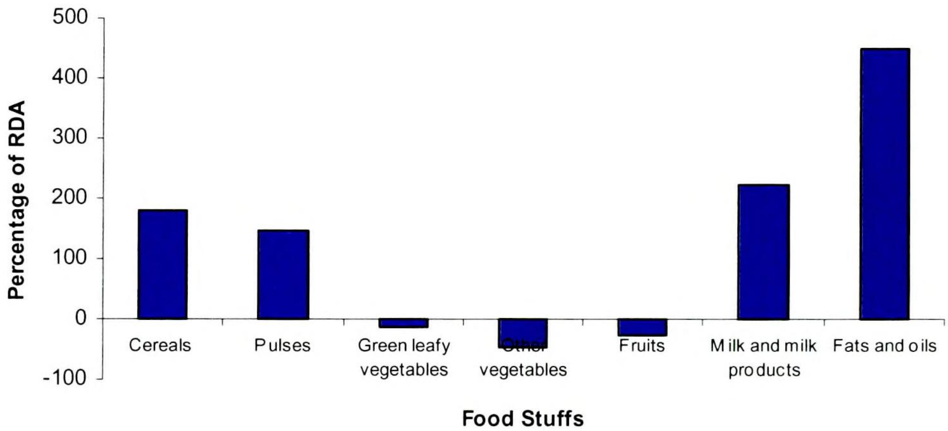

Figure 9
Percentage adequacy of food intake of hyperlipidemic subjects

Consumption of cereals, milk and milk products, fats and oils were found to be excess by 178.6,147.5,225 and 450 per cent respectively than the suggested values given by Ghafoorunissa and Krishnasamy (2007). The consumption of green leafy vegetables (13.3%), other vegetables (45%) and fruits (25%) were inadequate when compared with the suggested values of Ghafoorunissa and Krishnasamy (2007).

2. Nutrient intake

Table X presents the data on the nutrient intake by the hyperlipidemic subjects .

TABLE X

NUTRIENT INTAKE OF HYPERLIPIDEMIC SUBJECTS

NUTRIENTS	Actual intake	Suggested allowance*
Energy (Kcal)	2153	1800
Carbohydrate (g)	265	245.50
Total fat (g)	65	15.00
Protein (g)	53.26	59.5
Dietary fiber (g)	31.56	34

* Ghafoorunissa and Krishnasamy., (2007)

The average energy intake of the hyperlipidemic subjects was 2153 Kcal as against the suggested values of 1800 Kcal. Similarly carbohydrate (65g) was also found to be in excess. Consumption of protein and dietary fiber was only 53.3 and 31.6g when compared with the suggested allowance of 59.5 and 34g when compared with suggested values given by Ghafoorunissa and Krishnasamy (2007).

3. Food and nutrient intake of the tuberculosis subjects

a. Food intake pattern

Table XI and Figure 10 present the data on the food intake by the tuberculosis subjects.

TABLE XI
FOOD INTAKE PATTERN OF TUBERCULOSIS SUBJECTS

Food stuff (g)	Actual intake	Per cent adequacy	Suggested allowance*
Cereals	200	-47.6	420
Pulses	25	-27.8	90
Green leafy vegetables	25	-25	100
Other vegetables	40	-40	100
Fruits	10	-10	100
Milk and milk products	100	-25	400
Fats and oils	15	-75	20

*WHO (2003)

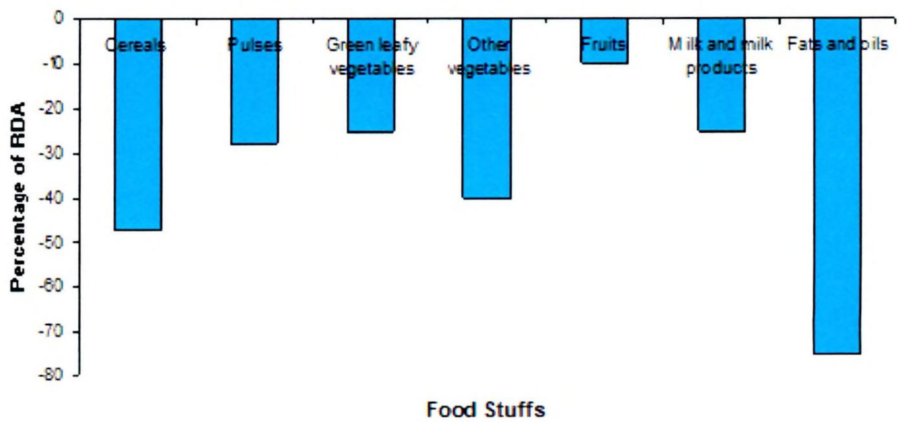

Figure 10
Percentage adequacy of food intake by tuberculosis subjects

Consumption of all foodstuffs including cereals, pulses, green leafy vegetables, other vegetables, fruits, milk and milk products and fats and oils were well below the suggested values of WHO (2003). A deficit of 27.8 per cent was observed in pulse consumption and 25 per cent deficit was observed in the milk and milk product consumption. Consumption of green leafy vegetables was at a deficit of 25 per cent and that of cereals was 47.6 per cent.

2. Nutrient intake

Table XII presents the data about the nutrient intake by the tuberculosis subject .

TABLE XII

NUTRIENT INTAKE OF TUBERCULOSIS SUBJECTS

Nutrients	Actual intake	Suggested allowance*
Energy (Kcal)	1756	2910
Carbohydrate (g)	159	290
Total fat (g)	20	30.2
Protein (g)	36	69

*WHO (2003)

The average energy intake of the tuberculosis subjects was 1756Kcal. The low energy value of diets of tuberculosis was mainly due to low intake of protein and carbohydrate intake which was only 36 and 159g respectively. The intake of carbohydrate (159 g) and fats (20g) was found to below the suggested values when compared with suggested values given by WHO (2003) .

E. Impact evaluation of individual and combined supplementation of wheat germ, bran and grass on diabetic, hyperlipidemic and tuberculosis subjects

1. Effect of supplementation on diabetic subjects

a. Changes in the physiological symptoms

The changes in the physiological symptom before and after supplementation of the diabetic subjects is depicted in Table XIII.

TABLE XIII
CHANGES IN PHYSIOLOGICAL SYMPTOMS OF DIABETEIC SUBJECTS

Symptoms*	Group DA		Group DB		Group DC		Group DD		Group DE		Group DF		Group DG	
	I	F	I	F	I	F	I	F	I	F	I	F	I	F
Polydypsia	10	1	12	2	10	1	11	2	10	-	10	1	9	9
Polyphagia	9	-	14	-	15	-	15	-	15	-	11	2	10	10
Polyuria	8	-	13	-	13	-	14	-	11	-	12	-	13	12
Nocturia	10	-	14	1	10	-	13	1	10	1	13	-	10	10
Constipation	12	-	10	-	9	-	10	-	15	-	10	-	5	5
Insomnia	-	-	1	1	-	-	-	-	1	1	-	-	-	-
Giddiness	-	-	-	-	-	-	-	-	-	-	-	-	2	2
Impaired vision	-	-	2	2	-	-	-	-	-	-	-	-	-	-

* Multiple responses I-Initial ; F-Final

Of the various clinical parameters which indicate diabetes, polyuria, polydypsia, polyphagia, nocturia and constipation were found to be the most frequently occurring symptoms in all the groups studied. In the initial phase it was found that 10,12,10,11,10,10,and nine subjects in groups DA,DB,DC,DD,DE,DF and DG expressed the occurrence of polydypsia and 9,14,15,15,11 and 10 subjects of the groups DA,DB,DC,DD,DE,DF and DG expressed polyphagia as a symptom. Around 10 to 15 subjects in the entire supplemented groups were suffering from constipation initially. After six months of supplementation with wheat germ, bran and grass individually and in combination there was a drastic reduction in the physiological symptoms expect for one or two subjects in whom symptoms like polyuria,polyphagia, nocturia and polydypsia were reduced. Further it was found that the subjects who expressed constipation as a problem expressed the relief of constipation after the supplementation. This can be owed to the high amount of soluble fiber in the supplements. As observed from the Table, there was not much improvement in the physiological symptoms in the control group. Many in the experimental groups studied desired to continue the supplementation even after the completion of the study due to the improvement shown in the various physiological symptoms.

b. Changes in biochemical picture

(i) Mean serum fasting and postprandial glucose and glycosylated hemoglobin levels

Tables XIV and Figures 11, 12 and 13 give the mean serum fasting and postprandial glucose and glycosylated hemoglobin levels at the initial and final phase of the study period and results of the statistical analysis.

TABLE XIV
CHANGES IN MEAN SERUM FASTING AND POSTPRANDIAL GLUCOSE AND GLYCOSYLATED HEMOGLOBIN LEVELS

(N=15/group)

Groups	Fasting Glucose (mg/dl)	Post prandial Glucose (mg/dl)	Glycosylated hemoglobin (%)
Group DA Initial	123.20 ±2.62	164.67 ±3.08	8.42 ±0.17
Final	101.13 ±0.92	123.93 ± 2.63	6.35 ±0.22
Difference	-22.07 ±2.91	-40.74 ± 4.20	-2.07 ±0.29
't' value	29.32**	37.17**	27.79**
Group DB Initial	123.60 ±2.72	164.60 ±2.61	8.37 ±0.14
Final	100.80 ±1.15	124.80 ±2.11	6.41 ±0.18
Difference	-22.80 ± 3.03	-39.80 ± 3.88	-1.96 ±0.25
t' value	29.16**	39.76**	30.85**
Group DC Initial	123.47 ±2.75	163.40 ±2.32	8.40 ±0.21
Final	101.40 ±1.77	115.47 ±1.88	6.41 ±0.10
Difference	-22.07 ±3.28	-47.93 ±3.17	-1.99 ±0.10
t' value	26.03**	38.12**	33.08**
Group DD Initial	123.80 ±2.37	163.67 ±2.06	8.39 ±0.28
Final	100.53 ±1.36	115.47 ±1.88	5.49 ±0.17
Difference	-23.27 ±2.71	-48.20 ±3.17	-2.90 ±0.17
t' value	42.05**	58.95**	30.17**
Group DE Initial	124.20 ±2.65	165.94 ±1.67	8.45 ±0.17
Final	95.87 ±1.46	113.07 ±1.62	5.26 ±0.12
Difference	-28.33 ± 2.71	-52.87 ±2.90	-3.19 ± 0.12
t' value	36.12**	70.61**	56.11**
Group DF Initial	123.47 ±2.53	165.67 ±1.95	8.41 ±0.15
Final	100.01 ±1.63	114.20 ±1.47	5.34 ±0.11
Difference	-23.46 ±2.29	-51.47 ±2.53	-3.07 ±0.11
't' value	39.53**	78.73**	64.59**
Group DG Initial	124.80 ±2.54	163.07 ±2.34	8.39 ± 0.21
Final	124.20 ±2.65	164.60 ±2.61	8.36 ±0.13
Difference	-0.60 ± 3.46	1.53 ±3.74	-0.03 ±0.13
't' value	0.65 NS	1.53 NS	0.65 NS
'F' value between groups	140.29**	409.83**	538.94**

** Significant at one per cent level; NS not significant

TUKEY VALUES FOR SERUM FASTING BLOOD GLUCOSE

Groups	Means in ascending order	Group DG	Group DA	Group DC	Group DB	Group DD	Group DF	Group DE
		0.60	**22.07**	**22.07**	**22.80**	**23.27**	**23.46**	**28.33**
DG	**0.60**	-	21.47	21.47	22.20	22.67	22.86	27.73
DA	**22.07**		-	0.00	0.73	1.20	1.39	6.26
DC	**22.07**			-	0.73	1.20	1.39	6.26
DB	**22.80**				-	0.47	0.66	5.53
DD	**23.27**					-	0.19	5.06
DF	**23.46**						-	4.87
DE	**28.33**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=3.27$

TUKEY VALUES FOR SERUM POSTPRANDIAL BLOOD GLUCOSE

Groups	Means in ascending order	Group DG	Group DB	Group DA	Group DC	Group DD	Group DF	Group DE
		1.53	**39.80**	**40.74**	**47.93**	**48.20**	**51.47**	**52.87**
DG	**1.53**	-	38.27	39.21	46.40	46.67	49.94	51.34
DB	**39.80**		-	0.94	8.13	8.40	11.67	13.07
DA	**40.74**			-	7.19	7.46	10.73	12.13
DC	**47.93**				-	0.27	3.54	4.94
DD	**48.20**					-	3.27	4.67
DF	**51.47**						-	1.40
DE	**52.87**							-

alpha =0.5;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=3.80$

TUKEY VALUES FOR SERUM GLYCOSYLATED HEMOGLOBIN

Groups	Means in ascending order	Group DG	Group DB	Group DC	Group DA	Group DD	Group DF	Group DE
		0.03	1.96	1.99	2.07	2.90	3.07	3.19
DG	0.03	-	1.93	1.96	2.04	2.87	3.04	3.16
DB	1.96		-	0.03	0.11	0.94	1.11	1.23
DC	1.99			-	0.08	0.91	1.08	1.20
DA	2.07				-	0.83	1.00	1.12
DD	2.90					-	0.17	0.29
DF	3.07						-	0.12
DE	3.19							-

alpha =0.5;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=0.19$* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970.

Serum fasting glucose levels

The mean serum fasting glucose levels ranged from 123.20 to 124.80mg/dl in the experimental and control groups as against the normal range of 70 to 99 mg/dl as quoted by Raghuram *et al.*, (2007). It is inferred that there was a reduction in fasting glucose levels on supplementation with wheat germ, bran and grass individually and in combination. Group DA supplemented with wheat germ and group DC supplemented with wheat grass had a minimal decrease of 22.07mg/dl. Group DB supplemented with wheat bran had a reduction of serum fasting glucose of 22.80mg/dl while group DF supplemented with wheat bran and grass had a reduction of 23.46mg/dl. Group DD supplemented with wheat germ and bran had a decrease of 23.27mg/dl while group DE supplemented with wheat germ and grass had a maximal decrease of 28.33mg/dl. The final values of the serum fasting glucose was significantly lower than the initial values ($P<0.01$) in all the experimental groups. The difference observed between the initial and final values of the control group DG was not significant.

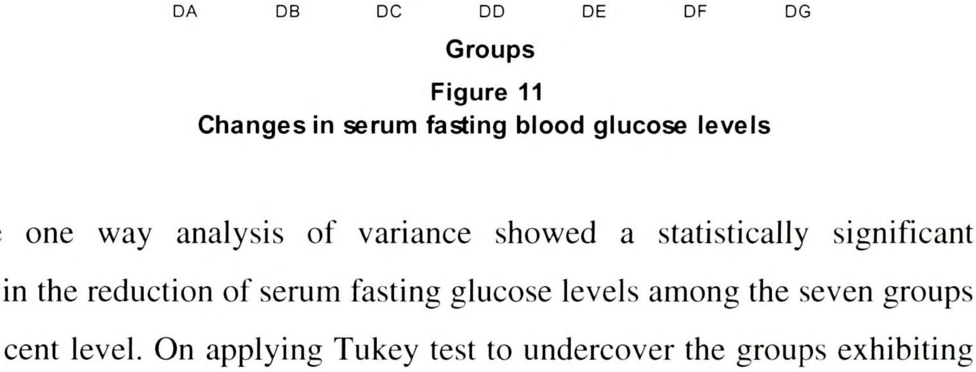

Figure 11
Changes in serum fasting blood glucose levels

The one way analysis of variance showed a statistically significant difference in the reduction of serum fasting glucose levels among the seven groups at one per cent level. On applying Tukey test to undercover the groups exhibiting the significance it was observed that the reduction in the fasting blood glucose in the groups DA,DC,DB,DD,DF and DE supplemented with wheat products proved significance over the group DG which served as the control group. Further the mean difference in the fasting blood glucose levels of group DE supplemented with wheat germ and bran proved significant over groups DA, DC, DB, DD and DF proving the combined potential of wheat germ and bran over the other test groups.

The maximum reduction in the fasting blood glucose levels as observed in group DE supplemented with wheat germ and grass could be substantiated with the study done by Potter *et al.*, (1981)where various test meals of varying dietary fiber content were tested on healthy subjects and these meals did decrease fasting blood glucose levels.

Serum postprandial glucose levels

The mean serum postprandial glucose levels of the experimental and control groups ranged from 184.40 to 188.53mg/dl whereas normal level ranges from 80 to 120 mg/dl as quoted by Raghuram *et al.*,(2007). The serum postprandial glucose levels reduced after individual as well as combined supplementation of wheat germ, bran and grass. The final mean serum postprandial glucose levels in the experimental groups after the supplementation ranged from 113.07 to 123.93 mg/dl. Among the supplemented groups, maximum reduction of 52.87mg/dl was noted in group DE supplemented with wheat germ and grass and the minimum reduction of 39.80mg/dl was noted in group DB supplemented with wheat bran. Group DA supplemented with wheat germ had a decrease of 40.74mg/dl while group DC supplemented with wheat grass had a decrease of 47.93 mg/dl. Group DD supplemented with wheat germ and bran had a decrease of 48.20 mg/dl. Group DF supplemented with wheat bran and grass showed a decrease of 51.47mg/dl which was almost closer to the decrease observed in group DE supplemented with wheat germ and grass. Decrease in serum postprandial glucose levels in the experimental groups were found to be significant at one per cent level whereas the change observed in the control group was not statistically significant.

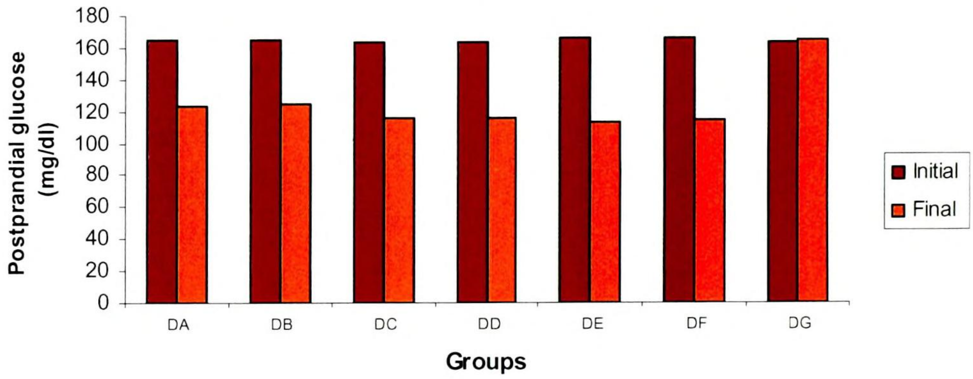

Figure 12
Changes in serum postprandial glucose levels

One way analysis of variance was performed and the difference between the groups was found to be significant at one per cent level. Tukey test revealed that the decreases observed in the mean serum postprandial glucose in groups DB,DA,DC,DD,DF and DE were significantly greater than group DG proving the potential of supplementation. Moreover the mean decrease observed in groups DC, DD and DF supplemented with wheat grass, wheat germ and bran and wheat bran and grass were significantly higher than that of groups DB and DA supplemented with wheat bran and wheat germ respectively. Group DE supplemented with wheat germ and grass pictured increased reduction and was significant over groups DB, DC, DA and DD proving the efficacy of supplementation of wheat germ and grass over the supplementation of wheat bran, wheat grass, wheat germ and wheat germ and bran respectively. From the results it could be confirmed that the combined supplementation of wheat germ and grass is more efficient in reducing serum postprandial glucose levels than the individual supplementation of wheat germ, bran and grass or combined supplementation of wheat germ and bran or wheat bran and grass.

The results of the present study are in accordance with another study conducted by Cara *et al* .,(1992) which showed that insulin dependent diabetics treated with wheat germ oil along w

ith insulin showed a glucose reduction of 20.35mg/dl. Murthy (2005) has demonstrated the beneficial potential of wheat grass powder on moderate diabetics by lowering the postprandial glucose by 19.25mg/dl.

Serum glycosylated hemoglobin levels

The glycosylated hemoglobin test (HbA_1C) is an excellent index of long term diabetes control. Unlike blood sugar which tends to fluctuate from day to day and even hour to hour, the HbA_1C test is a true index of the average blood glucose control during previous 2-3 months. HbA_1C test is done in the laboratory rapidly and precisely using the "gold standards" of the HbA_1C testing. The interpretation of the test results are as follows: normal - below 5.6 per cent, good control -5.6 to 7 per cent, fair control – 7 to 8 per cent, unsatisfactory control -8 to 10 per cent and poor control -above 10 per cent (American Diabetic Association, 2007).The mean initial glycosylated hemoglobin levels of the subjects in both the experimental and the control groups were between 8.39 to 8.45 per cent (unsatisfactory control). The impact evaluation showed that the HbA_1C had lowered by the individual supplementation of wheat germ, bran and grass as well as combinations of supplementation of wheat germ and bran, wheat germ and grass and wheat bran and grass over a period of six months. Group DA supplemented with wheat germ had a reduction of 2.07 per cent with the final values at 6.35 per cent which was considered as 'good control'.

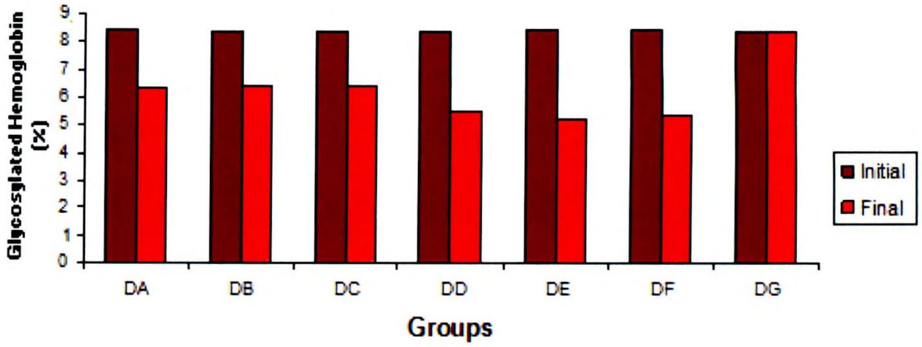

Figure 13
Changes in serum glycosylated hemoglobin levels

A similar effect was noted in groups DB and DC which had a reduction of 1.96 and 1.99 per cent respectively with the final values at 6.41 (good control). In all the three combined supplementation groups namely DD, DE and DF the final values were 5.49, 5.26 and 5. 34 per cent respectively which according to the gold standards set by the American Diabetic Association (2007) were normal. Further maximum reduction of the HbA_1C was noted in group DE supplemented with wheat germ and grass. The reduction between the mean initial and final values of HbA_1C in the all the supplemented groups were significant at one per cent level. No such significance was observed in the control group. Further to this Tukey test was applied to identify which pair of treatment differed significantly. It could be observed that the mean reduction in the HbA_1C of all the supplemented groups were significant over group DG which served as the control group. It could be also noted from the Table that the mean differences of groups DD and DF supplemented with wheat germ and bran and wheat bran and grass respectively were significant over groups DB, DC and DA revealing that the supplementation of wheat germ and bran and wheat bran and grass were better over the individual supplementation of wheat bran, grass and germ respectively. Further group DE supplemented with wheat germ and grass showed significant reduction in HbA_1C over groups DB, DC, DA and DD proving that the combined supplementation of wheat germ and grass supplementation is better than individual supplementation of wheat germ, bran and grass and combined supplementation of wheat germ and bran. It could also be noted that there was no significance between groups DF and DE proving both wheat germ and grass and wheat bran and grass supplementation had a similar impact on the blood glucose levels. Study by Yukiko *et al.*, (2007) proved that HbA_1C reduced on supplementation with Antioxidant Biofactor (AOB) a mixture of commercially available fermented grain foods which includes wheat.

(ii) Mean serum superoxide dismutase , malondialdehyde, glutathione reductase and glutathione peroxidase levels

Table XV and Figures 14 to 17 give the mean serum superoxide dismutase, malondialdehyde, glutathione reduced and glutathione peroxidase at the initial and final phase of the study period along with the mean reduction in their levels.

TABLE XV
CHANGES IN MEAN SERUM SUPERODIXE DISMUTASE , MALONDIALDEHYDE,GLUTATHIONE REDUCED AND GLUTATHIONE PEROXIDASE LEVELS

(N=15/group)

Groups	Superoxide dismutase (U/g Hb)	Malondialdehyde (µM/L)	Glutathione reduced (µM/L)	Glutathione peroxidase (U/g Hb)
Group DA Initial	3321.56 ±2.21	4.39 ± 0.10	41.37 ±0.19	26.61±0.16
Final	3509.89 ± 2.46	4.01 ±0.01	57.25 ±0.27	51.33 ±0.11
Difference	188.33 ±2.10	-0.38 ±0.10	15.88 ±0.33	24.72 ±0.22
't' value	335.41**	14.75**	185.36**	433.97**
Group DB Initial	3351.85 ±2.20	4.30 ±0.10	45.37 ±0.18	26.40 ±0.16
Final	3449.78 ± 5.05	4.02 ± 0.01	54.22 ±0.51	53.64 ±0.40
Difference	97.93 ± 5.06	-0.28 ± 0.10	8.85 ±0.51	27.24 ±0.48
t' value	74.91**	10.90**	66.65**	64.61**
Group DC Initial	3369.42 ±3.98	4.43 ±0.09	45.35 ±0.19	26.24 ±0.44
Final	3605.69 ±2.71	3.86 ± 0.08	55.82 ±0.89	53.64 ±0.40
Difference	236.27 ±1.17	-0.57 ± 0.13	10.47 ±0.90	27.40 ±0.48
t' value	755.26**	16.35**	45.18**	23.33**
Group DD Initial	3458.13 ±1.67	4.44 ± 0.09	45.32 ±0.16	26.47 ±0.49
Final	3709.73 ±1.74	3.95 ± 0.08	54.73 ±0.75	52.67 ±1.03
Difference	251.60 ±1.98	-0.49 ± 0.13	9.41 ±0.81	26.20±1.19
t' value	475.24**	17.52**	44.91**	85.75**
Group DE Initial	3385.55 ±2.51	4.40 ± 0.10	45.37 ± 0.18	26.47 ±0.31
Final	3647.68 ±1.26	3.31 ± 0.09	64.15 ± 0.56	59.40 ±0.52
Difference	262.13 ±1.41	-1.09 ± 0.14	18.78 ±0.55	32.93 ±0.68
t' value	695.52**	30.06**	131.62**	187.92**
Group DF Initial	3285.47 ±2.56	4.42 ± 0.08	45.40 ±0.20	26.47 ±0.31
Final	3453.47 ±9.05	3.59 ±0.27	53.40 ±0.15	53.18 ±0.99
Difference	168 ±8.86	-0.83± 0.31	8.00 ±0.24	26.71 ±1.03
't' value	73.47**	10.51**	131.38**	62.69**
Group DG Initial	3249.26 ±3.33	4.43 ±0.09	45.32 ±0.16	26.53 ±0.12
Final	3248.08±1.84	4.40 ±0.10	45.37 ±0.18	26.55 ±0.14
Difference	-1.18 ±4.03	-0.03 ± 0.14	0.05 ±0.23	0.02 ±0.21
't' value	1.18 ^{NS}	0.80 ^{NS}	0.91 ^{NS}	0.34 ^{NS}
'F' value between groups	588.03**	68.46**	1683.66**	3460.13**

** Significant at one per cent level; ^{NS} not significant

TUKEY VALUES FOR SERUM SUPEROXDIXE DISMUTASE

Groups	Means in ascending order	Group DG	Group DB	Group DF	Group DA	Group DC	Group DD	Group DE
		1.18	97.93	168.00	188.33	236.27	251.60	262.13
DG	1.18	-	96.75	166.82	187.15	235.09	250.42	260.95
DB	97.93		-	70.07	90.40	138.34	153.67	164.20
DF	168.00			-	20.33	68.27	83.60	94.13
DA	188.33				-	47.94	63.27	73.80
DC	236.27					-	15.33	25.86
DD	251.60						-	10.53
DE	262.13							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=16.79$

TUKEY VALUES FOR SERUM MALONDIALDEHYDE

Groups	Means in ascending order	Group DG	Group DB	Group DA	Group DD	Group DC	Group DF	Group DE
		0.03	0.28	0.38	0.49	0.57	0.83	1.09
DG	0.03	-	0.25	0.35	0.46	0.54	0.80	1.06
DB	0.28		-	0.10	0.21	0.29	0.55	0.81
DA	0.38			-	0.11	0.19	0.45	0.71
DD	0.49				-	0.08	0.34	0.60
DC	0.57					-	0.26	0.52
DF	0.83						-	0.26
DE	1.09							-

alpha =0.5 $D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=0.19$

TUKEY VALUES FOR SERUM GLUTATHIONE REDUCED

Groups	Means in ascending order	Group DG	Group DF	Group DB	Group DD	Group DC	Group DA	Group DE
		0.05	8.00	8.85	9.41	10.47	15.88	18.78
DG	0.05	-	7.95	8.80	9.36	10.42	15.83	18.73
DF	8.00		-	0.85	1.41	2.47	7.88	10.78
DB	8.85			-	0.56	1.62	7.03	9.93
DD	9.41				-	1.06	6.47	9.37
DC	10.47					-	5.41	8.31
DA	15.88						-	2.90
DE	18.78							-

alpha =0.5 $D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=0.19$* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970.

TUKEY VALUES FOR GLUTATHIONE PEROXIDASE

Groups	Means in ascending order	Group DG	Group DA	Group DD	Group DF	Group DB	Group DC	Group DE
		0.02	24.72	26.20	26.71	27.24	27.40	32.93
DG	0.02	-	24.70	26.18	26.69	27.22	27.38	32.91
DA	24.72		-	1.48	1.99	2.52	2.68	8.21
DD	26.20			-	0.57	1.04	1.20	6.73
DF	26.71				-	0.53	0.69	6.22
DB	27.24					-	0.16	5.69
DC	27.40						-	5.53
DE	32.93							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=0.79$* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970.

Serum superoxide dismutase levels

The mean initial serum superoxide dismutase levels of the experimental and the control groups ranged from 3321.56 to 3458.13 U/gHb whereas the normal range is 3800 to 5500U/g Hb (Winterbourn *et al.,* 1975).There was an increase in the serum superoxide dismutase levels on supplementation. Maximum increase of 262.13 U/g Hb was observed in group DE supplemented with wheat germ and grass while the slightest increase of 97.83 U/g Hb was noted in group DB supplemented with wheat bran. Group DA supplemented with wheat germ had registered an increase of 188.33 U/g Hb while group DC supplemented with wheat grass recorded an increase of 236.27 U/g Hb. The final values of the serum superoxide dismutase when compared with the initial values were found to be significantly (P<0.01) greater in the experimental groups. A negligible change from the initial and final values of serum superoxide dismutase observed in the control group was not statistically significant. One way analysis of variance conducted to substantiate the significance between the groups also proved to be significant at one per cent level. Results of Tukey test indicated that the mean differences in the levels of serum superoxide dismutase of the supplemented groups DB,DF,DA,DC,DD and DE were significantly greater than the group DG which served as the control. Further it could be noted that the mean incremental differences observed in group DA supplemented with wheat germ was significantly greater over groups DB and DF which were supplemented with wheat bran and wheat bran and grass respectively. Moreover the mean incremental difference in groups DC and DD supplemented with wheat grass and wheat germ and grass respectively was also significant over groups DB, DF and DA supplemented with wheat bran, wheat bran and grass and wheat germ respectively. The mean incremental difference of the serum superoxide dismutase of Group DE supplemented with wheat germ and grass was significant over groups DB, DF, DA and DC which were supplemented with wheat bran, wheat bran and grass and wheat germ and wheat grass respectively proving the potential of combined supplementation of wheat germ and grass.

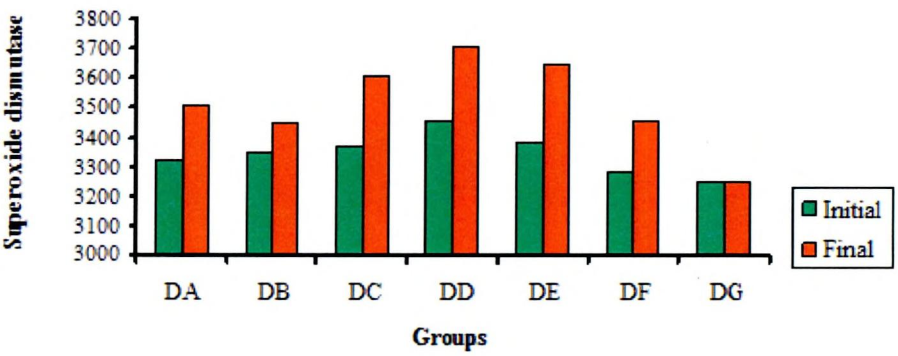

Figure 14
Changes in serum superoxide dismutase levels

Tukey test revealed that supplementation with wheat germ and grass resulted in significantly greater increments in serum superoxide dismutase over the supplementation of wheat bran and grass and what bran only. Superoxide dismutase is one of the most important of antioxidant enzymes. It catalyses the dismutation of superoxide radicals to hydrogen peroxide and molecular oxygen. It acts 10000 times faster than the spontaneous rate of superoxide radical dismutation (Droge *et al.,*2005). In this study, serum superoxide dismutase level significantly increased in the supplemented groups ($P<0.01$) as compared to the control group. The lower level of initial serum superoxide dismutase could be due to its direct inactivation by its products H_2O_2 or O.- 2 anion itself and / or development of auto antibodies against serum superoxide dismutase (Kurian and Scofield,2003). A study by Boros *et al.*,(2005) proved that wheat germ extract decreases glucose uptake and RNA ribose formation and increased superoxide dismutase level in pancreatic adenocarcinoma cells.

Serum malondialdehyde levels

Yet another comprehensible utility of the supplementation undertaken was the demonstrably distinct decrease of mean serum malondialdehyde levels. The initial mean serum malondialdehyde levels in both the experimental and control groups ranged from 4.30 to 4.44 μM/L which was much higher than the normal range of 0.15 to 0.69 μM/L (Varley *et al.,* 1986). The impact evaluation showed that the serum malondialdehyde had lowered by the individual supplementation of wheat germ, bran and grass as well as combinations of wheat germ and bran, wheat germ and grass and wheat bran and grass over a period of six months. Group DA supplemented with wheat germ had a reduction of 0.38 μM/L while group DB supplemented with wheat bran had a reduction of 0.28 μM/L and group DC supplemented with wheat grass had a decrease of 0.57 μM/L. Groups including group HE supplemented with wheat germ and grass had a maximum reduction of 1.09 μM/L followed by group DF supplemented with wheat bran and grass with a decrease of 0.83 μM/L.

Group DD supplemented with wheat germ and bran had a decrease of 0.49 μM/L in the mean serum malondialdehyde after six months of supplementation. The reduction observed in the final values of mean serum malondialdehyde in all the supplemented groups were significant at one per cent level. It could be observed from the Tukey test that in all the supplemented groups the mean serum malondialdehyde reduced significantly than the control group. Further it could be noted from the Tukey table that group DD supplemented with wheat germ and bran showed a significant betterment in the reduction of serum malondialdehyde over group DB supplemented with wheat bran. Group DC supplemented with wheat grass showed significant reduction of serum malondialdehyde when compared with groups DB and DA which were supplemented with wheat bran and wheat germ respectively.

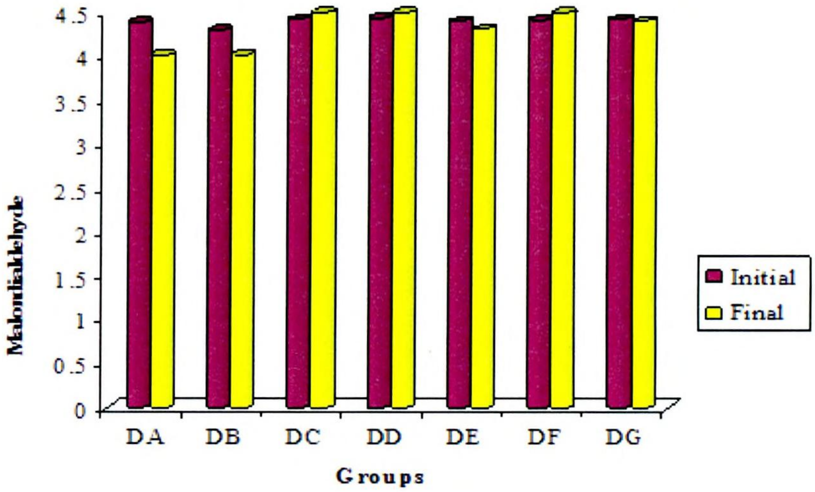

Figure 15
Changes in serum malondialdehyde levels

Group DF supplemented with wheat bran and grass also proved significant reduction in serum malondialdehyde when compared against groups DB,DA,DD and DC proving that supplementation of wheat bran and grass was even better than supplementation of wheat bran, germ, wheat germ and bran and wheat grass respectively. Group DE supplemented with wheat germ and grass showed significant reduction of serum malondialdehyde over the groups DB,DA,DD,DC and DF which were supplemented with wheat bran, germ, wheat germ and bran, wheat grass and bran and grass respectively proving supplementation of wheat germ and grass is more beneficial in reducing the lipid peroxidation.

Through the statistical analysis it is clearly defined that group DE supplemented with wheat germ and grass had a maximum reduction. The observation is in par with a study conducted Gonalez and Gonalez,(2005) who revealed that alpha lipolic acid plus vitamin E from wheat germ helps to reduce oxidative damage by reducing levels of lipid peroxidation and malondialdehyde concentration in ischemic and other oxidation-related pathological conditions.

Serum glutathione reduced levels

The mean initial glutathione reduced levels of the experimental and the control groups ranged from 41.37 to 45.40μM/L whereas normal serum glutathione reduced is in the range of 50-80 μmol/l (Beutler *et al.*,1963).There was an increase in the glutathione reduced levels on supplementation. Maximum increase of 18.78 μM/L was observed in group DE supplemented with wheat germ and grass while the least increase of 8.00 μM/L was noted in group DF supplemented with wheat bran and grass. Group DA supplemented with wheat germ had registered an increase of 15.88 μM/L while group DB supplemented with what bran recorded an increase of 8.85 μM/L. Group DC supplemented with wheat grass had recorded an increase of 10.47 μM/L. The final values of the glutathione reduced when compared with the initial values were found to be significantly (P<0.01) greater in the experimental groups. A negligible change observed in the control group was not statistically significant. One way analysis of variance conducted to substantiate the significance between the groups also proved to be significant at one per cent level.

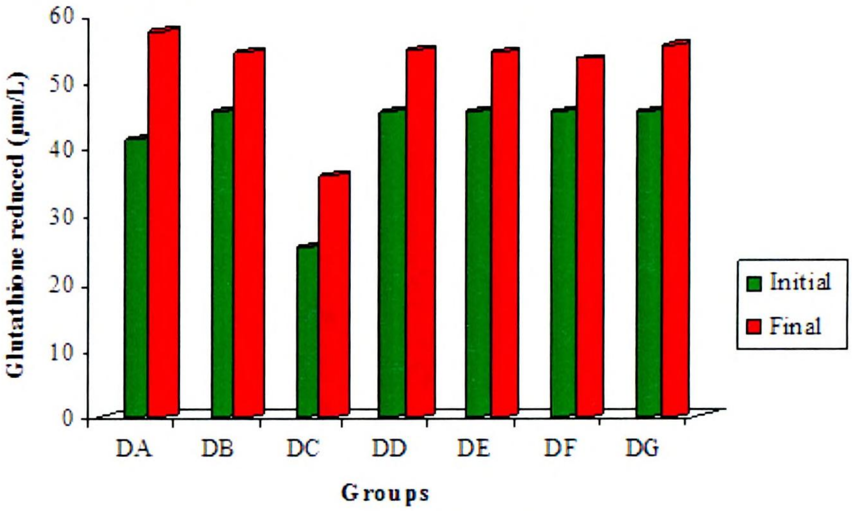

Figure 16
Changes in serum glutathione reduced levels

Results of Tukey test too indicated that the mean differences of glutathione reduced recorded in the supplemented groups were significantly greater than that of the control group. Further it could be noted that the mean incremental differences observed in groups DB and DD supplemented with wheat bran and wheat germ and bran respectively were significant over group DF which was supplemented with wheat bran and grass and moreover the mean incremental difference in group DC which was supplemented with wheat grass also was significant over groups DF,DB and DD. The mean incremental difference of the serum glutathione reduced of Group DA supplemented with wheat germ was significant over groups DF, DB,DD and DC. The mean increment in glutathione reduced of group DE supplemented with wheat germ and grass proved significance over the other supplemented groups namely DF, DB, DD, DC and DA proving the potential of combined supplementation of wheat germ and grass.

It was interesting to note that supplementation with wheat germ and grass resulted in significantly greater increments in glutathione reduced over the supplementation of wheat bran and grass and wheat bran only. According to Djujic *et al.*,(2000) wheat germ supplementation increased the levels of reduced glutathione, the most important antioxidant metabolite that plays an important role in maintaining good levels of glutathione peroxidase activity which is the main enzyme involved in removing the H_2O_2 generated from dismutation of superoxide anions by superoxide dismutase. Glutathione peroxidase is also the co-factor of several reducing enzymes such as dehydroascorbate reductase and endoperoxide isomerase. The results of the study conducted by Tiedge *et al.*,(1997) proved that wheat grass also reduces the lipid peroxidation rate in cancer patients by acting as a good chain breaking antioxidant, which reacts with peroxy radicals formed in propagation phase of lipid peroxidation to form carbon centered radical, which reacts readily and reversibly with oxygen to form a new chain – carrying peroxyl radicals which are highly stable forms than reactive oxygen species (ROS).

Serum glutathione peroxidase levels

The mean initial serum glutathione peroxidase of the diabetic subjects in all the groups were in the range of 26.47 to 26.61U/g Hb as against the normal value of 49 to 93 U/g Hb quoted by Paglia and Valentine (1967).Six months of supplementation of wheat products including wheat germ, bran ,grass and their combinations brought about a marked increase in the serum glutathione peroxidase values in all the supplemented groups. Group DA supplemented with wheat germ had an increase of 24.72 U/g Hb in the serum glutathione peroxidase levels while group DB supplemented with wheat bran resulted in a minimal increase of 27.24 U/g Hb. Group DC supplemented with wheat germ and grass had an increase of 27.40 U/g Hb among the supplemented groups while group DD supplemented with wheat germ and bran had an increase of 26.20 U/g Hb. Group DE supplemented with wheat germ and grass had a maximum increase of 32.93 U/g Hb followed by group DF supplemented with wheat bran and grass which had an increase of only 26.71 U/g Hb in the serum glutathione peroxidase levels. Group DG which served as the control group had an increase of 0.65 U/g Hb over a period of six months without any supplementation. The final values of the serum glutathione peroxidase was significantly lower than the initial values (P<0.01) in all the groups except the control group HG.

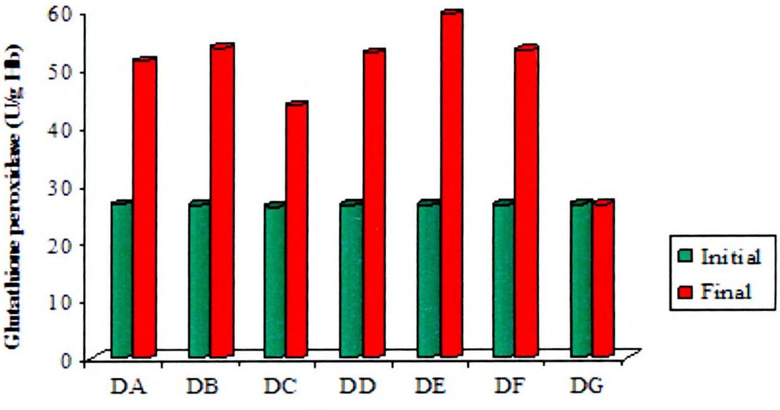

Figure 17
Changes in serum glutathione peroxidase

The one way analysis of variance showed significance in the reduction of serum glutathione peroxidase levels among the seven groups at one per cent level. On applying Tukey test to undercover the groups exhibiting the significance it was observed that the mean incremental difference of serum glutathione peroxidase in the groups DF,DB,DD,DC,DA and DE were significantly greater than that of control group HG. Groups DB and DD supplemented with wheat bran and wheat germ and bran had resulted in a significantly greater increments in the mean serum glutathione peroxidase levels than the group DF which was supplemented with wheat germ and grass. The increase in mean serum glutathione peroxidase of group DC supplemented with wheat grass showed significance over the groups DF,DB and DD. The increments in the mean difference of group DA supplemented with wheat germ showed significance over the remaining six groups .The results are in par with the study of Eric and Bright (1951) who has isolated the enzyme glutathione reducatase which oxidizes glutathione by triphoshopyridine nucleotide in aqueous extract of wheat germ.

(iii) Mean serum vitamin C and vitamin E levels

Table XVI and Figures 18 and 19 give the mean serum vitamin C and vitamin E levels at the initial and final phase of the study period along with the mean increments in their levels.

TABLE XVI
CHANGES IN MEAN SERUM VITAMIN C AND VITAMIN E LEVELS
(N=15/group)

Groups	Vitamin C (mg/dl)	Vitamin E (mg/dl)
Group DA Initial	1.02±0.02	0.80±2.08
Final	1.18±0.02	1.69 ±1.20
Difference	0.16±0.03	0.89 ±0.85
't' value	19.95**	3.92**
Group DB Initial	1.07 ±0.02	0.81 ±2.08
Final	1.18 ±0.02	1.59 ±3.08
Difference	0.11 ±0.04	0.78±0.81
t' value	10.29**	3.60**
Group DC Initial	1.07 ±0.15	0.82 ±2.02
Final	1.45 ±0.03	1.62 ±2.01
Difference	0.38 ±0.17	0.80 ±0.93
t' value	8.36**	3.2**
Group DD Initial	1.07 ±0.02	0.79 ±2.08
Final	1.17 ±0.02	1.63 ±2.01
Difference	0.10 ±0.01	0.84 ±0.81
t' value	37.40**	3.88**
Group DE Initial	1.02±0.02	0.85 ±2.03
Final	1.47 ±0.18	1.84 ±3.01
Difference	0.45 ±0.03	0.99 ±1.06
t' value	56.10**	3.49**
Group DF Initial	1.06 ±0.13	0.82 ±2.03
Final	1.42 ±0.03	1.46 ±2.01
Difference	0.36 ±0.13	0.64 ±1.08
't' value	10.36**	2.21*
Group DG Initial	1.09 ±0.11	0.81±1.02
Final	1.07 ±0.09	0.83 ±2.01
Difference	-0.02 ±0.10	0.02 ±3.01
't' value	0.75 ^{NS}	0.02 ^{NS}
'F' value between groups	49.57**	3.37*

** significant at one per cent level;* significant at five per cent level; ^{NS} not significant

TUKEY VALUES FOR SERUM VITAMIN C

Groups	Means in ascending order	Group DG	Group DD	Group DB	Group DA	Group DF	Group DC	Group DE
		0.02	**0.10**	**0.11**	**0.16**	**0.36**	**0.38**	**0.45**
DG	**0.02**	-	0.08	0.09	0.14	0.34	0.36	0.43
DD	**0.10**		-	0.01	0.06	0.26	0.28	0.35
DB	**0.11**			-	0.05	0.25	0.27	0.34
DA	**0.16**				-	0.20	0.22	0.29
DF	**0.36**					-	0.02	0.09
DC	**0.38**						-	0.07
DE	**0.45**							-

alpha =0.5 $D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=0.09$

TUKEY VALUES FOR SERUM VITAMIN E

Groups	Means in ascending order	Group DG	Group DF	Group DB	Group DC	Group DD	Group DA	Group DE
		0.02	**0.64**	**0.78**	**0.80**	**0.84**	**0.89**	**0.99**
DG	**0.02**	-	0.62	0.76	0.78	0.82	0.87	0.97
DF	**0.64**		-	0.14	0.16	0.20	0.25	0.35
DB	**0.78**			-	0.02	0.06	0.11	0.21
DC	**0.80**				-	0.04	0.09	0.19
DD	**0.84**					-	0.05	0.15
DA	**0.89**						-	0.10
DE	**0.99**							-

alpha =0.5 $D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=0.61$* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970.

Serum vitamin C levels

The mean initial serum vitamin C levels ranged from 1.02 to 1.09 mg/dl in the experimental and control groups as against the normal range of 0.40 to 1.50 mg /dl as quoted by Kurl *et al.*, (2002). It is inferred that there was an increase in serum vitamin C levels on supplementation with wheat germ, bran and grass individually and in combinations. Groups DA, DB and DC supplemented with wheat germ, bran and grass individually recorded an increase of 0.16, 0.11 and 0.38 mg /dl respectively. Group DD supplemented with wheat germ and bran had an increase of 0.10 mg /dl while group DF supplemented with wheat bran and grass had an increment of 0.36 mg /dl only. Group DE supplemented with wheat germ and grass had the maximum increase of 0.45 mg /dl. The final values of the serum vitamin C were significantly (P<0.01) greater than the initial values in all the experimental groups. No significant difference was observed between the initial and final values of the control group DG.

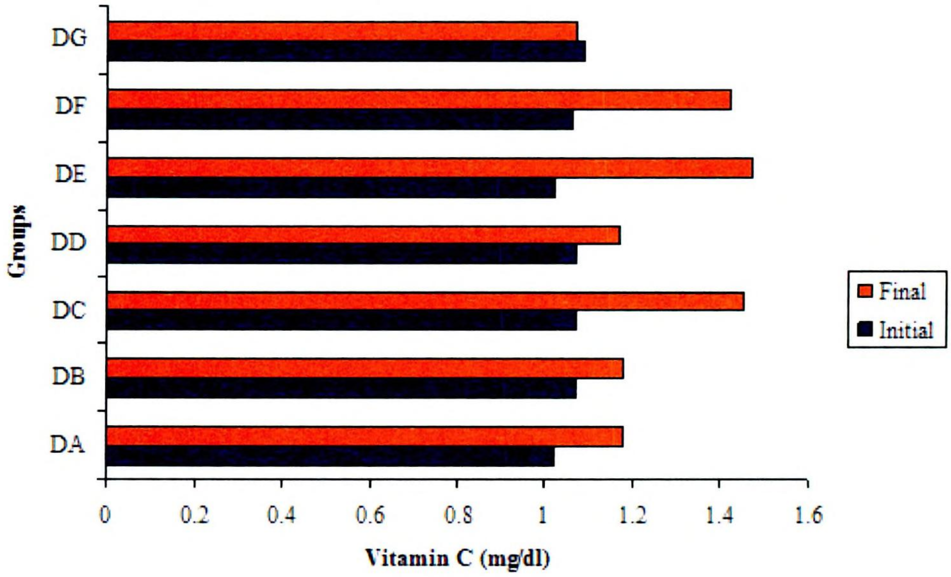

Figure 18
Changes in serum vitamin C levels

The one way analysis of variance showed a statistically significant difference in the increase of serum vitamin C levels among the seven groups at one per cent level. Tukey test exhibited significance in the serum vitamin C levels in the supplemented groups DB,DA,DF,DC and DE over the control group. Further the mean difference of the serum vitamin C levels of groups DF and DC supplemented with wheat germ and bran and wheat grass respectively proved to be significant over groups DD,DB and DA supplemented with wheat germ and bran, wheat bran and wheat germ respectively. The mean increments of group DE supplemented with wheat germ and grass was significantly greater over groups DD,DB , DA and DC supplemented with wheat germ and bran, wheat bran ,wheat germ and wheat grass respectively. From the results it could be inferred that supplementation of wheat germ and grass is more beneficial than other combinations.

Serum vitamin E levels

The mean initial serum vitamin E levels ranged from 0.79 to 0.85 mg/dl in the experimental and control groups as against the normal range of 1.35 to 2.50 mg/dl as quoted by Baker *et al.*,(1988). It is inferred that on supplementation with wheat germ, bran and grass individually and in combinations proved an increase in serum selenium levels. Group DA supplemented with wheat germ had an increase of 0.89 mg/dl, group DB supplemented with wheat bran had a minimal increase of 0.78 mg/dl and group DC supplemented with wheat grass had an increase of 0.80 mg/dl. Group DD supplemented with wheat germ and bran had an increase of serum vitamin E levels of 0.84 mg/dl while group DF supplemented with wheat germ and grass had an increment of 0.64 mg/dl. Group DE supplemented with wheat germ and bran had a maximal increase of 0.99 mg/dl. The final values of the serum selenium was significantly greater than the initial values ($P<0.05$) in all the experimental groups. The difference observed between the initial and final levels of the control group DG was not found to be statistically significant.

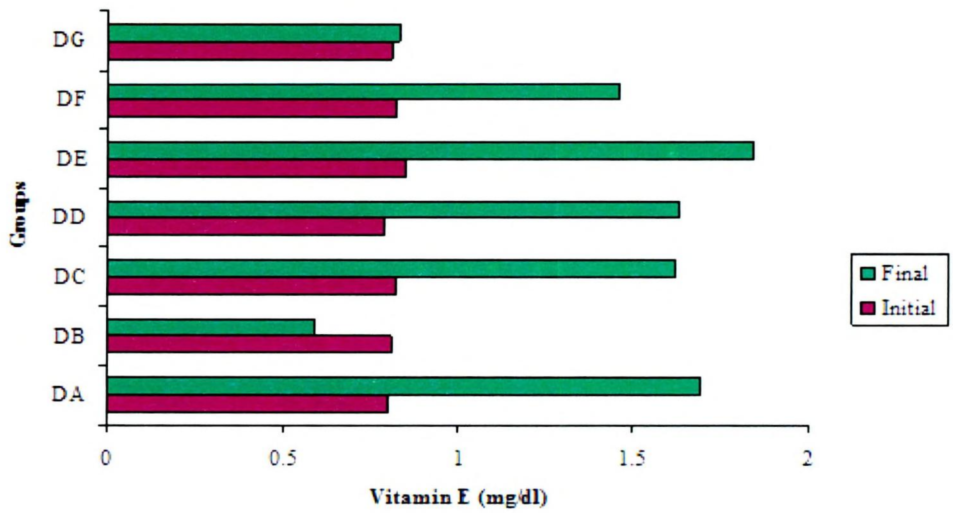

Figure 19
Changes in serum vitamin E levels

The one way analysis of variance showed a statistically significant difference in the increase of serum vitamin E levels among the seven groups at five per cent level. On applying Tukey test to undercover the groups exhibiting the significance it was observed that the increment in the serum vitamin E levels in all the supplemented groups were significantly greater than that of the control group. According to a study by Stampfer and Kelfe (1993) dietary supplementation of wheat germ is less effective than the synthetic supplements in coronary heart disease men. Similarly wheat product supplementation did not have much impact on the vitamin E levels of the diabetics.

(iv) Changes in mean serum zinc, selenium and copper levels

The changes in mean serum zinc, selenium and copper levels of the diabetic subjects before and after supplementation is given in Table XVII and Figures 20,21 and 22.

TABLE XVII
CHANGES IN MEAN SERUM ZINC, SELENIUM AND COPPER LEVELS
(N=15/group)

Groups	Zinc (µg/l)	Selenium (µg/l)	Copper (µg/l)
Group DA			
Initial	554.80±1.70	121.13±.46	944.33±2.94
Final	609.53±2.27	135.09 ±1.46	941.47 ±2.29
Difference	54.73±2.45	13.96 ±1.49	-2.86 ±1.97
't' value	83.54**	35.04**	5.43**
Group DB			
Initial	541.33±1.85	120.24 ±9.32	943.47 ±1.35
Final	571.87 ±2.29	133.44 ±9.32	941.27 ±1.30
Difference	30.54±1.38	13.20 ±10.65	-2.20 ±1.31
t' value	82.77**	4.64**	6.28**
Group DC			
Initial	547 ±12.34	122.20 ±6.16	946.67 ±1.72
Final	541.20 ± 1.17	127.34 ±8.32	943.40 ±1.13
Difference	5.80 ±2.12	5.14 ±6.48	-3.27 ±1.59
t' value	10.23**	2.96**	7.69**
Group DD			
Initial	543.87 ±1.10	120.11 ±1.66	948.60 ±1.40
Final	607.00 ±1.03	137.91 ±1.62	944.67 ±1.87
Difference	63.13 ±2.33	17.80 ±1.54	-3.93 ±1.17
t' value	91.30**	43.23**	12.56**
Group DE			
Initial	559.26 ±1.43	121.25 ±1.64	953.47 ±2.00
Final	615.49 ±1.77	139.45 ±1.83	947.73 ±1.42
Difference	56.23 ±1.14	18.20 ±1.46	-5.74 ±1.93
t' value	184.47**	46.62**	11.12**
Group DF			
Initial	541.55 ±1.00	122.02 ±2.59	944.87 ±1.38
Final	572.79 ±1.72	134.62 ±2.95	941.20 ±1.43
Difference	31.24 ±1.91	12.60 ±2.73	-3.67±1.00
't' value	61.17**	17.26**	13.73**
Group DG			
Initial	560.27 ±1.70	121.12±1.25	951.33 ±1.37
Final	-560.93 ±1.78	121.19 ±1.26	951.87 ±1.18
Difference	0.66±1.54	0.07 ±1.79	-0.54 ±0.59
't' value	1.60 NS	0.15 NS	1.27 NS
'F' value between groups	15.36**	8.48**	16.09**

** significant at one per cent level; NS not significant

TUKEY VALUES FOR SERUM ZINC

Groups	Means in ascending order	Group DG	Group DC	Group DB	Group DF	Group DA	Group DE	Group DD
		0.66	**5.80**	**30.54**	**31.24**	**54.73**	**56.23**	**63.13**
DG	**0.66**	-	5.14	28.94	29.88	54.07	55.57	62.47
DC	**5.80**		-	24.74	25.44	48.93	50.43	57.33
DB	**30.54**			-	0.70	24.19	25.69	32.59
DF	**31.24**				-	23.49	24.99	31.89
DA	**54.73**					-	1.50	8.4
DE	**56.23**						-	6.9
DD	**63.13**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=24.58$

TUKEY VALUES FOR SERUM SELENIUM

Groups	Means in ascending order	Group DG	Group DC	Group DB	Group DF	Group DA	Group DD	Group DE
		0.07	**5.14**	**12.60**	**13.20**	**13.96**	**17.80**	**18.20**
DG	**0.07**	-	5.07	12.53	13.13	13.89	17.73	18.13
DC	**5.14**		-	7.46	8.06	8.82	12.66	13.06
DF	**12.60**			-	0.60	1.36	5.20	5.60
DB	**13.20**				-	0.76	4.60	5.00
DA	**13.96**					-	3.84	4.24
DD	**17.80**						-	0.40
DE	**18.20**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=15.17$

TUKEY VALUES FOR SERUM COPPER

Groups	Means in ascending order	Group DG	Group DC	Group DB	Group DF	Group DA	Group DD	Group DE
		0.54	**2.20**	**2.86**	**3.27**	**3.67**	**3.93**	**5.74**
DG	**0.54**	-	1.66	2.32	2.73	3.13	3.39	5.20
DB	**2.20**		-	0.66	1.07	1.47	1.73	3.54
DA	**2.86**			-	0.41	0.81	1.07	2.88
DC	**3.27**				-	0.40	0.66	2.47
DF	**3.67**					-	0.26	2.07
DD	**3.93**						-	1.81
DE	**5.74**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=1.78$* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970.

Serum zinc levels

The mean initial serum zinc levels ranged from 541.33 to 560.27μg/l in the experimental and control groups as against the normal range of 565-580 μg /l as quoted by Klevay (2002). It is inferred that there was a gradual increase in serum zinc levels on supplementation with wheat germ, bran and grass individually and in combinations. Groups DA, DB and DC supplemented with wheat germ/bran/grass individually recorded an increase of 54.73, 30.54 and 5.80 μg /l respectively. Group DD supplemented with wheat germ and bran had a maximal increase of 63.13 μg /l of serum zinc while group DF supplemented with wheat bran and grass had an increment of 31.24 μg /l. Group DE supplemented with wheat germ and grass had an increase of 56.23 μg /l. The final values of the serum zinc was significantly lower than the initial values (P<0.01) in all the experimental groups. No significant difference was observed between the initial and final values of the control group DG.

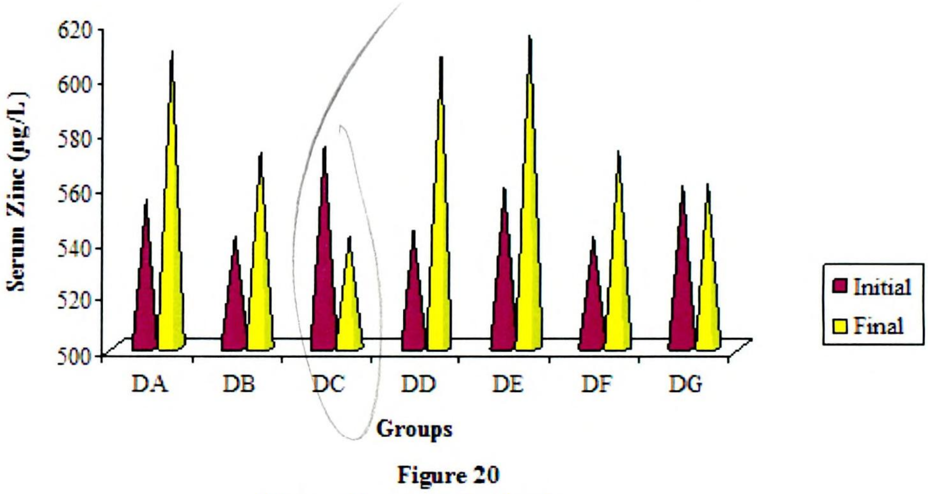

Figure 20
Changes in serum zinc levels

The one way analysis of variance showed a statistically significant difference in the increase of serum zinc levels among the seven groups at one per cent level. Through Tukey test the serum zinc levels in all the supplemented groups proved significance over the control group. Further the mean difference of the serum zinc levels of groups DB, DF and DA supplemented with wheat bran, wheat germ and bran and wheat germ respectively proved to be significant over group DC supplemented with wheat grass.

The mean increments of groups DE and DD supplemented with wheat germ and grass and wheat germ and bran were significantly greater over groups DC,DB and DF which were supplemented with wheat grass, wheat bran and wheat germ and bran respectively. From the results it could be inferred that supplementation of wheat germ and bran and wheat germ and grass is more beneficial than other combinations.

Kang and Zhon (2005) has revealed that zinc from wheat germ helps to reduce oxidative damage by reducing levels of lipid peroxidation and malondialdehyde concentration in ischemic and other oxidation-related pathological conditions.

Serum selenium levels

The mean serum selenium levels ranged from 121.12 to121.25 µg/l in the experimental and control groups as against the normal range of 140-150 µg /l as quoted by Freidun *et al.*,(2007). It is inferred that on supplementation with wheat germ, bran and grass individually and in combinations resulted in an increase in serum selenium levels. Group DA supplemented with wheat germ had an increase of 13.96 µg /l, group DB supplemented with wheat bran had an increase of 13.20 µg /dl and group DC supplemented with wheat grass had a minimal increase of 5.14 µg /dl. Group DD supplemented with wheat germ and bran had an increase of serum selenium of 17.80 µg /dl while group DF supplemented with wheat germ and grass had an increment of 12.60 µg /dl. Group DE supplemented with wheat germ and bran had a maximal increase of 18.20 µg /dl. The final values of the serum selenium was significantly greater than the initial values ($P<0.01$) in all the experimental groups except in the control group DG.

The one way analysis of variance showed a statistically significant difference in the increase of serum selenium levels among the seven groups at one per cent level. On applying Tukey test to undercover the groups exhibiting the significance it was observed that the increment in the serum zinc levels in all the supplemented groups were significantly greater than that of the control group. Further the mean differences in the mean serum selenium of groups DA ,DD and DE supplemented with wheat germ and wheat germ and bran respectively were proved to be significant over groups DC,DB and DF supplemented with wheat grass, wheat bran and wheat germ and grass respectively. From the results it could be inferred that supplementation of wheat germ individually or in combination with bran or grass is

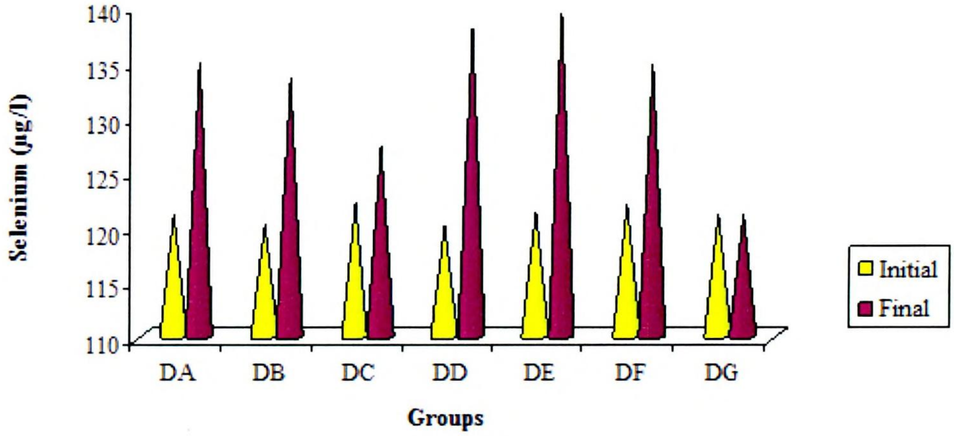

Figure 21
Changes in serum selenium levels

beneficial. Wheat germ supplementation in patients undergoing radiation therapy for rectal cancer had improved selenium levels which resulted in improved quality of life and reduced the appearance of secondary cancers (Carmia *et al*., 2004).

Serum copper levels

Initially mean serum copper levels ranged from 943.47 to 953.47 µg/l in the experimental and control groups as against the normal range of 950-1100µg /l as quoted by Klevay (1987). There was a decrease in serum copper levels with supplementation with wheat germ, bran and grass individually and in combinations. Group DA supplemented with wheat germ had a decrease of 2.86µg /l, group DB supplemented with wheat bran had a reduction of 2.20µg /l and group DC supplemented with wheat grass had a minimal decrease of 3.27 µg /l. Group DD supplemented with wheat germ and bran had a decrease of serum copper by 3.93µg /l while group DF supplemented with wheat germ and grass had a reduction of 3.67 µg /l. Group DE supplemented with wheat germ and bran had a maximal reduction of 5.74 μg /l. The final values of the serum copper were significantly lesser than the initial values (P<0.01) in all the experimental groups. No significant difference was observed between the initial and final values of the control group DG.

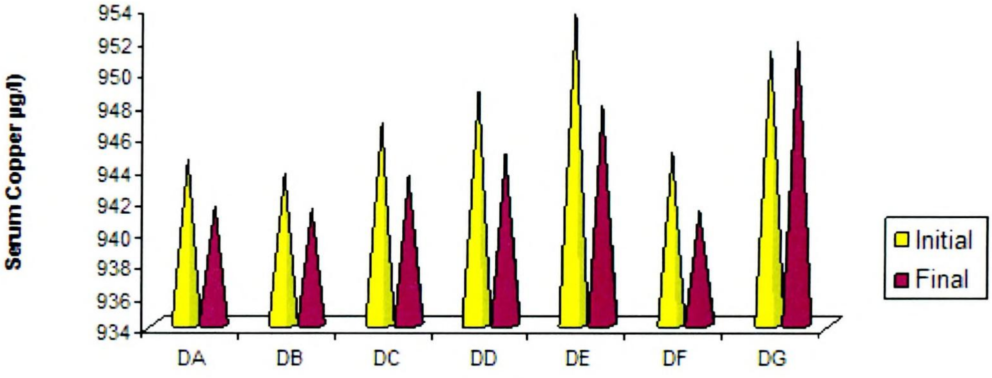

Figure 22
Changes in serum copper levels

The one way analysis of variance showed a statistically significant difference in the decrease of serum zinc levels among the seven groups at one per cent level. On applying Tukey test to undercover the groups exhibiting the significance it was observed that the reduction in the serum copper levels in the groups DB,DF,DA,DD and DE supplemented with wheat products proved significance over the group DG which served as the control group. Further the mean difference of the serum copper of group DE supplemented with wheat germ and bran proved significance over groups DB,DA,DC,DF and DD supplemented with wheat bran, wheat germ, wheat grass, wheat bran and grass and wheat germ and bran respectively.

Zinc and copper are minerals required by the human body. They are necessary components of many enzymes. Levels of zinc and copper in the body are regulated by a protein called metallothionein. As a result of this regulation, copper levels decrease as zinc levels increase and vice versa (Lonnerdal,1998).

(iv) Comparison of the effect of different supplements on the blood parameters

Table XXIV summarizes the results of the supplementation study on the blood parameters among the diabetic subjects.

TABLE XVIII
COMPARISON OF THE EFFECT OF DIFFERENT SUPPLEMENTS ON BLOOD PARAMETERS

Parameter	Rank					
	1	2	3	4	5	6
Fasting blood glucose (mg/dl)	DE	DF	DD	DB	DC,DA	-
Postprandial blood glucose (mg/dl)	DE	DF	DD	DC	DA	DB
Glycosylated hemoglobin (%)	DE	DF	DD	DA	DC	DB
Superoxide dismutase (U/gHb)	DE	DF	DC	DD	DA	DB
Malondialdehyde (μM/L)	DE	DA	DC	DD	DB	DF
Glutathione reduced (μM/L)	DE	DA	DC	DD	DB	DF
Glutathione peroxidase (U/g Hb)	DE	DA	DD	DF	DB	DC
Vitamin C (mg/dl)	DD	DE	DA	DF	DB	DC
Vitamin E(mg/dl)	DE	DD	DA	DF	DB	DC
Serum Zinc (μg/L)	DD	DE	DA	DF	DB	DC
Serum selenium(μg/L)	DE	DD	DA	DF	DB	DC
Serum copper(μg/L)	DE	DD	DF	DC	DA	DB

It could be clearly depicted from the Table that group DE supplemented with wheat germ and grass proved its efficiency in maintaining the blood parameters which were altered in diabetes except in the serum vitamin C and vitamin E levels which were best improved in group DD which was supplemented with wheat germ and bran. Group DF supplemented with wheat bran and grass was followed by group DE in maintaining the fasting blood glucose, postprandial blood glucose, Glycosylated hemoglobin and superoxide dismutase levels. Supplementation of wheat bran and grass individually proved less efficiency in maintaining the blood parameters than the combined supplementation of wheat germ, bran and grass.

(v) Changes in fasting blood glucose, post prandial glucose and glycosylated hemoglobin levels after the withdrawal of supplementation

Table XIX gives the final and three months after the withdrawal of supplementation data of fasting blood glucose, postprandial glucose and glycosylated hemoglobin levels.

TABLE XIX
CHANGES IN BLOOD PARAMETERS AFTER THE WITHDRAWAL OF SUPPLEMENTATION

(N=5/group)

Groups	Fasting glucose (mg/dl)	Post prandial glucose (mg/dl)	Glycosylated hemoglobin (%)
Group DA			
Final	101.13±0.92	123.93±2.63	6.35±0.22
A3M	113.20±1.32	143.60±2.36	7.41±0.17
Difference	12.07±1.57	19.67±2.81	1.06±0.29
‘t’ value	17.22**	15.68**	8.19**
Group DB			
Final	100.80 ±1.15	124.80 ±2.11	6.41 ±0.18
A3M	113.07±1.54	141.67±1.68	7.34±0.14
Difference	12.27±1.88	16.87±2.96	0.93±0.19
t’ value	14.62**	12.77**	10.96**
Group DC			
Final	101.40 ±1.77	115.47 ±1.88	6.41 ±0.10
A3M	113.27±1.49	143.60±2.23	7.34±0.14
Difference	11.87±2.25	28.13±1.35	0.93±0.19
t’ value	11.82**	46.67**	10.96**
Group DD			
Final	100.53 ±1.36	115.47 ±1.88	5.49 ±0.17
A3M	113.27±1.49	143.13±2.03	7.34±0.14
Difference	12.74±1.91	27.66±2.89	1.85±0.23
t’ value	14.94**	21.44**	18.01**
Group DE			
Final	95.87 ±1.46	113.07 ±1.62	5.26 ±0.12
A3M	112.93±1.33	143.67±2.26	7.38±0.14
Difference	17.06±2.53	30.60±1.02	2.12±0.22
t’ value	15.10**	67.20**	21.59**
Group DF			
Final	100.01 ±1.63	114.20 ±1.47	5.34 ±0.11
A3M	113.27±1.62	144.07±2.81	7.34±0.14
Difference	13.26±4.59	29.87±3.09	2.00±1.10
‘t’ value	6.47**	21.65**	4.07**

A3M: After three months of withdrawal of supplementation ;** significant at one per cent level

Serum fasting glucose levels

It is inferred from the Table that there was a gradual increase in fasting glucose levels after the withdrawal of supplementation. Group DA showed an increase of 12.07mg/dl. Group DB had an increase of serum fasting glucose of 12.27mg/dl while group DF had an increase of 13.26 mg/dl. Group DD had an increase of 12.74mg/dl while group DE had a maximal increase of 17.06 mg/dl. Three months after the withdrawal of supplementation the values of the serum fasting blood glucose were significantly higher than the final values ($P<0.01$) in all the experimental groups.

Serum postprandial glucose levels

The serum postprandial glucose levels increased after withdrawal of supplementation of individual as well as combined supplementation of wheat germ, bran and grass. Among the supplemented groups, maximum increase of 30.60mg/dl was noted in group DE and the minimum increase of 16.87 mg/dl was noted in group DB. Group DA had an increase of 19.67mg/dl while group DC had an increase of 28.3 mg/dl. Groups DD and DF showed increases of 27.66mg/dl and 29.87mg/dl respectively. Increase in serum postprandial glucose levels in the experimental groups were found to be significant at one per cent level.

Serum glycosylated hemoglobin levels

The mean final glycosylated hemoglobin levels of the subjects in both the experimental and control groups were between 5.26 to 6.4 per cent(good control). The HbA1C had increased in all the groups over a period of three months after the withdrawal of supplements. Group DA had an increase from 6.35 to 7.41 per cent. Group DB showed an increase from 6.41 to 7.34 per cent. Maximum increase from 5.26 to 7.38 per cent was noted in group DE followed by group DD with an increase from 5.49 to 7.34 per cent. Increases in serum glycosylated hemoglobin levels in the experimental groups were found to be significant at one per cent level.

In this study, the individual and combination of wheat germ, bran and grass were supplemented to diabetic subjects for a period of six months and the impact was evaluated. The findings of the clinical trial revealed that wheat germ, bran and grass possessed hypoglycemic, altering lipid peroxidation and antioxidant activity. The impact of supplementation with the supplements were found to be effective in reducing blood glucose, glycosylated hemoglobin, malondialdehyde and copper levels. Further the supplementation also helped to increase the superoxide dismutase, glutathione reductase, glutathione peroxidase, vitamin C, zinc and selenium levels. It could be further emphasized that supplementation of wheat germ and grass was very effective in balancing the diabetic blood parameters followed by wheat bran and grass. It is essential to substantiate that rather than individual supplementation of wheat germ, bran and grass, combination supplements including wheat germ and bran, wheat germ and grass and wheat bran and grass are more effective.

2. Effect of supplementation on selected hyperlipidemic subjects

a. Changes in biochemical picture

(i) Changes in mean serum total cholesterol, triglyceride, LDL-cholesterol, HDL- cholesterol and VLDL-cholesterol levels

Table XX and Figures 23 to 27 gives the mean serum total cholesterol, triglyceride, LDL-cholesterol, HDL- cholesterol and VLDL-cholesterol at the initial and final phase of the study period and the results of the statistical appraisal of the data.

TABLE XX
CHANGES IN MEAN SERUM TOTAL CHOLESTEROL, TRIGLYCERIDES, LDL-C, HDL-C AND VLDL-C LEVELS

(N=15/group)

Groups	Total Cholesterol (mg/dl)	Triglyceride (mg/dl)	LDL cholesterol (mg/dl)	HDL cholesterol (mg/dl)	VLDL cholesterol (mg/dl)
Group HA					
Initial	213.80±2.14	188.53±2.26	142.13±1.64	34.23±1.60	37.70±0.89
Final	178.53±0.64	155.33±1.54	101.60±1.40	43.10±1.30	31.06±0.62
Difference	-35.27±2.15	-33.20±2.46	-40.53±2.29	8.87±2.10	-6.64±0.99
't'value	63.42**	38.91**	68.41**	21.89 **	26.08**
Group HB					
Initial	214.73±0.88	186.13±2.07	143.27±1.33	33.87±1.77	37.26±0.25
Final	170.33±0.49	142.73±1.22	102.07±1.83	41.67±0.82	28.53±0.64
Difference	-44.40±1.30	-43.40±2.06	-41.20±2.21	7.80±1.74	-8.73±0.71
't'value	127.74**	78.34**	72.19**	17.36**	47.28**
Group HC					
Initial	216.07±0.88	185.33±2.13	145.27±4.86	33.73±1.87	37.07±0.39
Final	193.60±0.63	152.53±1.81	128.40±0.99	40.53±0.52	30.47±0.64
Difference	-22.47±1.41	-32.80±3.19	-16.87±5.00	6.80±2.01	-6.60±0.69
't'value	61.82**	29.00**	13.07**	13.12**	37.10**
Group HD					
Initial	204.67±3.60	184.67±3.04	143.47±1.18	35.13±1.78	36.87±1.06
Final	155.80±38.81	132.60±1.40	102.07±1.83	44.67±0.99	26.52±0.28
Difference	-48.87±37.68	-52.07±2.94	-41.40±2.21	9.54±1.60	-10.35±1.08
't'value	5.02**	3.16**	49.33**	18.51**	36.96**
Group HE					
Initial	214.13±1.06	185.27±3.03	143.20±1.26	34.20±1.90	37.07±1.16
Final	185.87±1.13	142.13±1.06	115.73±1.98	39.73±1.44	28.40±0.63
Difference	-28.26±1.22	-43.14±3.07	-27.47±2.50	5.53±2.36	-8.67±0.82
't'value	89.53**	10.10**	42.50**	9.10**	41.11**
Group HF					
Initial	214.07±0.26	185.93±2.46	143.80±1.57	32.87±2.07	37.42±0.17
Final	181.07±1.03	146.00±1.41	107.13±3.54	38.57±0.52	29.20±0.41
Difference	-33.00±1.13	-39.93±2.34	-36.67±4.37	5.70±2.10	-8.22±0.42
't'value	112.72**	37.84**	32.50**	28.98**	67.24**
Group HG					
Initial	215.27±1.79	184.40±2.56	143.60±1.45	32.87±1.41	36.87±0.64
Final	216.00±6.08	186.13±2.07	143.80±1.57	33.60±2.20	36.20±0.86
Difference	0.73±3.06	1.73±3.31	0.20±1.97	-0.73±1.56	-0.67±1.11
't'value	0.93^{NS}	0.34^{NS}	0.39^{NS}	0.65^{NS}	1.16^{NS}
'F' Value	17.60**	532.19**	321.60**	2622.735**	11995.54**

** significant at one per cent level; ^{NS} not significant

TUKEY VALUES FOR SERUM TOTAL CHOLESTEROL

Groups	Means in ascending order	Group HG	Group HC	Group HE	Group HF	Group HA	Group HB	Group HD
		0.73	**22.47**	**28.26**	**33.00**	**35.27**	**44.40**	**48.87**
HG	**0.73**	-	20.74	27.53	32.27	34.34	43.67	48.14
HC	**22.47**		-	5.79	10.53	12.80	21.93	26.40
HE	**28.26**			-	4.74	7.01	16.14	20.61
HF	**33.00**				-	2.27	11.40	15.87
HA	**35.27**					-	9.13	13.60
HB	**44.40**						-	4.47
HD	**48.87**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=15.96$

TUKEY VALUES FOR SERUM TRIGLYCERIDES

Groups	Means in ascending order	Group HG	Group HC	Group HA	Group HF	Group HE	Group HB	Group HD
		1.73	**32.80**	**33.20**	**39.93**	**43.14**	**43.40**	**52.07**
HG	**1.73**	-	31.07	31.47	38.20	41.41	41.67	50.34
HC	**32.80**		-	0.40	7.13	10.34	10.60	19.27
HA	**33.20**			-	6.73	9.94	10.20	18.87
HF	**39.93**				-	3.21	3.47	12.14
HE	**43.14**					-	0.26	8.93
HB	**43.40**						-	8.67
HD	**52.07**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=2.94$

TUKEY VALUES FOR SERUM LDL-CHOLESTEROL

Groups	Means in ascending order	Group HG	Group HC	Group HE	Group HF	Group HA	Group HB	Group HD
		0.20	**16.87**	**27.47**	**36.67**	**40.53**	**41.20**	**41.40**
HG	**0.20**	-	16.67	27.27	36.47	40.33	41.00	41.20
HC	**16.87**		-	10.60	19.80	23.66	24.33	24.53
HE	**27.47**			-	9.20	13.06	13.73	13.93
HF	**36.67**				-	3.86	4.53	4.73
HA	**40.53**					-	0.67	0.87
HB	**41.20**						-	0.20
HD	**41.40**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=3.45$ * Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970

TUKEY VALUES FOR SERUM HDL-CHOLESTEROL

Groups	Means in ascending order	Group HG	Group HF	Group HB	Group HC	Group HE	Group HA	Group HD
		0.27	**5.53**	**5.70**	**6.80**	**7.80**	**8.87**	**9.54**
HG	**0.27**	-	5.26	5.43	6.53	**7.53**	8.60	9.27
HF	**5.53**		-	0.17	1.27	2.27	3.34	4.01
HB	**5.70**			-	1.11	2.10	3.17	3.84
HC	**6.80**				-	1.00	2.07	2.74
HE	**7.80**					-	1.07	1.74
HA	**8.87**						-	0.67
HD	**9.54**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=2.33$

TUKEY VALUES FOR SERUM VLDL- CHOLESTEROL

Groups	Means in ascending order	Group HG	Group HC	Group HA	Group HF	Group HE	Group HB	Group HD
		0.67	**6.06**	**6.64**	**8.22**	**8.67**	**8.73**	**10.35**
HG	**0.67**	-	5.39	5.97	7.55	8.00	8.06	9.68
HC	**6.60**		-	0.04	1.62	2.07	2.13	3.75
HA	**6.64**			-	1.58	2.03	2.09	3.71
HF	**8.22**				-	0.45	0.51	2.13
HE	**8.67**					-	0.06	1.68
HB	**8.73**						-	1.62
HD	**10.35**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=0.90$* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970

Serum total cholesterol levels

The mean initial serum total cholesterol level of the hyperlipidemic subjects in all the groups were in the range of 204.67 to 215.27mg/dl as against the normal value of < 200mg/dl quoted by Raghuram *et al* .,(2007) and Ghafforunisa and Krishnaswamy (2007).Six months of supplementation of wheat products including wheat germ, bran, grass and their combinations brought about a marked reduction in the serum total cholesterol values in all the supplemented groups. Group HA supplemented with wheat germ had a decrease of 35.27mg/dl in the serum total cholesterol levels while group HB supplemented with wheat bran had a reduction of 44.10mg/dl. Group HC supplemented with wheat germ and grass had a minimal decrease of 22.47mg/dl among the supplemented groups while group HD supplemented with wheat germ and bran had the maximum reduction of 48.87mg/dl. Group HE supplemented with wheat germ and grass had a decrease of 28.26mg/dl followed by group HF supplemented with wheat bran and grass which had a decrease of only 33mg/dl in the serum total cholesterol level. Group HG which served as the control group had an increase of 0.73mg/dl over a period of six months without any supplementation. The final values of the serum total cholesterol were found to be significantly lower than the initial values (P<0.01) in all the experimental groups except the control group HG.

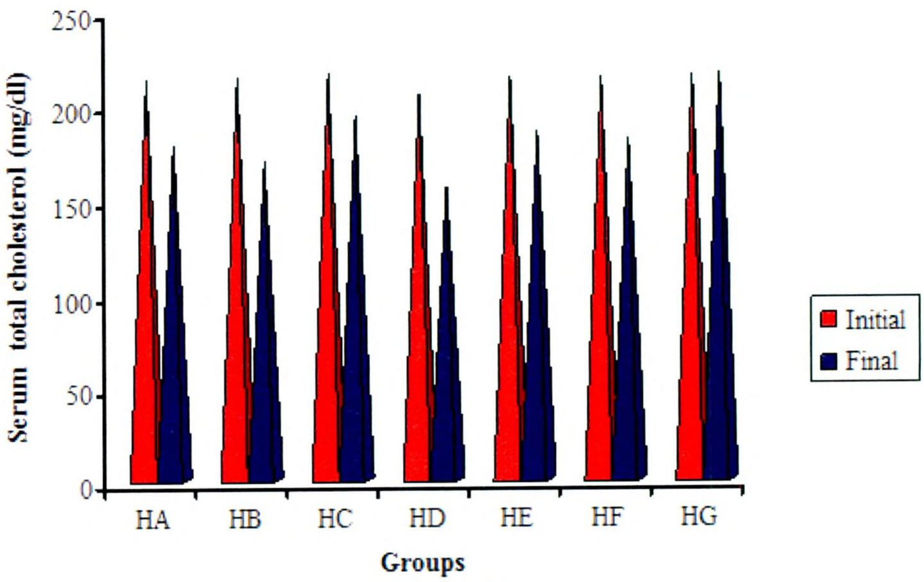

Figure 23

Changes in serum total cholesterol levels

The one way analysis of variance showed a statistically significant difference in the reduction of serum total cholesterol levels among the groups at one per cent level. On applying tukey test to undercover the groups exhibiting the significance it was observed that the mean difference in the serum total cholesterol of the groups HC, HE, HF, HA, HB and HD registered significance when compared with group HG (control group) proving that the groups that were supplemented individually and in combination of wheat germ, bran and grass showed improvements than the non supplemented group

HG. Groups HB and HD supplemented with wheat bran and wheat germ and bran respectively had shown a statistically significant decrease in the mean total cholesterol when compared against the groups HC and HE which were supplemented with wheat grass and wheat germ and grass respectively.

The maximum reduction in mean serum total cholesterol was noted in group HD which had a combined supplementation of wheat germ and bran. Similarly maximum reduction was recorded in group HB which was individually supplemented with wheat bran. Study by Vorster *et al.*, (1986) proved the role of dietary fibre in oat bran through supplementation of oat bran as tablets in lowering total cholesterol. Similarly the dietary fibre in wheat bran might have posed to decrease the serum total cholesterol as 100g of wheat bran contains 42.8g of dietary fiber. According to Khaw *et al.*, (1987) cereal fiber reduces cholesterol on regular intake. Further a study conducted by Ostlund *et al.*, (2003) demonstrated that purified plant sterols (phytosterol) have cholesterol lowering effect. Similarly the phytosterol in wheat germ might have been the useful component in lowering cholesterol levels. Dietary fiber and phytosterol prevents the oxidation of LDL- cholesterol thereby reduces total cholesterol (Mohammed,2000).

Serum triglyceride levels

The mean serum triglyceride levels ranged from 184.40 to 188.53mg/dl whereas normal serum triglyceride level is less than 150mg/dl (Ghafforunissa and Krishnasamy, 2007). The serum triglyceride levels reduced after individual supplementation with wheat germ, bran and grass and also in combination. The final values of the mean serum triglyceride levels in the experimental groups after the supplementation ranged from 132.60 to 155.33mg/dl. Among the supplemented groups, maximum reduction of 52.07mg/dl was noted in group HD supplemented with wheat germ and bran and the minimum reduction of 32.80mg/dl was noted in group HC supplemented with wheat grass. Group HA supplemented with wheat germ had a decrease of 33.20mg/dl while group HB supplemented with wheat bran had a decrease of 43.40mg/dl. Group HE supplemented with wheat germ and grass had a decrease of 43.14 mg/dl almost nearing the decrease recorded in group HB supplemented with only wheat bran. Group HF supplemented with wheat bran and grass showed a decrease of 39.33mg/dl which was much below the decrease observed in group HB supplemented with wheat bran. Decrease in serum triglyceride levels in the experimental groups were found to be significant at one per cent level whereas the change observed in the control group was not statistically significant.

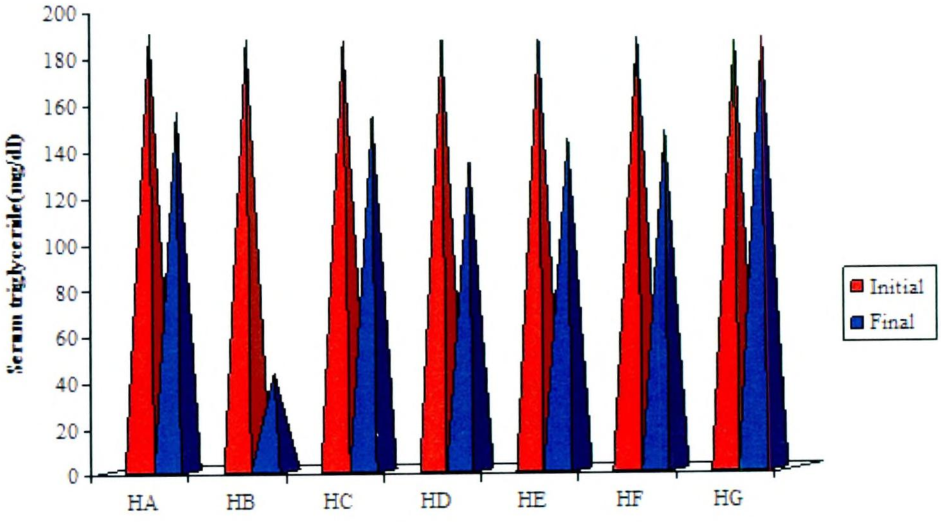

Figure 24
Changes in serum triglyceride levels

Further to 't' test one way analysis of variance was performed and found to be significant at one per cent level between the groups. On applying tukey test to discern the group exhibiting the maximum significance, it was revealed that the mean differences of serum triglyceride in groups HC, HA, HF, HE, HB and HD were significantly greater than that of group HG proving the potential of supplementation. Moreover the mean difference of group HF supplemented with wheat germ and grass showed significantly greater reduction in serum triglyceride levels than group HC and HA supplemented with wheat grass and wheat germ respectively. Groups HE and HB supplemented with wheat bran and grass and wheat bran respectively pictured significant difference when compared with groups HC, HA and HF proving the supplementation of wheat bran and grass and wheat bran respectively to be more efficient over the supplementation of wheat grass , wheat germ individually and wheat bran and grass in combination. The mean differences in group HD supplemented with wheat germ and bran recorded significant reduction of serum triglyceride over the groups HC, HA, HF, HE and HB proving the beneficial supplementation of wheat germ and bran over the supplementation of wheat grass, wheat germ, wheat bran and grass, wheat germ and grass and wheat bran.

Similar to the results of serum total cholesterol maximum reduction of 52.07mg/dl was noted in group HD supplemented with wheat germ and bran where individual supplementation of wheat bran in group HB had a reduction of 43.40 mg/dl and group HA supplemented with wheat germ had a reduction of 38.91mg/dl. The reduction in groups HB and HD supplemented with wheat bran and wheat germ and bran respectively was in par with the study conducted by Swain *et al.*,(1990) where the serum triglyceride levels lowered on supplementation with wheat bran and oat bran on normolipidemic males. The serum triglyceride lowering potential of wheat germ in groups HA, HD and HE could be supported by the study of Martine *et al.*, (1992) where short-term supplementation of wheat germ lowered plasma triglyceride levels in rats.

Serum LDL-cholesterol level

Yet another comprehensible utility of the supplementation was the demonstrably distinct decrease of mean serum LDL-cholesterol levels. The initial mean serum LDL-cholesterol levels in both the experimental and control groups ranged from 142.13 to 145.27mg/dl which was much higher than the normal levels of < 130mg/dl (Raghuram *et al.*, 2007). The impact evaluation showed that the serum LDL – cholesterol had lowered by the individual supplementation of wheat germ, bran and grass as well as combinations of wheat germ and bran, wheat germ and grass and wheat bran and grass over a period of six months. Group HA supplemented with wheat germ had a reduction of 40.53mg/dl while group HB supplemented with wheat bran had a reduction of 41.20mg/dl and group HC supplemented with wheat grass had a minimal decrease of 16.87mg/dl. In the group HD supplemented with wheat germ and bran had a maximum reduction of 41.40mg/dl followed by group HF supplemented with wheat bran and grass with a decrease of 36.67mg/dl. Group HE supplemented with wheat germ and grass had a decrease of 27.47mg/dl in the mean LDL-cholesterol after six months of supplementation. The reduction in the serum LDL-cholesterol level of all the experimental groups except the control group were significant at one per cent level. Further to this tukey test was applied to identify which pair of treatment differed significantly. It could be observed that in all the supplemented groups including HC, HE, HF, HA, HB and HD the mean serum LDL-cholesterol reduced significantly than group HG which served as the control group. Further it could be noted from the tukey table that group HE supplemented with wheat germ and grass showed a significant betterment in the reduction of LDL- cholesterol over group HC supplemented with wheat grass. Group HF supplemented with wheat bran and grass showed a significant reduction of serum LDL- cholesterol when compared with groups HC and HE which were supplemented with wheat grass and wheat germ and grass respectively.

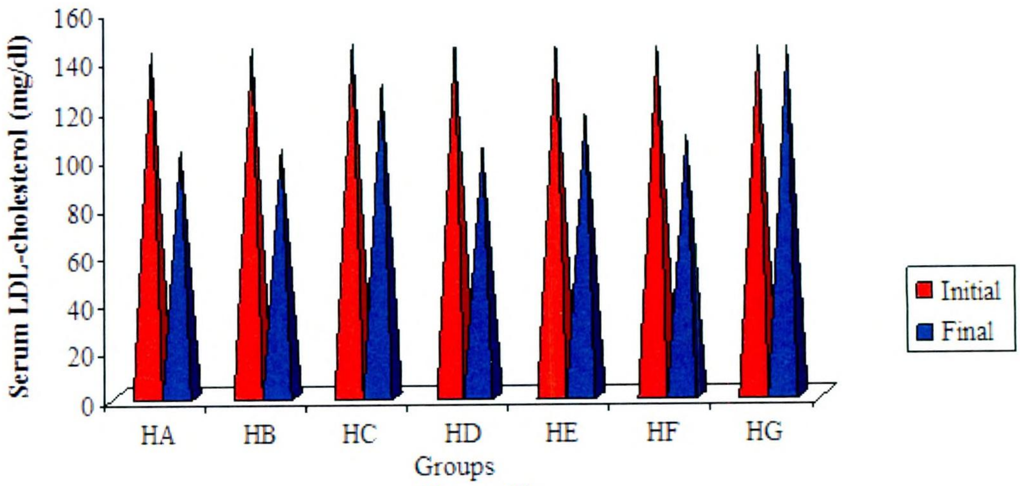

Figure 25
Changes in serum LDL-cholesterol levels

Groups HA and HB supplemented with wheat germ and wheat bran respectively resulted in significant reduction in LDL- cholesterol as against groups HC,HE and HF proving individual supplementation of wheat germ and bran to be even better than supplementation of wheat bran and grass. Groups HA, HB and HD supplemented with wheat germ, wheat bran and wheat germ and bran respectively showed significant reduction of LDL - cholesterol over the groups HC, HE and HF which were supplemented with wheat grass, wheat germ and grass and wheat bran and grass respectively proving supplementation of wheat germ and bran individually or in combination is beneficial.

Through the statistical analysis it is clearly defined that the group DD supplemented with wheat germ and bran had a maximum reduction followed by the individual supplements namely wheat germ and wheat bran. The present investigation is in par with a study conducted by Venketasan *et al*., (2007) where the potential of a fibre cocktail of fenugreek, guar gum and wheat bran reduced oxidative modification of LDL induced by an atherogenic diet in rats. Further a French study found that eating 30 grams, or about a quarter of a cup of raw wheat germ a day for 14 weeks lowered LDL cholesterol by 7.2 per cent (Illman *et al*., 2005). Wheat germ success against LDL cholesterol could stem for its antioxidant powers. Studies show that fiber and antioxidants from foods prevented LDL particles from becoming oxidized (Kacprzaks *et al*.,2002). Oxidized LDL - cholesterol presents a much greater danger to health. When a fat such as LDL- cholesterol undergoes oxidation, it is more prone to collect in blood vessels to form plaque. Over time, the plaques narrows the blood vessels or unleashes a clot, which can result in a heart attack or stroke.

Serum HDL-cholesterol levels

The mean initial HDL-cholesterol levels of the experimental and the control groups was around 35mg/dl whereas normal HDL –cholesterol levels should be more than 50mg/dl (Ghafforunissa and Krishnasamy,2007).There was an increase in the HDL-cholesterol levels on supplementation. Maximum increase of 9.4 mg/dl was observed in group HD supplemented with wheat germ and bran while the least increase of 5.53mg/dl was noted in group HF supplemented with wheat bran and grass. Group HA supplemented with wheat germ had registered an increase of

8.87mg/dl while group HB supplemented with wheat bran recorded an increase of 7.80mg/dl and group HC supplemented with wheat grass had recorded an increase of

5.70mg/dl. The increments in the HDL-cholesterol were found to be significant (P<0.01) in the experimental groups.

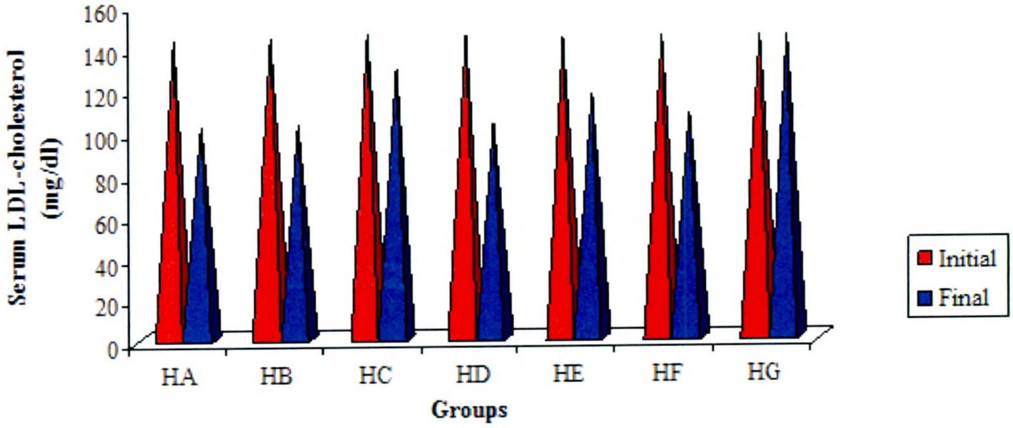

Figure 26
Changes in serum HDL-cholesterol levels

The negligible change from the initial value observed in the control group was not statistically significant. One way analysis of variance conducted to substantiate the significance between the groups also proved to be significant at one per cent level. On applying tukey test to undercover the groups exhibiting maximum significance, it was noted that the mean differences of serum HDL cholesterol of the supplemented groups HF, HB, HC, HE, HA and HD were significantly greater than the group HG which served as the control group. Further it could be noted that the mean incremental differences observed in groups HE and HA supplemented with wheat bran and grass and wheat germ respectively were significant over group HF which was supplemented with wheat bran and grass and moreover the mean incremental difference in group HA was also significant over group HB supplemented with wheat bran. The mean incremental difference of the serum HDL cholesterol of group HD supplemented with wheat germ and bran was significantly greater than the groups HF, HB and HC which were supplemented with wheat germ and bran, wheat bran and wheat grass respectively proving the potential of combined supplementation of wheat germ and bran.

The tukey test revealed that supplementation with wheat germ and bran resulted in significantly greater increments in HDL cholesterol over the supplementation of wheat bran and grass or what bran alone. This could be further supported by a study by Jeane *et al.,(*1987) where HDL cholesterol increased by five per cent for every five per cent decrease in total cholesterol on pectin and wheat bran supplementation. Further according to Brenda *et al.,* (1987), there was a negative correlation between HDL- cholesterol and cardiovascular disease risk. Dietary supplementation with wheat germ and bran have shown a positive effect on HDL cholesterol levels, thereby proving to be an effective treatment for hyperlipidemics.

Serum VLDL-cholesterol

On pondering over the serum VLDL - cholesterol levels of the experimental and the control groups it could be noted that the mean initial VLDL cholesterol levels did vary from 36.87 to 37.70mg/dl which were above the normal values of less than 35mg/dl (Ghafforunisa and Krishnasamy,2007).After supplementation of wheat germ, bran and grass both individually as well as in combinations there was a reduction in VLDL-cholesterol levels in all the experimental groups. Group HA supplemented with wheat germ had a reduction of 6.64mg/dl while group HB supplemented with wheat bran had a reduction of 8.73mg/dl and group HC supplemented with wheat grass showed a minimal reduction of 6.06mg /dl among the supplemented groups. Group HF supplemented with wheat bran and grass had a reduction of 8.22mg/dl while group HE supplemented with wheat bran and grass had a decrease of 8.67mg/dl. Maximum reduction of 10.35mg/dl was observed in group HD supplemented with wheat germ and bran. Students 't' test revealed one per cent level of significance in the reduction observed in all the experimental groups whereas the difference observed in the control group was not found to be significant. The one way analysis of variance showed a statistical significance at one per cent level between the groups. Further to this tukey test helped to identify which pair of treatment differed significantly. The mean difference of serum LDL cholesterol in the groups HC, HA, HF, HE, HB and HD registered significance when compared with group HG (control group) proving that the groups that were supplemented individually and in combination of wheat germ, bran and grass showed improvements than the non- supplemented group HG. Further groups HF,HE and HB supplemented with wheat germ and grass, wheat bran and grass and wheat bran respectively showed a significant reduction in the mean VLDL cholesterol levels ($P<0.01$) over the groups HC and HA which were supplemented with wheat grass and wheat germ respectively. Interestingly group HD supplemented with wheat germ and bran showed a maximum reduction in the VLDL-cholesterol which was significant over groups HC, HA, HF, HE, HB and HD proving the potential of combined supplement of wheat germ and bran.

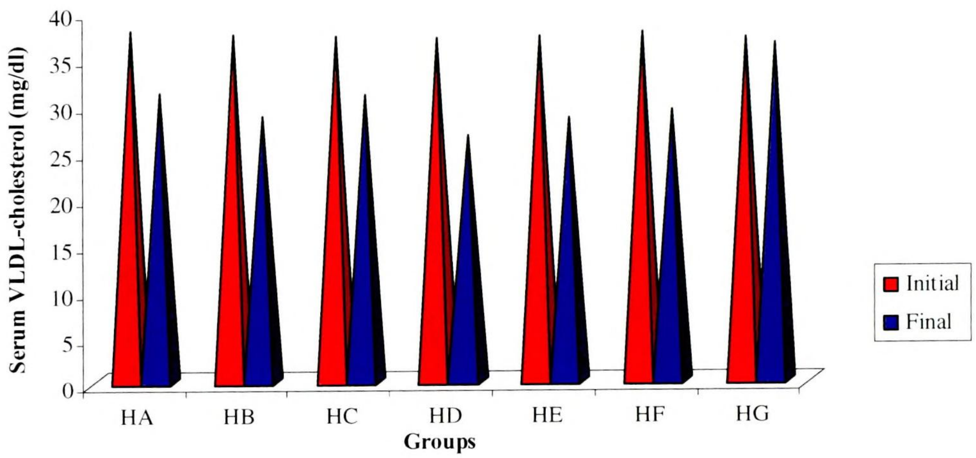

Figure 27
Changes in serum VLDL-cholesterol levels

Similar to other parameters it could be culled out that a combined supplementation of wheat germ with bran has proved to lower serum VLDL-cholesterol levels. The results are in par with the study conducted by Illman *et al.*, (1985) where 30g of wheat germ supplemented for a period of 14 weeks reduced VLDL-cholesterol . Further a study by Venketesan *et al.*,(2007) also has proved a reduction in VLDL cholesterol where fiber cocktail of fenugreek, guar gum and wheat bran were supplemented as atherogenic diet to LDL induced rats.

(ii) Changes in mean Apo enzymes A_1 and B levels

The changes in mean Apo enzymes A_1 and B levels before and after supplementation is given in Table XXI and Figures 28 and 29.

TABLE XXI
CHANGES IN MEAN SERUM APO ENZYME A_1 AND B LEVELS
(N=5/groups)

Groups	Apo enzyme A_1 (mg/dl)	Apo enzyme B (mg/dl)
Group HA		
Initial	103.07±2.18	120.89±1.25
Final	131.74±1.49	101.87±0.83
Difference	28.67±2.94	-19.02±1.60
't'value	36.47**	96.40**
Group HB		
Initial	103.00±3.62	123.03±1.25
Final	119.98±1.76	102.47±2.47
Difference	16.98±3.99	-20.56±2.69
't'value	15.83**	56.81**
Group HC		
Initial	102.93±2.65	121.43±1.33
Final	122.85±1.10	102.47±0.94
Difference	19.92±2.19	-18.96±1.23
't'value	34.02**	116.24**
Group HD		
Initial	103.13±2.13	122.34±1.23
Final	137.88±1.46	103.09±1.27
Difference	34.75±2.88	-19.25±1.33
't'value	45.13**	110.37**
Group HE		
Initial	102.34±2.40	121.23±1.71
Final	127.23±1.49	82.25±1.74
Difference	24.89±2.91	-38.98±2.72
't'value	31.99**	56.19**
Group HF		
Initial	103.33±2.13	121.34±1.12
Final	122.98±1.80	75.41±1.26
Difference	19.65±2.70	-45.93±2.09
't'value	13.37**	82.19**
Group HG		
Initial	103.60±2.37	121.90±1.18
Final	103.46±2.31	120.77±1.79
Difference	-0.14±1.82	-1.13±2.24
't'value	0.29^{NS}	1.88^{NS}
'F' Value	254.26**	820.84**

** significant at one per cent level; ^{NS} not significant

TUKEY VALUES FOR SERUM APOENZYME A_1

Groups	Means in ascending order	Group HG	Group HB	Group HF	Group HC	Group HE	Group HA	Group HD
		0.14	**16.98**	**19.65**	**19.92**	**24.89**	**28.67**	**34.75**
HG	**0.14**	-	16.84	19.51	19.78	24.75	28.53	34.61
HB	**16.98**		-	2.67	2.94	7.91	11.69	17.77
HF	**19.65**			-	0.27	5.24	9.02	15.10
HC	**19.92**				-	4.97	8.75	14.83
HE	**24.89**					-	3.78	9.86
HA	**28.67**						-	6.08
HD	**34.75**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=12.50$

TUKEY VALUES FOR SERUM APOENZYME B

Groups	Means in ascending order	Group HG	Group HC	Group HA	Group HD	Group HB	Group HE	Group HF
		1.13	**18.96**	**19.02**	**19.25**	**20.56**	**38.98**	**45.93**
HG	**1.13**	-	17.83	17.89	18.12	19.43	37.85	44.80
HC	**18.96**		-	0.06	0.29	1.60	20.02	26.97
HA	**19.02**			-	0.23	1.54	19.96	26.91
HD	**19.25**				-	1.31	19.73	26.68
HB	**20.56**					-	18.40	25.37
HE	**38.98**						-	6.95
HF	**45.93**							-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=2.98$ *Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970

Serum apoenzyme A_1

The mean initial serum apoA_1level of the hyperlipidemic subjects in all the groups were in the range of 102.34 to 103.60 mg/dl as against the normal value of 107 - 177 mg/dl as quoted by Rifai (1988) and Gordon *et al.,* (1977).Six months of supplementation of wheat products including wheat germ, bran ,grass and their combinations brought about a marked increase in the serum apoenzyme A_1 levels in all the supplemented groups. Group HA supplemented with wheat germ had an increase of 28.67g/dl in the serum apoenzyme A_1 levels while group HB supplemented with wheat bran had a minimal increase of 16.98 mg/dl. Group HC supplemented with wheat germ and grass had an increase of 19.92mg/dl while group HD supplemented with wheat germ and bran had the maximum increase of 34.75mg/dl. Group HE supplemented with wheat germ and grass had an increase of 24.89mg/dl followed by group HF supplemented with wheat bran and grass with an of only 19.65mg/dl of serum apoenzyme A_1 levels. Group HG which served as the control group had an increase of 0.14mg/dl over a period of six months. The final values were significantly greater than the initial values ($P<0.01$) in all the experimental groups except the control group HG where the difference was not statistically significant.

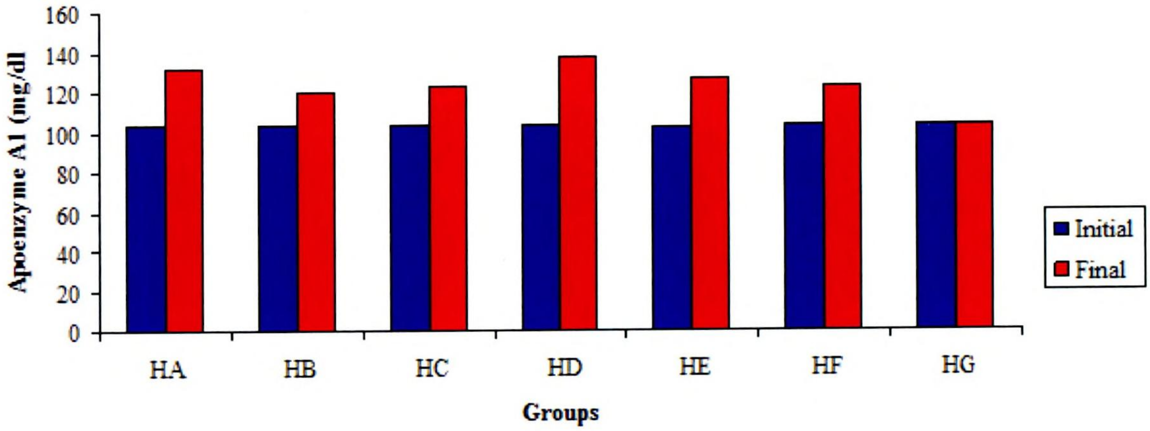

Figure 28
Changes in serum Apoenzyme A1 levels

The one way analysis of variance showed a statistically significant difference in the reduction of serum apoenzymes A_1 levels among the seven groups at one per cent level. On applying tukey test to undercover the groups exhibiting the significance it was observed that the mean incremental difference of serum apoenzyme A_1 in the groups HF, HB, HC, HE, HA and HD was significant over the control group HG . Group HD supplemented with wheat germ and bran had shown a statistically significant increase in the mean apoenzyme A_1 over the groups HF and HB which were supplemented with wheat germ and grass and wheat bran respectively. The maximum increase in mean serum apoenzyme A_1 was noted in group HD which had a combined supplementation of wheat germ and bran. Similarly maximum increase was recorded in group HB which was individually supplemented with wheat bran. Study by Brussard *et al.,* (1980) demonstrated an increase of enzyme Apo A_1 with increase in the HDL concentration. Further the study by Cara *et al.*,(1992) proved the increase of apoenzyme A_1 level when fed with oat bran, rice bran, wheat fiber, and wheat germ on postprandial lipemia in healthy adults.

Serum apoenzyme B

On contemplating at the initial mean serum apoenzyme B levels of the subjects in the experimental and control groups, it is apparent that levels did range from 120.89 to 123.03mg/dl and normal serum apoenzyme B level ranges from 70-130mg/dl (Rifai,1988). The serum apoenzyme B levels reduced after individual as well as combined supplementation with wheat germ, bran and grass. The final values of the mean serum apoenzyme B levels in the experimental groups after the supplementation ranged from 75.41 to 120.77 mg/dl. Among the supplemented groups, maximum reduction of 83.09mg/dl was noted in group HD supplemented with wheat germ and bran and the minimum reduction of 19.25mg/dl was noted in group HC supplemented with wheat grass. Group HA supplemented with wheat germ had a decrease of 19.02mg/dl while group HB supplemented with wheat bran had a decrease of 20.56mg/dl. Group HE supplemented with wheat germ and grass had a decrease of 38.95 mg/dl. Group HF supplemented with wheat bran and grass showed a decrease of 45.93mg/dl which was much below the decrease observed in group HB supplemented with wheat bran.

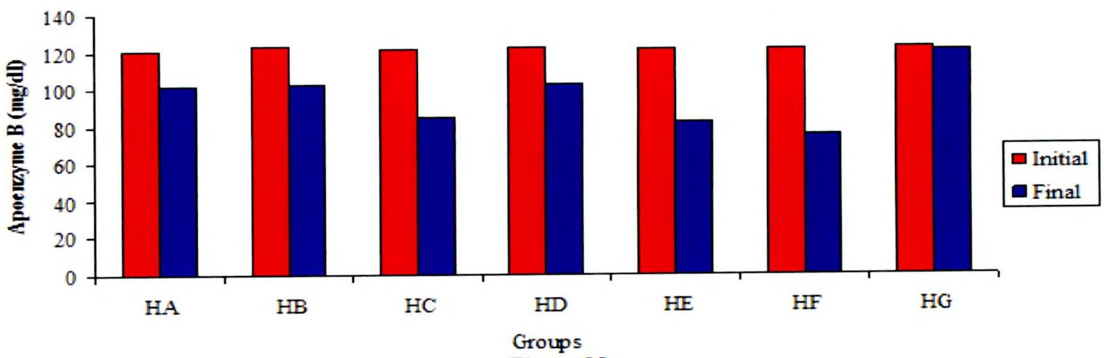

Figure 29
Changes in serum apoenzyme B levels

Decreases in serum apoenzyme B level in the experimental groups were found to be significant at one per cent level whereas the change observed in the control group was not statistically significant. Further to 't' test, one way analysis of variance was performed and found to be significant at one per cent level between the groups. On applying tukey test to discern the group exhibiting the maximum significance, it was revealed that the mean differences of serum apoenzyme B in groups HC, HE, HF, HA, HB and HD were significantly lower than that of group HG proving the potential of supplementation. Moreover the mean difference of groups HF supplemented with wheat germ and grass was significantly greater than the group HC and HE supplemented with wheat grass and wheat germ and bran respectively. Groups HA and HB supplemented with wheat germ and wheat bran respectively pictured significant difference when compared with groups HC, HE and HF proving the beneficial impact of supplementation of wheat grass , wheat germ and grass and wheat bran and grass respectively .The mean differences in group HD supplemented with wheat germ and bran recorded significant reduction of serum apoenzyme B over the groups HC, HE and HF proving the benefits of supplementing wheat germ and bran over wheat grass, wheat germ and grass and wheat bran and grass respectively.

Similar to the results of serum LDL – cholesterol, maximum reduction (82.19mg/dl) of apoenzyme B was noted in group HD supplemented with wheat germ and bran. Individual supplementation of wheat bran in group HB had a reduction of 82.17mg/dl and group HA supplemented with wheat germ had a reduction of 79.20mg/dl. Similar results were obtained by Cara *et al.*,(1992) where the serum apoenzyme B lowered on supplementation with wheat bran and oat bran on normolipidemic males.

(iii) Changes in mean lipoprotein ratios

Table XXII presents the changes in the lipoprotein concentrations ratios before and after the supplementation study.

TABLE XXII

CHANGES IN MEAN LIPOPROTEIN RATIO

Groups	LDL-C/HDL-C*		HDL/Total cholesterol**		ApoB /ApoA-I***	
	Initial	Final	Initial	Final	Initial	Final
HA	4.15	2.91	0.16	0.20	0.84	0.57
HB	4.19	2.94	0.16	0.20	0.84	0.72
HC	4.31	3.72	0.16	0.19	0.84	0.60
HD	4.36	3.07	0.16	0.21	0.84	0.53
HE	4.23	3.33	0.16	0.19	0.85	0.72
HF	4.09	3.02	0.16	0.20	0.85	0.72
HG	4.37	4.28	0.15	0.16	0.86	0.85

Normal range * Desired level (low risk): 0.5-3.0 ; Borderline level (moderate risk): 3.0-6.0; Elevated level (high risk): >6.0

** Ideal level: ≥0.24;low:<0.24;very dangerous:<0.10

***apoB/apoA-I ratio <0.75

The mean initial LDL-C to HDL-C ratios of the experimental and the control groups ranged from 4.09 to 4.37 as against the desired range of 0.5 to 3.0. Group HA supplemented with wheat germ had a decrease of 1.24 followed by group HB by 1.25. Maximum reduction in the ratio of 1.29 in the ratio was noted in group HD supplemented with wheat germ and bran and minimal reduction was noted in group HC supplemented with wheat grass.

The HDL to total cholesterol ratio ranged from 0.15 to 0.16 initially and after the supplementation period the final values ranged from 0.16 to 0.21 whereas the ideal value is ≥0.24. Similarly the apoB to $ApoA_1$ ratio ranged from 0.84 to 0.86 initially and after six months of supplementation the ratio ranged between 0.53 and 0.72 which was found to be well within the normal range of reference i.e <0.75.

Thus the findings of the study revealed that individual and combined supplementation of wheat germ ,bran and grass for a period of six months reduced serum total cholesterol, triglycerides, LDL-cholesterol and VLDL cholesterol levels and increased HDL-cholesterol levels. It is highly encouraging to note that the combination of wheat germ and bran (60g of wheat germ and 20g of wheat bran) brought about the maximum benefits in lowering the lipid profile of the hyperlipidemic groups.

(iv) Comparison of the impact of different supplements on the blood parameters assessed

Table XXIII summarizes the different supplements used for supplementation study on the blood parameters assessed on the experimental groups.

TABLE XXIII
COMPARISON OF THE DIFFERENT SUPPLEMENTS ON BLOOD PARAMETERS

Parameters	Rank					
	1	2	3	4	5	6
Total cholesterol (mg/dl)	HD	HB	HA	HF	HE	HC
Serum triglyceride (mg/dl)	HD	HB	HE	HF	HA	HC
Serum LDL-cholesterol (mg/dl)	HD	HB	HA	HF	HE	HC
Serum HDL-cholesterol (mg/dl)	HD	HA	HE	HC	HB	HF
Serum VLDL- cholesterol (mg/dl)	HD	HB	HE	HF	HA	HC
Serum Apoenzyme A_1(mg/dl)	HD	HA	HE	HC	HB	HF
Serum Apoenzyme B (mg/dl)	HD	HB	HA	HF	HE	HC

From the above Table it could be depicted that group HD supplemented with wheat germ and bran proved to decrease total cholesterol, serum triglyceride, LDL-cholesterol, VLDL-cholesterol and Apoenzyme A_1 and increased HDL-cholesterol and Apoenzyme B levels. Further group HB supplemented with wheat bran was ranked second in reducing total cholesterol, serum triglyceride, LDL-cholesterol. It could also be noted that group HC and HF supplemented with wheat grass and wheat bran and grass proved minimum beneficiary in maintaining the blood profile in hyperlipidemia.

(v) Changes in blood parameters after withdrawal of supplementation in hyperlipidemic subjects

Table XXIV gives the final and three months after the withdrawal of supplementation data on total cholesterol , serum triglyceride, LDL-cholesterol and HDL-cholesterol levels.

TABLE XXIV
CHANGES IN BLOOD PARAMETERS ATER THE WITHDRAWAL OF SUPPLEMENTATION **(N=5)**

Groups	Total cholesterol (mg/dl)	Serum triglyceride (mg/dl)	Serum LDL-cholesterol (mg/dl)	Serum HDL-cholesterol (mg/dl)
Group HA				
Final	178.53±0.64	155.33±1.54	101.60±1.40	43.10±1.30
A3M	189.56±1.12	178.44±2.01	126.35 ±2.02	37.22±0.98
Difference	11.03±2.21	23.11±1.26	24.75±1.56	-5.88±1.63
't' value	11.18**	41.08**	35.54**	8.08**
Group HB				
Final	170.33±0.49	142.73±1.22	102.07±1.83	41.67±0.82
A3M	188.86±1.43	167.55±1.06	124.73±1.97	35.86±1.12
Difference	18.53±2.01	24.82±1.89	22.66±1.77	-5.81±1.53
t' value	20.65**	29.42**	28.68**	8.51**
Group HC				
Final	193.60±0.63	152.53±1.81	128.40±0.99	40.53±0.52
A3M	186.43±1.23	169.49±1.86	136.77±1.65	35.76±1.12
Difference	7.17±1.35	16.96±1.65	8.37±2.31	-4.77±1.56
t' value	11.90**	23.02**	8.12**	6.84**
Group HD				
Final	155.80±38.81	132.60±1.40	102.07±1.83	44.67±0.99
A3M	179.54±2.01	165.43±2.12	129.45±1.78	32.54±1.10
Difference	23.74±1.22	32.83±2.33	27.38±1.56	-12.13±2.48
t' value	43.59**	31.56**	39.96**	10.96**
Group HE				
Final	185.87±1.13	142.13±1.06	115.73±1.98	39.73±1.44
A3M	198.53±1.36	169.45±1.88	131.44±1.25	33.68±1.89
Difference	12.66±1.55	27.32±2.03	15.71±1.86	-6.05±1.44
t' value	18.30**	30.15**	18.92**	9.41**
Group HF				
Final	181.07±1.03	146.00±1.41	107.13±3.54	38.57±0.52
A3M	195.12±2.13	171.45±2.41	136.44±2.01	31.45±1.03
Difference	14.05±1.53	25.45±1.45	29.31±1.63	-7.12±1.85
't' value	20.57**	65.64**	67.25**	8.62**

A3M: After three months of withdrawal of supplementation ;** significant at one per cent level

Serum total cholesterol levels

It is inferred from the Table that there was an increase in the total cholesterol levels after the withdrawal of supplementation. Group HA showed an increase of 11.03mg/dl. Group HB had an increase of serum total cholesterol of 18.53mg/dl while group HF had an increase of 14.05 mg/dl. Group HD had a maximal increase of 23.74mg/dl while group HE had an increase of 12.66 mg/dl. After three months of withdrawal of supplementation values of the serum total cholesterol raised significantly compared to the final values ($P<0.01$) in all the experimental groups.

Serum triglyceride levels

The serum triglyceride levels increased after withdrawal of supplementation of individual as well as combined supplementation of wheat germ, bran and grass. Among the supplemented groups, maximum increase of 32.83mg/dl was noted in group HD and the minimum increase of 16.96 mg/dl was noted in group HC. Group HA had an increase of 23.11mg/dl while group HB had an increase of 24.82 mg/dl. Group HE had an increase of 27.32 mg/dl. Group HF showed an increase of 25.45mg/dl. Increase in serum triglyceride levels in the experimental groups were found to be significant at one per cent level.

Serum LDL-cholesterol levels

It is inferred from the Table that there was an increase in the LDL-cholesterol levels after the withdrawal of supplementation. Group HA showed an increase of 24.75mg/dl. Group HB had an increase of serum LDL-cholesterol of 22.66mg/dl while group HF had an increase of 29.31 mg/dl. Group HD had a maximal increase of 27.38mg/dl while group HE had an increase of 15.71 mg/dl. The after three months of withdrawal of supplementation values of the serum LDL-cholesterol were significantly higher than the final values ($P<0.01$) in all the experimental groups.

Serum HDL-cholesterol levels

The serum HDL-cholesterol levels decreased after withdrawal of supplementation of individual as well as combined supplementation of wheat germ, bran and grass. Group HA showed a decrease of 5.88mg/dl. Group HB had an increase of serum HDL-cholesterol of 5.81mg/dl while group HF had a decrease of 7.12 mg/dl. Group HD had a maximal decrease of 12.13mg/dl while group HE had a decrease of 6.05 mg/dl. The serum fasting blood glucose values after three months of withdrawal of supplementation were found to be significantly higher than the final values ($P<0.01$) in all the experimental groups.

Thus, the findings of the study revealed that individual and combination of wheat germ, bran and grass supplemented to hyperlipidemic subjects for a period of six months reduced total cholesterol, serum triglyceride, LDL-cholesterol, VLDL-cholesterol and increased HDL-cholesterol levels. It was observed that than the individual supplementation of wheat germ, bran and grass and combination of supplements including wheat germ and bran, wheat germ and grass and wheat bran and grass was more effective. It could be further emphasized that supplementation of wheat germ and bran was very effectively in balancing the hyperlipidemic blood than the other combined supplements.

3. Effect of individual and combined supplementation of wheat germ, bran and grass on selected tuberculosis subjects

a. Changes in the physiological symptoms

The physiological symptoms before and after treatment of the tuberculosis subjects is depicted in Table XXV.

TABLE XXV
CHANGES IN PHYSIOLOGICAL SYMPTOMS OF TUBERCULOSIS SUBJECTS*

Symptoms	Group TA		Group TB		Group TC		Group TG	
	I	F	I	F	I	F	I	F
Cough with sputum	13	2	15	5	15	3	15	12
Dry cough	12	5	13	3	15	9	15	12
Loss of weight	14	1	13	-	15	-	15	2
Loss of appetite	13	-	5	-	15	-	15	15
Chest pain	10	4	10	4	15	6	15	13
Shortness of breath	11	3	14	4	10	2	15	12
Low grade fever	13	-	13	-	12	1	13	13
Night sweats	11	4	10	3	11	9	10	10
Anorexia	14	-	11	2	15	-	15	15
Clubbing	1	1	1	1	4	4	3	5
Peripheral lymphadenopathy	2	2	3	3	2	2	7	9
Hemoptysis	1	1	-	-	-	-	8	8

* Multiple responses

Of the various clinical parameters indicative of tuberculosis, cough with sputum was present in 13 subjects in group TA, 15 subjects each in groups TB, TC and TD. After the supplementation it was noted that group TA supplemented with wheat germ, group TB supplemented with wheat grass and group TC supplemented with wheat germ and grass had only two, five and three subjects with cough and sputum respectively. Dry cough was prevalent among 12,13 and 15 subjects in groups TA,TB and TC respectively whereas after the supplementation period it was noted that only five, three and five subjects respectively had dry cough. Loss of weight greatly reduced at the end of the study among all the supplemented subjects. The loss of appetite which was present initially disappeared completely in the supplemented groups after the supplementation period. Chest pain, shortness of breath, low grade fever and anorexia was present in most of the subjects before supplementation which was not present after the supplementation. Such improvements were not observed among the control group subjects.

b. Changes in biochemical picture

(i) Changes in the total protein, albumin and globulin levels

Table XXVI and Figures 30 and 31 presents the changes in the serum total protein and albumin levels observed among the tuberculosis subjects after supplementation.

TABLE XXVI
CHANGES IN MEAN SERUM TOTAL PROTEIN AND ALBUMIN LEVELS (N=15/group)

Groups	Total protein (g/dl)	Albumin (g/dl)
Group TA Initial	7.4±1.24	3.0±0.13
Final	8.8±1.46	3.5±0.03
Difference	1.4±0.96	0.5±0.22
't' value	14.50**	9.68**
Group TB Initial	7.4±0.74	3.1±0.51
Final	8.4±5.36	3.3±0.68
Difference	1.0±0.20	0.2±0.15
t' value	2.26**	5.16**
Group TC Initial	7.6±2.77	3.0±0.26
Final	9.4±1.12	3.7±0.19
Difference	1.8±3.53	0.7±0.31
t' value	9.25**	8.73**
Group TD Initial	7.1±1.24	3.1±0.91
Final	7.5±2.51	3.2±0.92
Difference	0.4±.2.12	0.1±0.18
t' value	2.46*	2.15*
'F' value between groups	8.83**	13.07**

** Significant at one per cent level;* Significant at five per cent level

TUKEY VALUES FOR SERUM TOTAL PROTEIN

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		0.4	**1.0**	**1.4**	**1.8**
TD	**0.4**	-	0.6	1	1.4
TB	**1.0**		-	0.4	0.8
TA	**1.4**			-	0.4
TC	**1.8**				-

alpha =0.5 ;D_{min}=Q^*_t(sqrt($MS_{within\ group}$))/(sqrt(S))=1.1

TUKEY VALUES FOR SERUM ALBUMIN

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		0.1	**0.2**	**0.5**	**0.7**
TD	**0.1**	-	0.1	0.4	0.6
TB	**0.2**		-	0.3	0.5
TA	**0.5**			-	0.2
TC	**0.7**				-

alpha =0.5D_{min}=Q^*_t(sqrt($MS_{within\ group}$))/(sqrt(S))=0.6* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970

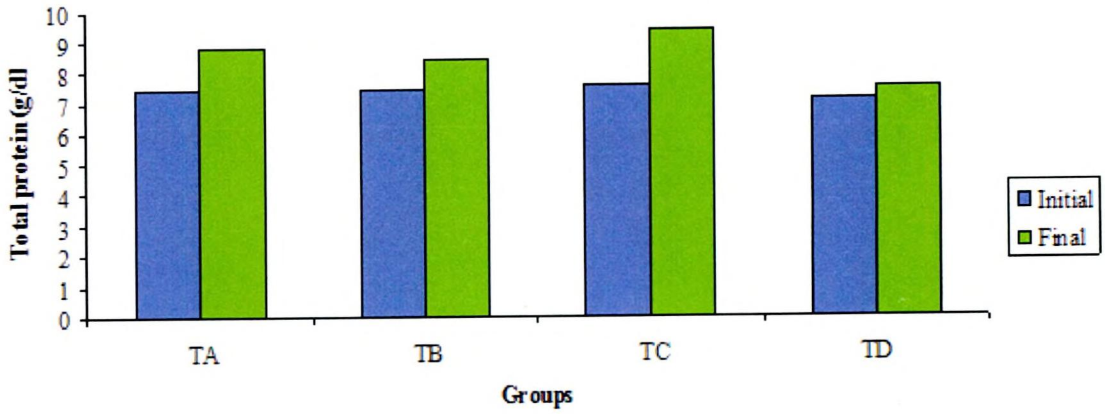

Figure 30
Changes in serum total protein level

Serum total protein levels

The mean initial serum total protein levels of the experimental and the control groups ranged from 7.1 to 7.6 g/dl which was in the lower range of the normal serum total protein levels i.e. 7-9g/dl (Varun *et al.*,2007).There was an increase in the serum total protein levels on supplementation. Maximum increase of 1.8 g/dl was observed in group TC supplemented with wheat germ and grass while an increase of 1.4g/dl was noted in group TA supplemented with wheat germ only. Group TB supplemented with wheat bran had registered an increase of 1g/dl. The final values of the serum total protein when compared with the initial values were found to be significantly greater (P<0.01) in the experimental and control groups. One way analysis of variance conducted to substantiate the significance between the groups also proved to be significant at one per cent level. On applying tukey test to undercover the groups exhibiting maximum significance, it was noted that only the mean differences of serum total protein of group TC supplemented with wheat germ and grass was significantly greater than that of group TD which served as the control group.

It was recorded that supplementation with wheat germ and grass resulted in significantly greater increments in serum total protein over the supplementation of wheat germ and wheat grass individually.

Serum albumin levels

The serum albumin levels of the experimental and the control groups ranged between 3 and 3.1g/dl whereas the normal range of the serum albumin is 35-50g/l as quoted by Varun *et al.*,(2007). On supplementation with wheat products including wheat germ, grass and their combination there was an improvement in the serum albumin levels after six months. Group TC supplemented with wheat germ and grass had a maximum increase of 0.7g/dl. Group TA supplemented with wheat germ had an increase of 0.5g/dl followed by group TB supplemented with wheat grass with a mean increase of 0.2g/dl. The changes observed in the mean serum albumin levels in the experimental and the control group were found to be significant at one per cent level .One way analysis of variance also proved to be significant at one per cent level between the four groups compared. The tukey test performed to cull out the most significantly differed group revealed that only the mean difference in group TC supplemented with wheat germ and grass was significantly greater over group TD which served as the control group.

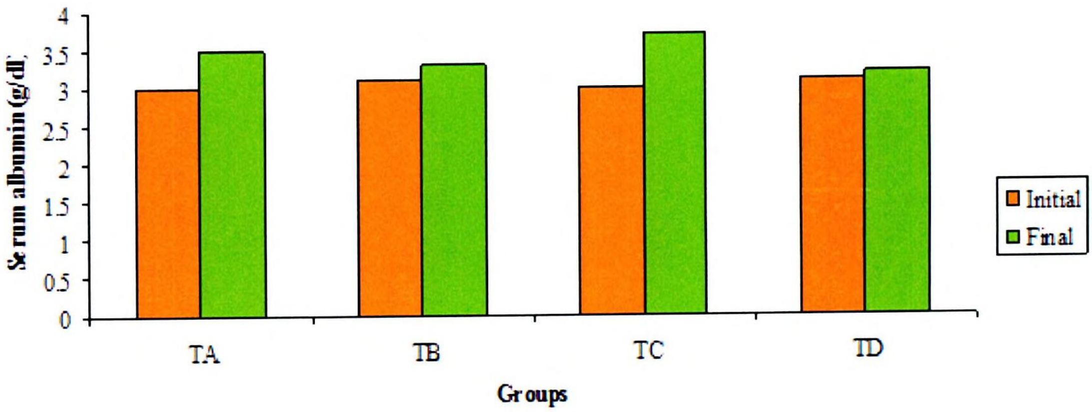

Figure 31
Changes in serum albumin level

In the present study lower levels of serum total protein and albumin present in the pulmonary tuberculosis subjects before the supplementation study. This agrees with Sasaki *et al.*,(1999) who stated that albumin and total protein were significantly lower in pulmonary tuberculosis. The increase in the total protein and the serum albumin levels on supplementation could be compared with the study of Robert *et al*, (2002) where there was a slight increase in total protein and serum albumin on supplementation of wheat germ to multi drug resistant tuberculosis subjects. Arinola and Igbi (1998) reported high levels of IgG and IgM in pulmonary tuberculosis. Nagayama *et al.*,(1999) also stated that hyperglobulinaemia in tuberculosis is one of the predictive factors for the development of residual pleural thickening in tuberculous pleurisy.

(ii) Changes in the TLC and CD_4 levels

Table XXVII and Figures 32 and 33 shows the changes in the TLC and CD_4 of tuberculosis subjects after supplementation.

TABLE XXVII
CHANGES IN SERUM TLC AND CD_4 LEVELS

(N=15/group)

Groups	TLC(cells/m^3)	CD_4(cells/m^3)
Group TA Initial	8615.87±214.81	324.13±3.83
Final	7747.47±114.53	501.73±0.96
Difference	-868.40±112.17	177.60±3.83
't' value	28.95**	173.43**
Group TB Initial	8863.80±210.13	334.07±3.85
Final	8298.73±111.79	477.40±5.32
Difference	-565.07±141.35	143.33±3.25
t' value	14.94**	164.94**
Group TC Initial	8423.73±218.23	322.93±2.25
Final	7335.13±115.00	573.93±0.25
Difference	-1088.60±119.19	251.00±3.56
t' value	34.16**	263.69**
Group TD Initial	8088.60±212.23	332.80±1.61
Final	7864.40±111.20	441.40±3.23
Difference	-224.20±129.25	108.60±3.29
t' value	6.49**	123.45**
'F' value between groups	16.60**	12.18**

** Significant at one per cent level

TUKEY VALUES FOR SERUM TLC

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		108.60	143.33	177.60	251.00
TD	108.60	-	34.73	69.00	142.40
TB	143.33		-	34.27	107.67
TA	177.60			-	73.40
TC	251.00				-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=126.61$

TUKEY VALUES FOR SERUM CD_4

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		224.20	565.07	868.00	1088.60
TD	224.20	-	340.87	643.80	864.40
TB	565.07		-	302.93	523.53
TA	868.00			-	220.60
TC	1088.60				-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=3.42$

Serum TLC levels

The mean initial serum TLC of the tuberculosis subjects in all the groups were in the range of 8088.60 to 8863.80 cells/m^3 as against the normal value of 1891-4800 cells/ m^3 as quoted by Robert and Jarlier,(2002). Six months of supplementation of wheat products including wheat germ, grass and their combination brought about a marked reduction in the serum TLC values in all the supplemented groups. Group TA supplemented with wheat germ had a decrease of 868.40 cells/m^3 in the serum TLC levels while group TB supplemented with wheat grass had a reduction of 565.07 cells/m^3 only. Group TC supplemented with wheat germ and grass had a maximal decrease of 1088.60 cells/m^3 among the supplemented groups. Group TD which served as the control group had a decrease of 224.20 cells/m^3 over a period of six months without any supplementation. The final values of the serum TLC were significantly lower than the initial values (P<0.01) in the experimental and control groups.

The one way analysis of variance also showed a statistically significant difference in the reduction of serum TLC levels among the four groups at one per cent level. Tukey test undercoverd that groups TB, TA and TC registered significantly lower reduction than the control group proving that the supplementation of wheat germ and grass individually and in combination resulted in improvements over the non supplemented group TD. Group TA supplemented with wheat germ had shown a statistically significant decrease in the mean TLC levels when compared against the group TB supplemented with wheat grass. Further the mean differences of group TC supplemented with wheat germ and grass showed significance over groups TB and TA supplemented with wheat grass and germ proving the combined supplementation of wheat germ and grass to be effective.

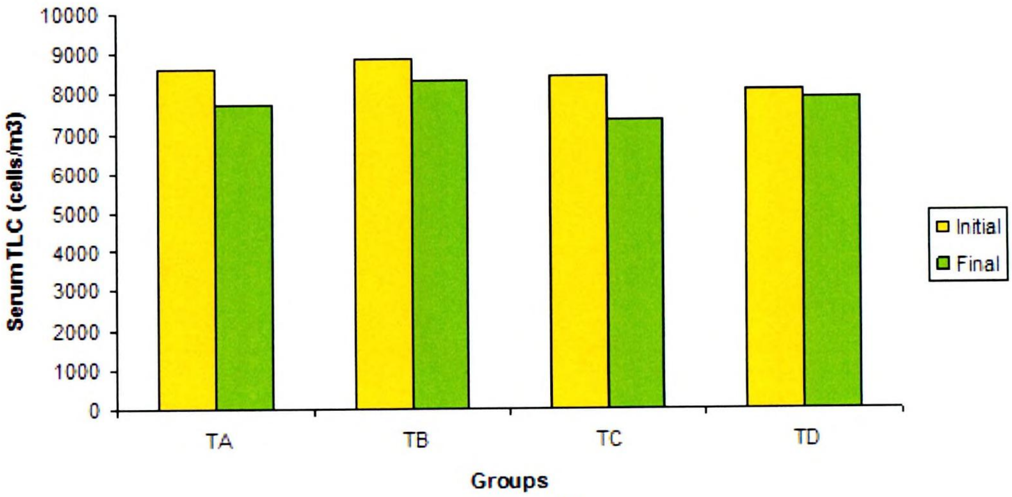

Figure 32
Changes in serum TLC levels

The elevated levels of TLC levels before the start of the supplementation study may be due to the chronic infection as TLC is accelerated in many diseases including pulmonary tuberculosis (Nefedov *et al.*,2007).

Serum CD_4 count

The initial mean CD_4 count of the selected subjects in the experimental and control groups ranged from 322.93 to 334.07 cells/m^3 whereas normal CD_4 level ranges from 690-2420cells/m^3 (Robert and Jarlier,2002).. The CD_4 levels increased after individual as well as combined supplementation with wheat germ and grass. The final values of the mean CD_4 levels in the experimental groups after the supplementation ranged from 441.40 to 573.93 cells/m^3. Among the supplemented groups, maximum increase of 251 cells/m^3 was noted in group TC supplemented with wheat germ and grass and the minimum increase of 143.33 cells/m^3 was noted in group TB supplemented with wheat grass. Group TA supplemented with wheat germ had an increase of 177.60 cells/$m^{3.}$ Increase in the initial and final values of CD_4 count in the experimental and control groups were found to be significant at one per cent level .

One way analysis of variance indicated significance between the groups at one per cent level. On applying tukey test it was observed that the mean difference of TLC of the groups TB, TA and TC were significantly higher than the control group TD. Group TA supplemented with wheat germ had shown a significantly improved TLC levels when compared to group TB supplemented with wheat grass. Further the mean difference in group TC supplemented with wheat germ and grass showed significance over the groups TB and TA supplemented with wheat grass and germ proving the combined supplementation of wheat germ and grass to be effective.

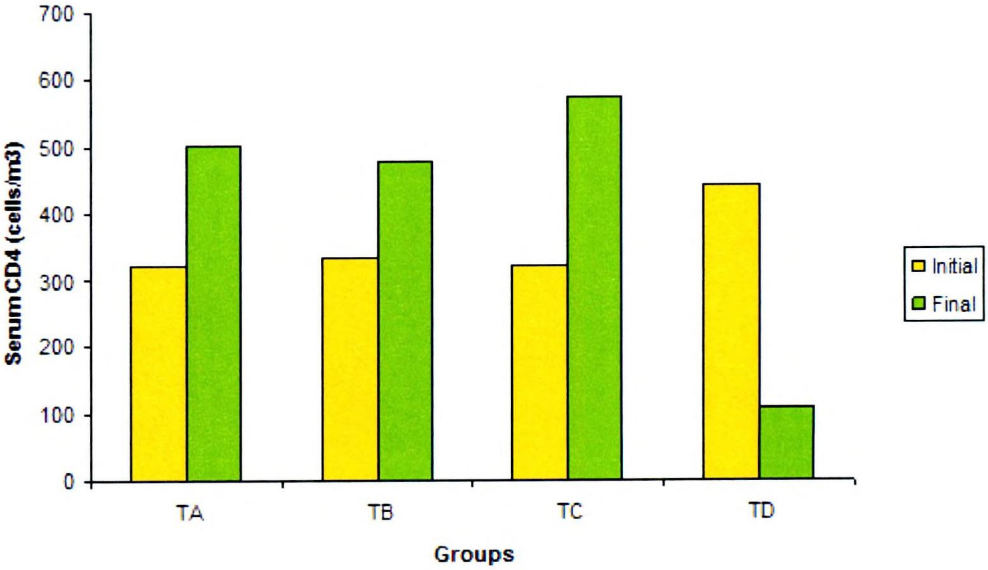

Figure 33
Changes in serum CD_4 levels

The increase in CD_4 count in all the supplemented groups showed a positive impact of supplementation. The study by Kalser (2006) reveals that as immunity builds in the body TLC count decreases with increase in CD_4 counts in tuberculosis affected HIV subjects. Similarly in the present study, there was a decrease in the TLC count with increase in CD_4 count proving the potential of combined supplementation of wheat germ and grass.

(iii) Serum superoxide dismutase, malondialdehyde, glutathione reduced, glutathione peroxidase and total antioxidant activity

The changes in mean serum superoxide dismutase, malondialdehyde, glutathione reduced ,glutathione peroxidase and total antioxidant activity after supplementation is given in Table XXVIII and Figures 34 to 38.

TABLE XXVIII
CHANGES IN MEAN SERUM SUPERODIXE DISMUTASE, MALONDIALDEHYDE, GLUTATHIONE REDUCED, GLUTATHIONE PEROXIDASE AND TOTAL ANTIOXIDANT ACTIVITY LEVELS

(N=15/group)

Groups	Superoxide dismutase (U/g Hb)	Malondialdehyde (µM/L)	Glutathione reduced (µM/L)	Glutathione peroxidase (U/g Hb)	Total antioxidant activity (mmol/l)
Group TA					
Initial	2785.42±5.87	4.26±0.15	44.23±0.12	22.53±1.18	0.53±0.98
Final	2935.65±2.19	2.01±0.01	53.48±1.21	42.27±0.79	0.98±0.42
Difference	150.23±3.27	-2.25±0.16	9.25±0.76	19.74±1.23	0.45±0.06
't' value	177.80**	52.59**	45.52**	72.15**	28.05**
Group TB					
Initial	2695.59±4.56	4.31±0.11	45.13±1.10	22.40±1.28	0.57±0.2
Final	2838.85±3.23	2.34±0.02	50.36±1.12	45.13±1.12	0.92±0.01
Difference	143.26±2.57	-1.97±0.14	5.23±0.99	22.73±1.22	0.35±0.01
t' value	215.73**	52.63	19.76**	8.37**	130.90**
Group TC					
Initial	2733.14±5.87	4.29±0.16	45.34±1.11	22.73±1.28	0.62±0.02
Final	2908.40±1.76	2.00±0.12	56.57±1.12	52.33±1.05	1.11±0.04
Difference	175.26±5.73	-2.29±0.13	11.23±1.15	29.60±1.30	0.49±0.06
t' value	203.68**	65.88**	36.52**	85.16**	91.63**
Group TD					
Initial	2795.35±4.88	4.31±0.13	45.53 ±0.16	22.47±1.19	0.53±0.01
Final	2924.58±5.42	3.87±0.12	43.37 ±0.18	27.73±1.12	0.56±0.01
Difference	129.23±1.95	-0.44±0.05	-2.16 ±0.23	5.26±1.02	0.03±0.02
t' value	256.47**	29.92**	33.33**	19.29	5.61
F' value between groups	5327.79**	422.86**	1628.99**	812.37**	8.21**

** significant at one per cent level

TUKEY VALUES FOR SERUM SUPEROXIDE DISMUTASE

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		129.23	**143.26**	**150.23**	**175.26**
TD	**129.23**	-	14.03	21	46.03
TB	**143.26**		-	6.97	32
TA	**150.23**			-	25.03
TC	**175.26**				-

alpha =0.5 ;D_{min}=Q^*_t(sqrt($MS_{within\ group}$))/(sqrt(S))=10.23

TUKEY VALUES FOR SERUM MALONDIALDEHYDE

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		0.44	**1.97**	**2.25**	**2.29**
TD	**0.44**	-	1.53	1.81	1.85
TB	**1.97**		-	0.28	0.32
TA	**2.25**			-	0.04
TC	**2.29**				-

alpha =0.5D_{min}=Q^*_t(sqrt($MS_{within\ group}$))/(sqrt(S))=1.09

TUKEY VALUES FOR SERUM GLUTATHIONE REDUCED

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		2.16	**5.23**	**9.25**	**11.23**
TD	**2.16**	-	3.07	7.09	9.07
TB	**5.23**		-	4.02	6.00
TA	**9.25**			-	1.98
TC	**11.23**				-

alpha =0.5D_{min}=Q^*_t(sqrt($MS_{within\ group}$))/(sqrt(S))=8.26* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970

TUKEY VALUES FOR SERUM GLUTATHIONE PEROXIDASE

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		5.26	22.73	19.74	29.60
TD	5.26	-	17.47	14.48	24.34
TB	22.73		-	2.99	6.87
TA	19.74			-	9.86
TC	29.60				-

alpha =0.5D_{min}=Q^*_t(sqrt($MS_{within\ group}$))/(sqrt(S))=1.14

TUKEY VALUES FOR SERUM TOTAL ANTIOXIDANT ACTIVITY

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		0.03	0.35	0.45	0.49
TD	0.03	-	0.32	0.42	0.46
TB	0.35		-	0.10	0.14
TA	0.45			-	0.04
TC	0.49				-

alpha =0.5D_{min}=Q^*_t(sqrt($MS_{within\ group}$))/(sqrt(S))=0.27* Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970

Serum superoxide dismutase

The mean initial serum superoxide dismutase levels of the experimental and the control groups ranged from 2733.14 to 2795.35U/gHb whereas normal superoxide dismutase levels ranged from 3800 to 5500U/g Hb (Winterbourn *et al.*, 1975).There was an increase in the serum superoxide dismutase levels on supplementation. Maximum increase of 175.26 U/g Hb was noted in group TC supplemented with wheat germ and grass while an increase of 143.26 U/g Hb was registered in group TB supplemented with wheat grass. The final values of the serum superoxide dismutase were found to be significantly greater than the initial values in all the groups studied (P<0.01). One way analysis of variance conducted to substantiate the significance between the groups also proved to be significant at one per cent level. Results of tukey test indicated that the mean differences of serum superoxide dismutase of the supplemented groups TB,TA and TC were significantly greater than the group TD which served as the control. Further it could be noted that the mean incremental differences observed in group TC supplemented with wheat germ and grass was significantly greater over group TB which was supplemented with wheat grass proving that potential of combined supplementation of wheat germ and grass.

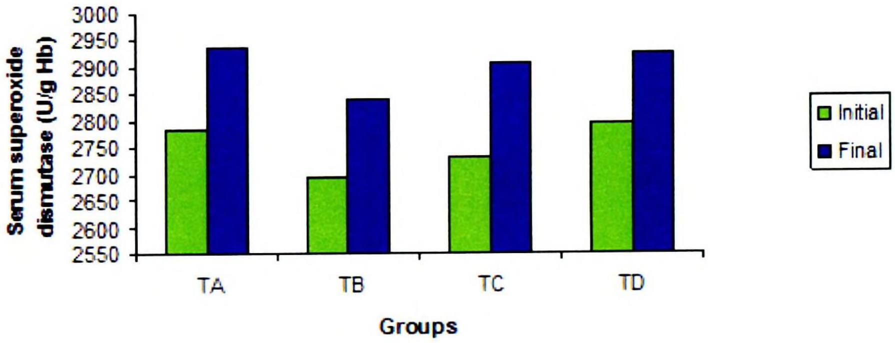

Figure 34
Changes in serum superoxide dismutase levels

Supplementation of wheat germ and grass resulted in significantly greater increments of serum superoxide dismutase over the supplementation of wheat germ and grass individually. Superoxide dismutase is one of the most important antioxidant enzymes. It catalyses the dismutation of superoxide radicals to hydrogen peroxide and molecular oxygen. It acts with a rate of 10000 times faster than the spontaneous rate of superoxide radical dismutation (Droge,2002). The decreased level of serum superoxide dismutase initially could be due to its direct inactivation by its products H_2O_2 or O.- 2 anion itself and / or development of auto antibodies against serum superoxide dismutase (Kurian and Scofield.,2003). A study by Boros *et al*.,(2001) proved that wheat germ extract decreases glucose uptake and RNA ribose formation and increased superoxide dismutase level in pancreatic adenocarcinoma cells.

Serum malondialdehyde

Yet another comprehensible utility of the supplementation was the demonstrably distinct decrease of mean serum malondialdehyde levels. The initial mean serum malondialdehyde levels in both the experimental and control groups ranged from 4.26 to 4.31 μM/L which was much higher than the normal range of 0.15 to 0.69 μM/L (Varley, 1986). The impact evaluation showed that the serum malondialdehyde had lowered by the individual and combined supplementation of wheat germ, bran and grass over a period of six months. Group TA supplemented with wheat germ had a reduction of 2.25 μM/L while group TB supplemented with wheat grass had a reduction of 1.97 μM/L and group TC supplemented with wheat germ and wheat grass had a decrease of 2.29 μM/L. The reduction in the mean serum malondialdehyde levels in the all the supplemented and control groups were significant at one per cent level. It could be further observed through tukey test that in all the supplemented groups the mean serum malondialdehyde reduced significantly compared to the control group. Further it could be noted that group TC supplemented with wheat germ and grass had a significantly greater reduction of serum malondialdehyde over groups TB and TA supplemented with wheat germ and grass individually.

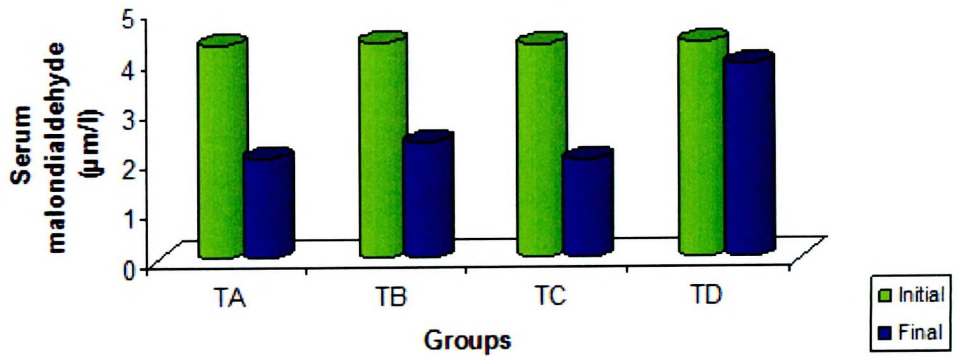

Figure 35
Changes in mean malonadialdehyde levels

It is clearly evident that the group TC supplemented with wheat germ and grass had a maximum reduction. The observation of the present investigation is in par with a study conducted Oscar and Rocio (2005) who explained that alpha lipoic acid plus vitamin E from wheat germ helps to reduce oxidative damage by reducing the levels of lipid peroxidation and malondialdehyde concentration in ischaemic and other oxidation-related pathological conditions.

Serum glutathione reduced

The mean initial glutathione reduced levels of the experimental and the control groups ranged from 44.23 to 45.34µM/L whereas normal serum glutathione reduced ranges from 50 to 80 µM/L (Beutler *et al.*,1963).There was an increase in the glutathione reduced levels on supplementation. Maximum increase of 11.23 µM/L was observed in group TC supplemented with wheat germ and grass while the increase of 5.23 µM/L was noted in group TB supplemented with wheat grass. Group TA supplemented with wheat germ had registered an increase of 9.25 µM/L.The final values of the glutathione reduced were found to be significantly greater (P<0.01) than the initial values in the experimental and control groups.

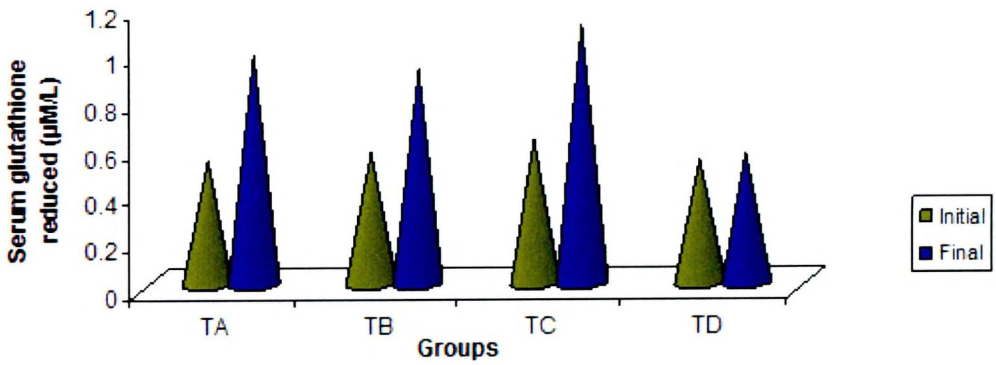

Figure 36
Changes in serum glutatihione reduced

One way analysis of variance conducted to substantiate the significance between the groups also proved to be significant at one per cent level. On applying tukey test to undercover the groups exhibiting maximum significance, it was noted that the mean increments of glutathione reduced of the supplemented groups TA,TB and TC were significantly greater than the control group TD.

It was noted from the tukey test that supplementation with wheat germ and grass resulted in significantly greater increments in glutathione reduced over the supplementation with wheat germ or grass alone. Mascio *et al.*,(1999) explain that wheat germ supplementation increases the level of reduced glutathione, the most important antioxidant metabolite that plays an important role in maintaining good levels of glutathione peroxidase activity which is the main enzyme involved in removing the H_2O_2 generated from dismutation of superoxide anions by superoxide dismutase. Glutathione peroxidase is also the co-factor of several reducing enzymes such as dehydroascorbate reductase and endoperoxide isomerase. Lenzsi *et al*,(1994) proved that wheat grass also reduces the lipid peroxidation rate in cancer patients by acting as a good chain breaking antioxidant. It reacts with peroxy radicals formed in propagation phase of lipid peroxidation to form carbon centered radical, which reacts readily and reversibly with oxygen to form a new chain – carrying peroxyl radicals which are highly stable forms than reactive oxygen species (ROS).

Serum glutathione peroxidase

The mean initial serum glutathione peroxidase of the tuberculosis subjects in all the groups were in the range of 22.40 to 22.73 U/g Hb as against the normal value of 49 to 93 U/g Hb as quoted by Pagalia and Valentine,(1967).Six months of supplementation of wheat products including wheat germ and grass and their combination brought about a marked increase in the serum glutathione peroxidase values in all the supplemented groups. Group TA supplemented with wheat germ had an increase of 19.74 U/g Hb in the serum glutathione peroxidase levels while group TB supplemented with wheat grass had an increase of 22.73U/gHb. Group

TC supplemented with wheat germ and grass had a maximum increase of 29.60 U/ gHb among the supplemented groups. Group TD which served as the control group had an increase of 5.26 U/g Hb over a period of six months. The final values of the serum glutathione peroxidase was significantly higher than the initial values (P<0.01) in all the experimental and the control group.

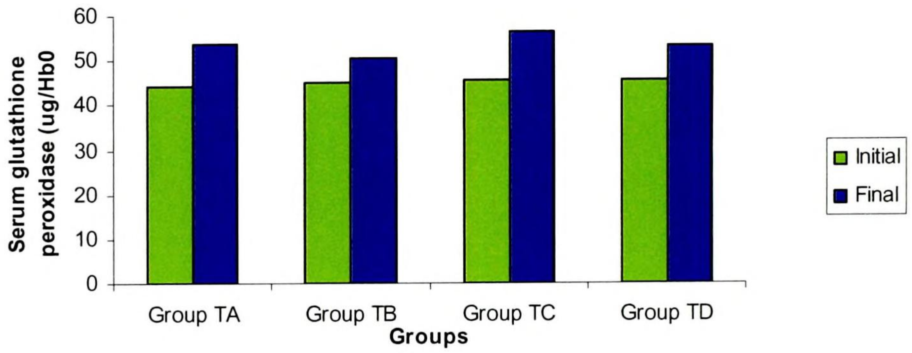

Figure 37
CHANGES IN SERUM GLUTATHIONE PEROXIDASE (U/g Hb)

The one way analysis of variance showed a statistically significant difference in the reduction of serum glutathione peroxidase levels among the four groups at one per cent level. On applying tukey test to undercover the groups exhibiting the significance it was observed that the mean incremental difference of serum glutathione peroxidase in the groups TA,TB and TC registered significance when compared with group TD (control group) proving that the groups supplemented individually and in combination of wheat germ and grass showed improvements over the non supplemented group TD. Group TC supplemented with wheat germ and grass had shown a statistically significant increase in the mean serum glutathione peroxidase when compared with the groups TB and TC which were supplemented with wheat grass and wheat germ respectively. The results are in par with the study of Osunda Hunsai (2006) who has isolated the enzyme glutathione peroxidase which oxidizes glutathione by triphosphopyridine nucleotide in aqueous extract of wheat germ.

Serum total antioxidant activity

The mean initial serum total antioxidant activity of the tuberculosis subjects in all the groups were in the range of 0.53 to 0.62mmol/l as against the normal range of 1.25 to 1.70 mmol/l as quoted by Miller *et al.*, (1993). Six months of supplementation of wheat products including wheat germ, grass and their combination brought about a marked increase in the total antioxidant values in all the supplemented groups. Group TA supplemented with wheat germ had an increase of 0.45mmol/l in the serum total antioxidant levels followed by group TB supplemented with wheat grass registering an increase of 0.35mmol/l. Group TC supplemented with wheat germ and grass registered an increase of 0.49mmol/l. The control group TD had an increase of 0.03mmol/l over a period of six months without any supplementation. The final values of the serum total antioxidant activity was significantly lower than the initial values ($P<0.01$) in all the experimental and control groups.

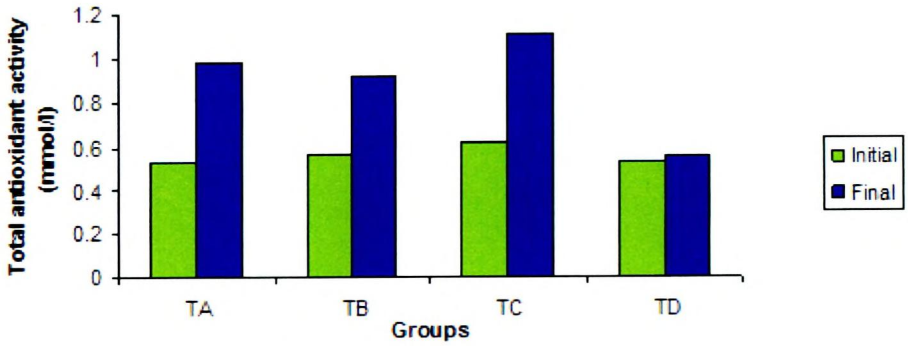

Figure 38
Changes in serum total antioxidant activity levels

The one way analysis of variance showed a statistically significant increase in the reduction of serum total antioxidant levels among the four groups at one per cent level. The mean incremental difference of serum total antioxidant activity in the groups TB,TA and TC registered significance over the group TD (control group).Similar findings were reported by Osunda Hunsai (2006) who demonstrated an increase of total antioxidant activity of pulmonary tuberculosis patients in Nigeria when a multivitamin mix contained wheat germ was supplemented.

(iv) Changes in serum zinc, selenium and copper levels

The changes in the serum zinc, selenium and copper levels of tuberculosis subjects after supplementation is given in Table XXIX and Figures 39 to 41.

TABLE XXIX
CHANGES IN MEAN SERUM ZINC, SELENIUM AND COPPER LEVELS
(N=15/group)

Groups	Serum zinc (µg/l)	Serum selenium (µg/l)	Serum copper (µg/l)
Group TA Initial	524.26±2.42	115.25±1.06	949.23±1.17
Final	596.53±2.10	127.09±1.96	922.12±1.38
Difference	72.27±2.11	11.84±1.16	-27.11±2.02
't' value	128.10**	39.50**	50.19**
Group TB Initial	527.40±1.29	116.95±1.20	948.56±1.33
Final	575.73±2.40	124.20±1.12	925.35±1.45
Difference	48.33±2.22	7.25±1.13	-23.21±1.95
't' value	81.43**	24.06**	44.52**
Group TC Initial	527.80±3.35	117.88±1.20	949.12±1.52
Final	615.87±2.35	132.45±1.33	919.44±1.02
Difference	88.07±2.33	14.57±1.80	-29.68±1.56
't' value	141.37**	31.33**	71.16**
Group TD Initial	528.93±1.18	119.75±1.79	949.53±1.18
Final	530.93±1.20	120.23±1.70	946.23±1.44
Difference	2.00±1.13	0.48±0.71	-3.30±1.33
't' value	6.62**	2.88*	9.28**
'F' value between groups	370.19**	460.10**	712**

** significant at one per cent level; * significant at five per cent level ;[NS] not significant

TUKEY VALUES FOR SERUM ZINC

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		2.00	**48.33**	**72.27**	**88.07**
TD	**2.00**	-	46.33	70.27	86.07
TB	**48.33**		-	23.94	39.74
TA	**72.27**			-	15.80
TC	**88.07**				-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=45.30$

TUKEY VALUES FOR SERUM SELENIUM

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		0.48	**7.25**	**11.84**	**14.57**
TD	**0.48**	-	6.77	11.36	14.09
TB	**7.25**		-	4.59	7.32
TA	**11.84**			-	2.73
TC	**14.57**				-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=7.08$

TUKEY VALUES FOR SERUM COPPER

Groups	Means in ascending order	Group TD	Group TB	Group TA	Group TC
		3.30	**23.21**	**27.11**	**29.68**
TD	**3.30**	-	19.91	23.81	26.38
TB	**23.21**		-	3.90	6.47
TA	**27.11**			-	2.57
TC	**29.68**				-

alpha =0.5 ;$D_{min}=Q^*_t(sqrt(MS_{within\ group}))/(sqrt(S))=1.70$ * Table 29 in E.S. Pearson and H.O. Hartley (Eds.),*Biometrika tables for statisticians* (3rd ed., Vol. 1), Cambridge University Press, New York, 1970

Serum zinc

The mean initial serum zinc levels ranged from 524.26 to 528.93 μg /l in the experimental and control groups as against the normal range 565 to 580 μg /l as quoted by Klevay (1984). It is inferred that there was a gradual increase in serum zinc levels on supplementation with wheat germ and grass individually and in combination. Group TA supplemented with wheat germ had an increase of 72.27 μg /l, group TB supplemented with wheat grass had an increase of 48.33 μg /dl and group TC supplemented with wheat germ and grass had a maximal increase of 88.07 μg /l . The final values of the serum zinc was significantly lower than the initial values (P<0.01) in all the experimental and control groups.

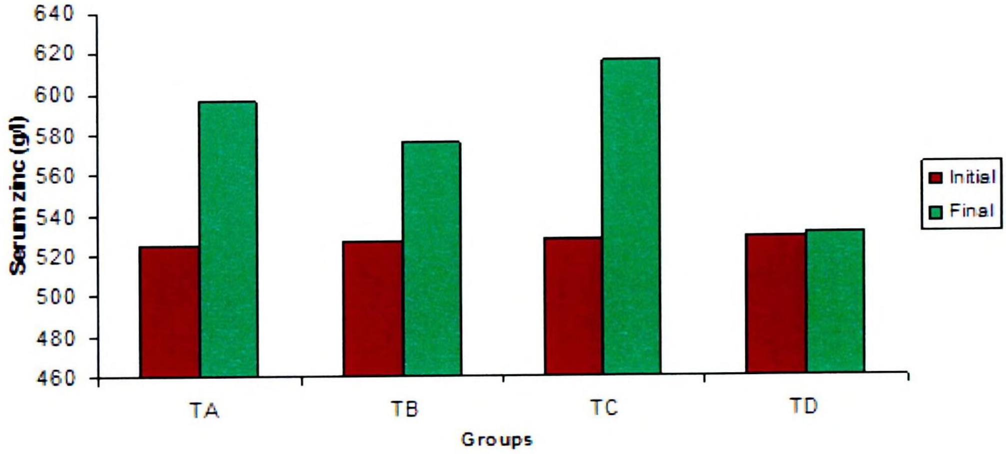

Figure39
Changes in serum zinc levels

The one way analysis of variance showed a statistically significant difference in the increase of serum zinc levels among the four groups at one per cent level. On applying tukey test to undercover the groups exhibiting the significance it was observed that the increment in the serum zinc levels in the groups TC,TB and TA supplemented with wheat products proved significance over the group TD which served as the control group.

Serum zinc levels increased in the tuberculosis subjects after the supplementation. The rapidly dividing cells of the immune system are sensitive to zinc deficiency. The role of zinc in the development and maintenance of a normally functioning immune system is well established (Weismann *et al.*,1990).

Serum selenium

The mean serum selenium levels ranged from 115.25-119.75 g /l in the experimental and control groups as against the normal range of 140-150 g /dl as quoted by Oster *et al.*,(1983). It is inferred that there was an increase in serum selenium levels on supplementation with wheat germ and grass individually and in combination. Group TA supplemented with wheat germ had an increase of 11.84 g /l, group TB supplemented with wheat grass had an increase of 7.25 g/l and group TC supplemented with wheat germ and wheat grass had a maximal increase of 14.57g /l. The final values of the serum selenium was significantly higher than the initial values (P<0.01) in all the experimental and control groups.

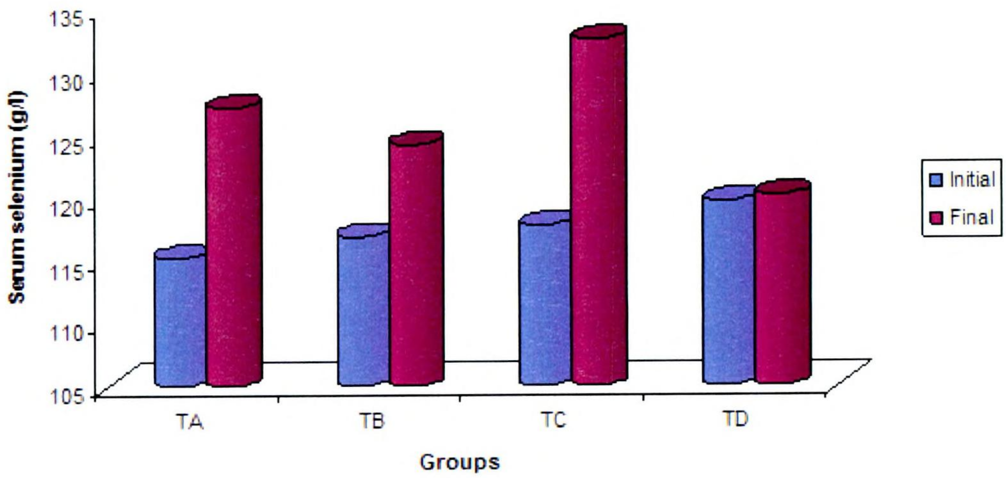

Figure 40
Changes in serum selenium levels

The one way analysis of variance showed a statistically significant difference in the increase of serum selenium levels among the four groups at one per cent level. On applying tukey test to undercover the groups exhibiting the significance it was observed that the increment in the serum selenium levels in the groups TB,TA and TC supplemented with wheat products proved significance over the group TD which served as the control group. Further the increments in groups TA and TC were significantly greater than the group TB supplemented with wheat grass.

Serum copper

The mean initial serum copper levels ranged from 948.56 to 949.53 g /l in the experimental and control groups as against the normal range of 950-1100g /l as quoted by Klevay (1987). There was a decrease in serum copper levels on supplementation with wheat germ and grass individually and in combination. Group TA supplemented with wheat germ had a decrease of 27.11 g /l, group TB supplemented with wheat grass had a reduction of 23.21 g /l and group TC supplemented with wheat germ and wheat grass had a maximal decrease of 29.68 g /l. The final values of the serum copper were significantly lower than the initial values (P<0.01) in all the experimental and control groups.

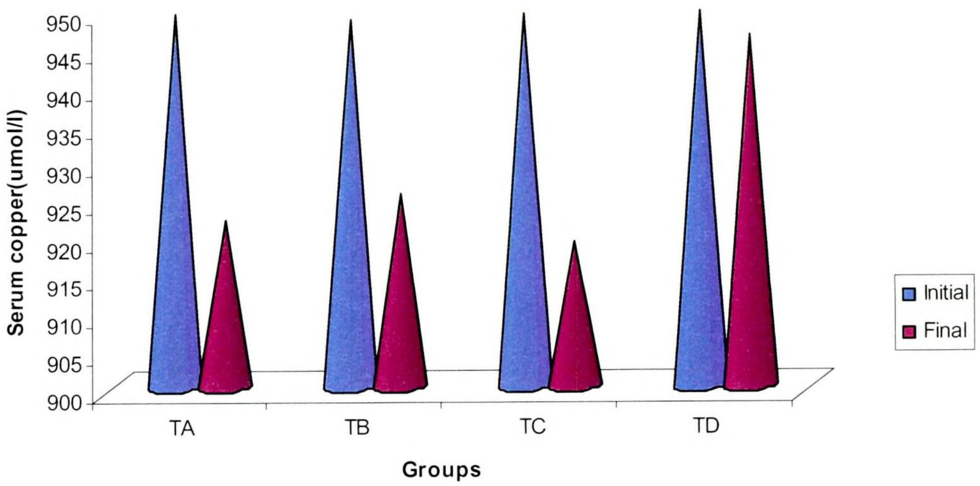

Figure 41
Changes in serum copper levels (g/l)

The one way analysis of variance showed a statistically significant difference in the decrease of serum copper levels among the four groups at one per cent level. On applying tukey test it was observed that the reduction in the serum copper levels in the groups TB,TA and TC supplemented with wheat products were significantly lower than the control group TD. Further the mean difference of the serum copper levels of groups TA and TC supplemented with wheat germ and germ and grass respectively proved significance over the group TB supplemented with wheat grass.

Serum copper level is elevated in chronic and acute illness . Increase in the level of copper may be due to reduction in the level of zinc (Prasad.,1985).With the decrease in copper levels it could be concluded from this study that wheat germ and grass can act as an adjuvant nutritional supplement in pulmonary tuberculosis .

(iv) Comparison of the impact of different supplements on the blood parameters assessed

Table XXX summarizes the impact of different supplements on the blood parameters assessed on the experimental groups.

TABLE XXX
COMPARISON OF THE EFFECT OF DIFFERENT SUPPLEMENTS ON BLOOD PARAMETERS

Parameter	Rank		
	1	2	3
Total protein (g/dl)	TC	TA	TB
Serum albumin (g/dl)	TC	TA	TB
TLC (cells/m^3)	TC	TA	TB
CD_4(cells/m^3)	TC	TA	TB
Superoxide dismutase (U/gHb)	TC	TA	TB
Malondialdehyde (μM/L)	TC	TA	TB
Glutathione reduced (μM/L)	TC	TA	TB
Glutathione peroxidase (U/g Hb)	TC	TA	TB
Serum zinc (μg/L)	TC	TA	TB
Serum selenium(μg/L)	TC	TA	TB
Serum copper(μg/L)	TC	TA	TB

It could be clearly depicted from the Table that group TC supplemented with wheat germ and grass proved its efficiency in maintaining the blood parameters which were altered in tuberculosis. Further group TA supplemented with wheat germ ranked second following by group TB reducing the blood parameters.

(iv) Changes in total protein and CD_4 levels after the withdrawal of supplementation

Table XXXI gives the final and after three months of withdrawal of supplementation data of serum total protein, albumin, TLC and CD4 count.

TABLE XXXI
CHANGES IN BLOOD PARAMETERS ATER WITHDRAWAL OF SUPPLEMENTATION

(N=5)

Groups	**Total protein g/dl**	**CD_4 (cells/m^3)**
Group TA Final	8.8±1.46	501.73±0.96
A3M	8.2±2.31	475.56±1.45
Difference	0.6±0.20	26.17±2.45
't' value	6.72**	39.95**
Group TB Final	8.4±5.36	477.40±5.32
A3M	8.1±4.12	398.75±2.14
Difference	0.3±0.31	78.65±3.89
t' value	2.96**	75.62**
Group TC Final	9.4±1.12	573.93±0.25
A3M	8.3±1.01	499.88±2.65
Difference	1.1±0.2	74.05±3.85
t' value	12.32**	71.93**

A3M: After three months of withdrawal of supplementation ;** significant at one per cent level

Serum total protein levels

It is inferred from the Table that there was a decrease in the total protein levels after the withdrawal of supplementation. Group TA showed a decrease of 0.6g/dl. Group TB had a decrease of serum total protein of 0.3g/dl while group TC had a decrease of 1.1g/dl.

Serum CD_4 levels

The serum CD_4 levels decreased after withdrawal of supplementation of individual as well as combined supplementation of wheat germ, bran and grass. Group TA showed a decrease of 26.17 cells/m^3. Group TB had a decrease of 78.65 cells/m^3 while group TC had a decrease of 74.05 cells/m^3. The after three months of withdrawal of supplementation values of the serum CD_4 levels was significantly higher than the final values ($P<0.01$) in all the experimental groups.

Thus, the findings of the study revealed that individual and combination of wheat germ and grass supplemented to tuberculosis subjects for a period of six months increased total protein, albumin and CD_4 levels and decreaseS the TLC levels. It is highly encouraging to note that rather than individual supplementation of wheat germ and grass combination of supplements including wheat germ and grass is more effective.