THE EFFECT OF PHYSICAL FITNESS EXERCISE ON THE DISTRIBUTION OF PLASMA HIGH DENSITY LIPOPROTEIN CHOLESTEROL AND OTHER LIPID FRACTIONS

BY

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ABSTRACT

The effect of participation in a physical fitness programme of moderate intensity on plasma lipoprotein cholesterols, total cholesterol, triglycerides and apolipoprotein-A levels was investigated.

Forty-nine inactive individuals participated in a sixteen week exercise programme that consisted of three sessions weekly with one and one-half hours in each session. The results were compared with those of a non-participating group.

Results suggest that a programme of physical exercise of moderate intensity of a relatively short duration (sixteen weeks) can have a beneficial effect by modifying the plasma lipid constituents in potentially high risk individuals, particularly those over 40 years who have low HDL-cholesterol blood levels.

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INTRODUCTION

More than a century ago, plasma cholesterol was implicated as the major component of atherosclerotic plaques.[34] In 1951 Barr et al[4] reported that coronary heart disease patients frequently exhibited several abnormalities in the distribution of lipoproteins in plasma. These included a tendency towards a reduction in alpha lipoprotein (High Density Lipoprotein) and an absolute increase in beta lipoprotein (Low Density Lipoprotein). Gofman and co-workers[17] in 1956 concluded from their study of serum lipids that plasma lipoprotein and cholesterol levels could be used to predict the clinical appearance of atherosclerosis.

In those early days, the study of lipoproteins and lipids was very simple. Only alpha and beta lipoproteins and total cholesterol were known. Present knowledge of this lipid system is more extensive; lipid components are now classified as chylomicrons, Very Low Density Lipoprotein (VLDL), Low Density Lipoprotein (LDL), High Density Lipoprotein (HDL), and their associated apolipoproteins[39] classified as A through E. Additionally, there are triglycerides, total-cholesterol and HDL-cholesterol.

Recent studies of Gordon et al[18] and others[1, 14, 27, 30, 34] have confirmed Barr's report that there is a significant negative correlation between HDL-cholesterol level and the development of coronary artery disease. At the same time there exists a positive correlationship between the incidence of coronary heart disease and the levels of LDL-cholesterol and triglycerides.

The popularity of the clinical application of HDL-cholesterol measurement is further enhanced by reports from other researchers[10, 12, 38, 19, 21, 25, 26, 28, 36] who suggested that factors such as strenuous physical training, smoking, weight loss, dieting, etc., can influence the HDL-cholesterol concentration. Wood[44] and Haskell[20] both have demonstrated that athletes have higher HDL-cholesterol and lower triglycerides and total-cholesterol levels when compared with more sedentary controls. However, most of these reports have dealt with individuals involved in strenuous physical training programmes of long duration in preparation for athletic competitions. There is scanty information on the extent to which lipid levels in sedentary individuals can be modified through participation in physical fitness programmes of modest duration and intensity.

In this study we investigated the relationship between the physical fitness exercise activity of men and women who participated in the fitness programme at the local fitness centres and their plasma levels of HDL-cholesterol, triglycerides, total-cholesterol, LDL-cholesterol and apolipoprotein-A.

Material and Method

Subjects

Forty-nine individuals employed in sedentary occupations were recruited through two local fitness centres. There were twenty-one females and twenty-eight males ranging in ages from 26 to 51 years and identified as the exercise group. Ninety per cent of these individuals had not previously participated in any regular form of organized physical fitness activity. The study began in December, 1980 and finished in April, 1981 (sixteen weeks).

The forty-seven non-participating subjects identified as the control group were hospital staff members. Their working day included considerable walking and normally they participated regularly in organized activities such as softball tournaments, run-a-mile race, morning exercise, etc. There were thirty females and seventeen males between the ages of 24 and 54 years. Their plasma lipid levels were determined on two occasions, in December, 1980 and in April, 1981.

All subjects, both exercise and control groups, were requested not to change their lifestyle (i.e. alcohol consumption, smoking and dietary habits) throughout the four months of the programme.

All were apparently healthy; they were not on medication and had normal biochemical profiles consisting of blood glucose, urea nitrogen, bilirubin, alkaline phosphatases, uric acid and aspartate aminotransferase.

Physical Fitness Exercise Programme

There were three exercise sessions of one and one-half hours duration per week. The exercise programme consisted of a fifteen minute warm-up exercise of low intensity stretching and sit-ups, followed by thirty minutes of running and jogging on an indoor track. The subjects were advised to monitor their exercise heart rate with ten second pulse counts several times during the exercise. The physical activity was regulated to maintain a heart rate for a period of thirty minutes at 70-85% of the age-adjusted maximum heart rate as described by Astrand.[2] A log was kept by each participant for the number of kilometers jogged. This allowed a fairly accurate check of the exercise levels maintained between sessions. The exercise session was supplemented by skipping, push-ups, swimming and weight lifting. There was a final fifteen minutes cool-down period after the exercise. The average jogging distance for each participant was fifteen kilometers per week.

Collection of Samples

Overnight fasting venous blood samples were collected from subjects in 10 ml. Vacutainer tubes (B.D. & Co. Canada Ltd.) containing an anti-coagulant (Ethylenediamine tetra-acetic acid). These tubes were gently mixed several times by inversion and then the blood was separated immediately by centrifugation at 2000 RPM at 4^oC. The plasma fractions were stored at -10^oC until analysis.

Blood samples were collected at enrollment to set the baseline level. Additional samples were taken after the fourth, eighth, twelfth and sixteenth weeks of the programme.

Analytical Methods

Total cholesterol was determined using a VP Bichromatic Computerized Analyzer (Abbott Laboratories, CA.). The enzymatic cholesterol reagents were supplied by Worthington Diagnostics. The cholesterol ester in the sample was enzymatically hydrolyzed to free cholesterol and free fatty acids. These products were oxidized by cholesterol oxidase to cholesten-3-one with the simultaneous production of hydrogen peroxide which was then quantitated by coupling with a chromogen. The resultant compound was measured at 500/600 mu.

Triglycerides analysis was performed on the SMA 12/60 multi-channel autoanalyzer (Technicon, Tarrytown, N.Y.). The procedure employed an Ultra-Violet enzymatic[40] reaction as modified by Kessler and Lederer. The method involved the enzymatic hydrolysis of triglycerides by lipase to glycerol and was followed by a series of reactions that terminated in the NADH-NAD indicator system. This final reaction was measured spectrophotometrically at 340 mu. Our method used two independent but interrelated channels for blanking purpose.

High-Density-Lipoprotein-Cholesterol procedure[46] involved the separation of the High Density Lipoprotein fraction from the other lipoproteins (i.e. VLDL and LDL fractions) by the addition of manganese chloride and heparin to the sample. The resulting precipitate was removed by centrifugation. The supernatant fluid containing the HDL-cholesterol was assayed with the Worthington enzymatic reagents in the Abbott VP analyzer.

Low-Density-Lipoprotein-Cholesterol was estimated according to the method of Friedwald.[13]

$$\text{LDL-chol.} = \text{Total-chol.} - \text{HDL-chol.} - \frac{\text{Triglycerides}}{5}$$

Apolipoprotein-A concentration was measured by the Standard Radial Immunodiffusion[41] technique. The reagent (M-partigen Apolipoprotein plate) was supplied by Behring Hoechst Inc. In this procedure, specific antiserum to apolipoprotein-A was impregnated in agarose gel. Immunoprecipitation was produced by addition of the sample. Apolipoprotein-A concentration was determined by the use of reference standards similarly tested.

Statistical Method

The student's t-distribution test for two sample observation was used to assess the significance of the difference between parameter means within and between groups.

$$t = \frac{(\bar{x}_1 - \bar{x}_2) - (u_1 - u_2)}{\sqrt{\left(\frac{1}{n_1} + \frac{1}{n_2}\right) \frac{(n_1 - 1)s_1^2 + (n_2 - 1)s_2^2}{n_1 + n_2 - 2}}}$$

with $n_1 + n_2 - 2$ degrees of freedom

Where $\bar{x}$ = Sample mean

s = Sample standard deviation

u = Population mean

n = Sample size.

To analyze the bivariate date (Table 111), the linear regression model was used to test the mathematical relationship between the variables and their correlation coefficient (r) by

$$y = a + bx$$

where $a = \bar{y} - b\bar{x}$

$$b = \frac{n\Sigma xy - \Sigma x\Sigma y}{n\Sigma x^2 - (\Sigma x)^2}$$

$$r = \frac{\Sigma xy - \frac{\Sigma x\Sigma y}{n}}{\sqrt{(\Sigma x^2 - \frac{(\Sigma x)^2}{n})(\Sigma y^2 - \frac{(\Sigma y)^2}{n})}}$$

Probability values were calculated from the t-Distribution table using the formula of

$$t = \sqrt{vr^2 / (1 - r^2)}$$

where $v = n - 2$

n = number of sample data points

r = correlation coefficient.

All statistical methods were performed with Texas Instruments Programmable 59 Calculator with PC - 100c Printer.

Due to the relatively small sample size and the nature of the biological parameter, a probability value of less than 0.075 was considered as statistically significant.

RESULTS

Changes in plasma lipid during the course of the 16-week exercise programme

Sample data of the experiment are shown in appendix (I).

Selection of control data for reduction of sample size is discussed in appendix (II).

The effect of exercise on various plasma lipid levels is shown in Table 1 and Figures I - V.

HDL-cholesterol It was quite apparent that there was a significant difference in HDL-cholesterol levels between female and male participants at most stages of our study. The HDL-cholesterol concentration increased numerically during the course of the exercise programme in both male and female groups. Within twelve weeks, the male exercise group had a significant increase which continued until termination of the programme. (43.6 ± 11.4 vs 48.9 ± 10.2 mg/dl, P = 0.03). The HDL-cholesterol in the female exercise group was significantly higher than the baseline level only after sixteen weeks of participation (50.3 ± 9.7 vs 54.9 ± 9.5 mg/dl, P = 0.07).

Total-Cholesterol There was no significant difference in total-cholesterol levels between the male and female exercise groups at any stage. Moreover, data did not show any significant changes throughout the sixteen-week programme. At the final week, the mean concentration of total-cholesterol was slightly lower, but this was not statistically significant.

TABLE I

CHANGES IN PLASMA LIPID LEVELS DURING A SIXTEEN WEEK PHYSICAL FITNESS PROGRAMME

	N	Sex	Baseline Mean mg/dl ± S.D.	4th Week Mean mg/dl ± S.D.	8th Week Mean md/dl ± S.D.	12th Week Mean mg/dl ± S.D.	16th Week Mean md/dl ± S.D.
HDL Cholesterol	21	Female	50.3 ± 9.7	51.2 ± 9.3	53.0 ± 10.0	52.5 ± 9.0	54.9* ± 9.5
	28	Male	43.6† ± 11.4	47.1 ± 10.6	45.6† ± 9.6	48.9* ± 10.2	48.0*† ± 10.5
Total Cholesterol	21	Female	233.3 ± 41.4	230.1 ± 43.4	228.0 ± 44.9	227.6 ± 44.6	221.0 ± 40.4
	28	Male	227.9 ± 47.4	226.2 ± 38.6	223.5 ± 42.8	223.0 ± 38.8	215.8 ± 37.4
Tri-glycerides	21	Female	142.3 ± 46.6	127.3 ± 45.9	128.0 ± 44.9	121.7 ± 42.4	122.9 ± 40.0
	28	Male	145.6 ± 37.8	132.3 ± 38.5	126.1* ± 35.2	124.9* ± 34.8	123.8* ± 38.4
LDL Cholesterol	21	Female	154.5 ± 37.9	153.4 ± 38.9	149.4 ± 40.9	150.9 ± 41.4	142.2 ± 37.2
	28	Male	155.2 ± 49.0	152.6 ± 40.3	152.8 ± 41.9	149.2 ± 39.3	143.0 ± 36.8
Apolipo-protein	21	Female	186.8 ± 13.0	186.2 ± 15.0	189.0 ± 14.2	188.7 ± 11.9	190.4 ± 12.3
	28	Male	185.1 ± 12.1	184.7 ± 12.6	187.8 ± 11.6	188.8 ± 9.4	189.6 ± 10.7

* Indicate $P < 0.075$ between intervals within gender groups

† Indicate $P < 0.075$ between gender groups within intervals

FIG. 1 MEAN HDL-CHOLESTEROL LEVEL ATTAINED AT INTERVALS OF THE EXERCISE PROGRAMME

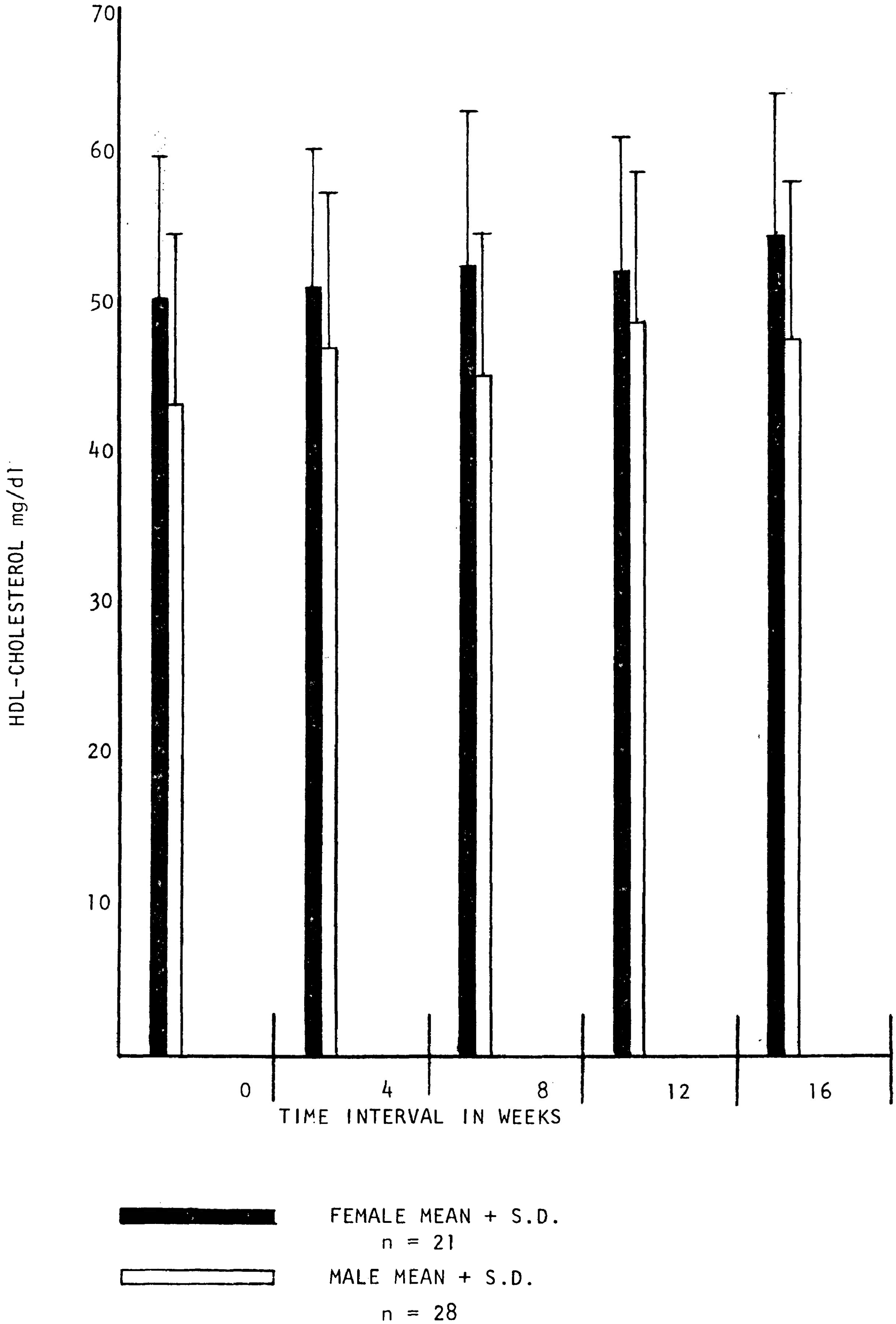


FIG.11 TOTAL CHOLESTEROL LEVEL ATTAINED AT INTERVALS OF THE EXERCISE PROGRAMME

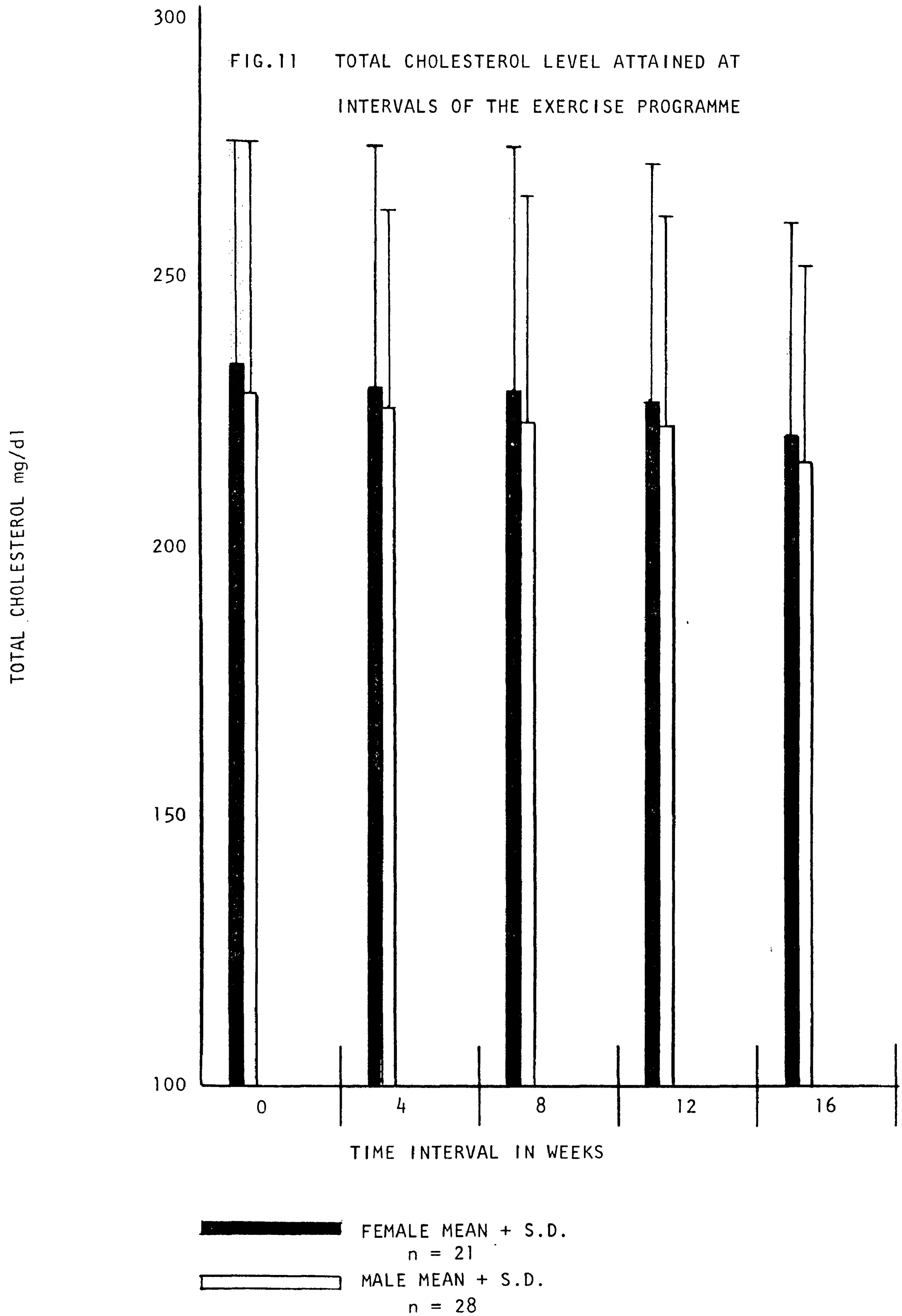


FIG. 111 TRIGLYCERIDES LEVEL ATTAINED AT INTERVALS OF THE EXERCISE PROGRAMME

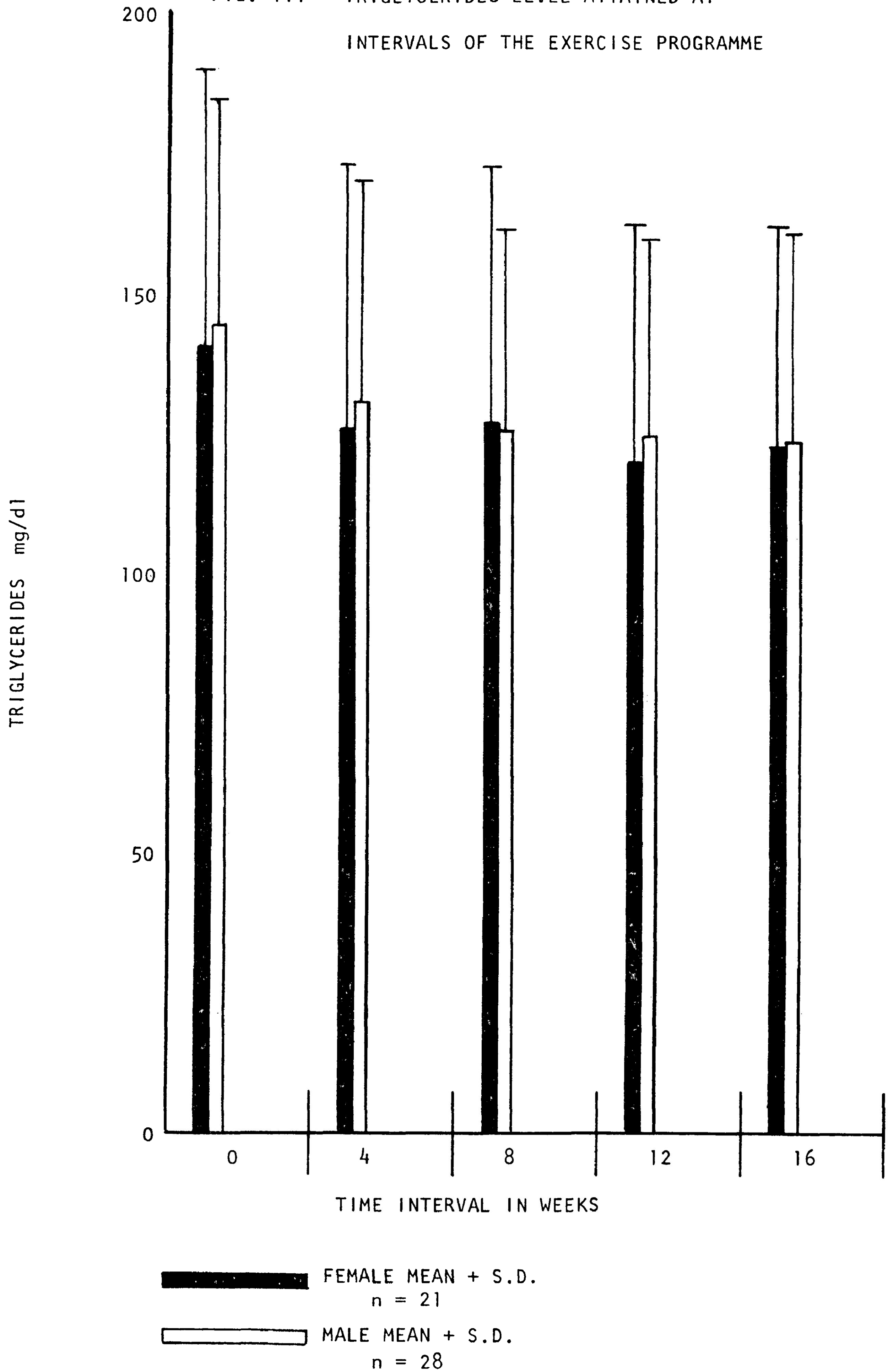


FIG. IV LDL-CHOLESTEROL LEVELS ATTAINED AT INTERVALS OF THE EXERCISE PROGRAMME

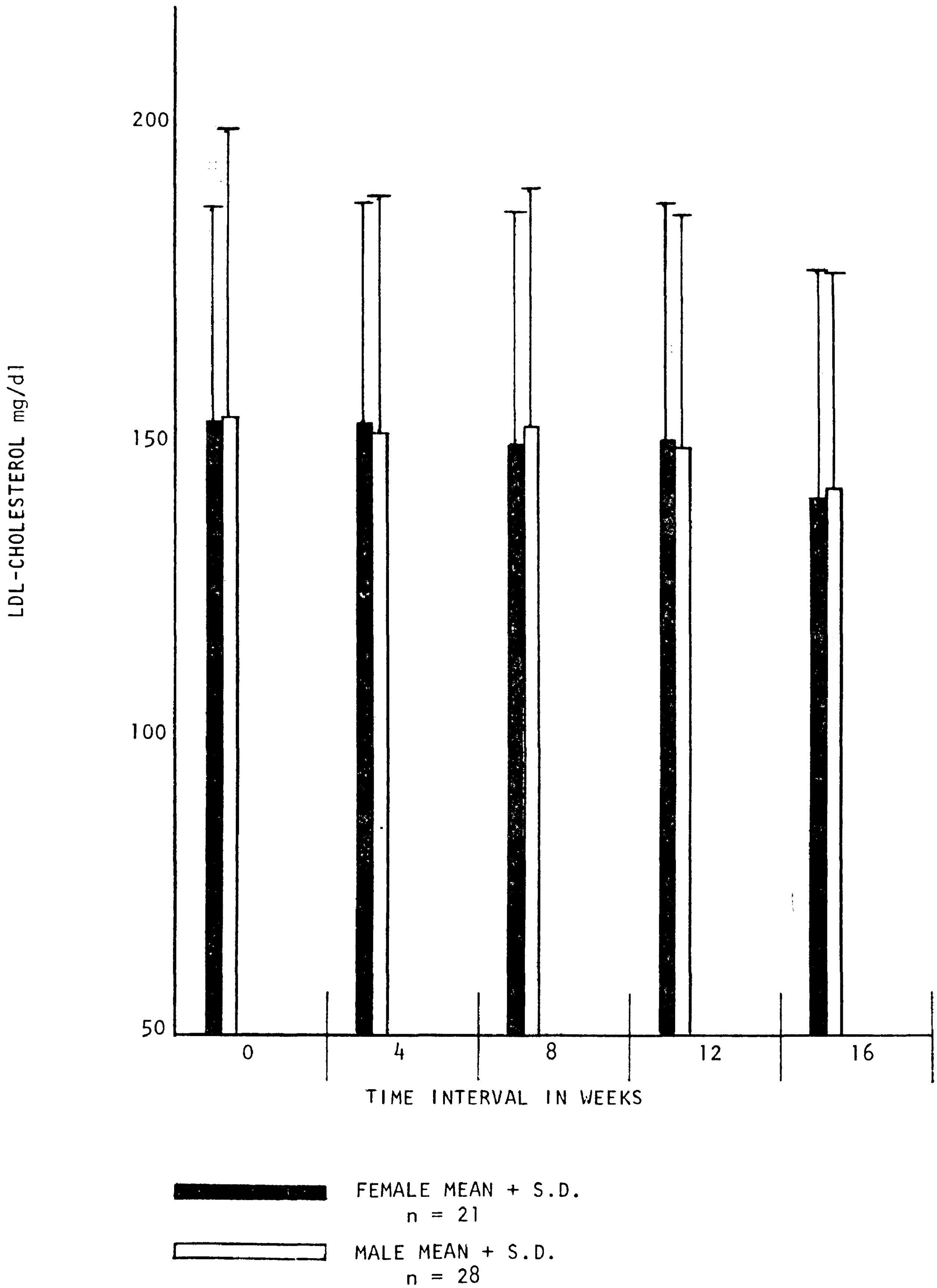
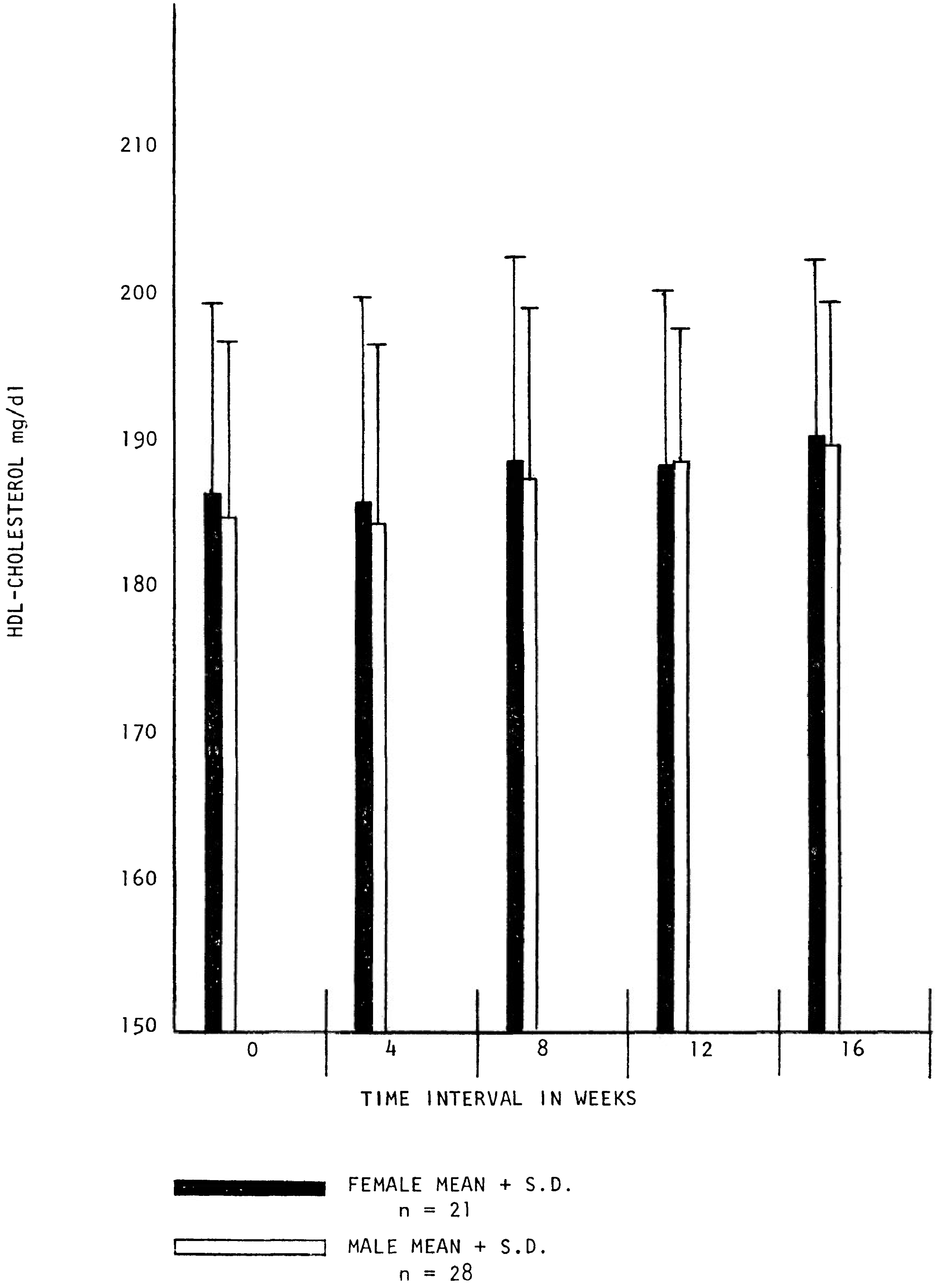


FIG. V APOLIPOPROTEIN-A LEVELS ATTAINED AT INTERVALS OF THE EXERCISE PROGRAMME



(Male = 227.9 ± 47.4 vs 215.8 ± 37.4.
Female = 233.3 ± 41.4 vs 221.0 ± 40.4 mg/dl).

<u>Triglycerides</u> The triglycerides level was significantly lower after eight weeks of exercise in the male group when compared with the baseline value (145.6 ± 37.8 vs 126.1 ± 35.2 mg/dl, P = 0.02). The female mean level gradually decreased as the exercise programme progressed. However, this change was too small to be statistically significant. No significant difference was observed between gender groups within intervals.

<u>LDL-Cholesterol</u> The LDL-cholesterol levels in both men and women were relatively constant during the course of the exercise programme. There was no significant change even at the sixteenth week between or within the gender groups. (Male = 155.2 ± 49.0 vs 143.0 ± 36.8. Female = 154.5 ± 37.9 vs 142.2 ± 37.2 mg/dl).

<u>Apolipoprotein-A</u> The mean level gradually increased as the exercise programme progressed. However, these changes were too small to be statistically significant even at the final week. (Male = 185.1 ± 12.1 vs 189.6 ± 10.7. Female = 186.8 ± 13.0 vs 190.4 ± 12.3 mg/dl). There was no significant difference between the male and female groups throughout the exercise programme and there was no significant change within the gender groups.

Comparison of plasma parameters of exercise group with non-participating group at baseline and at the sixteenth week

As shown in Table II, at the baseline there was a noticeable difference in HDL-Cholesterol levels between the exercise group and the non-participating group (Male = 43.6 $\pm$ 11.1 vs 49.2 $\pm$ 5.5, P = 0.03, Female = 50.3 $\pm$ 9.7 vs 55.4 $\pm$ 7.2 mg/dl. P = 0.02). At the end of the exercise programme, this difference had disappeared (Male = 48.0 $\pm$ 10.3 vs 50.4 $\pm$ 5.6, Female = 54.9 $\pm$ 9.5 vs 56.0 $\pm$ 8.0 mg/dl).

Initially, there was a statistically significant difference in total-cholesterol levels between the female exercise group and non-participating group (233.3 $\pm$ 41.4 vs 206.8 $\pm$ 42.4 mg/dl, P = 0.02). However, there was no significant difference between the male groups.

The triglycerides levels for either sex show no significant difference at initial and final stages.

The initial LDL-cholesterol level was significantly higher in the female exercise group, but not in the final stage. There was no significant difference in either initial or final LDL-cholesterol between the exercise and non-participating male groups.

No significant differences were observed between the groups in either initial or final apolipoprotein-A levels.

TABLE II

COMPARISON OF THE BASELINE AND FINAL PLASMA LIPID LEVELS BETWEEN THE EXERCISE GROUP AND NON-PARTICIPATING GROUP*

		Baseline			Final		
		Exercise Group	Non-Participating Group	Difference	Exercise Group	Non-Participating Group	Difference
HDL Cholesterol mg/dl	Female	50.3 ± 9.7	55.4 ± 7.2	Significant P = 0.02	54.9 ± 9.5	56.0 ± 8.0	Not significant P = 0.33
	Male	43.6 ± 11.4	49.2 ± 5.5	Significant P = 0.03	48.0 ± 10.3	50.4 ± 5.6	Not significant P = 0.20
Total-Cholesterol mg/dl	Female	233.3 ± 41.4	206.8 ± 42.9	Significant P = 0.02	221.0 ± 40.4	211.4 ± 39.7	Not significant P = 0.20
	Male	227.9 ± 47.4	215.6 ± 30.9	Not significant P = 0.18	215.8 ± 37.4	213.6 ± 27.1	Not significant P = 0.42
Tri-glycerides mg/dl	Female	142.3 ± 46.6	126.6 ± 37.1	Not significant P = 0.09	122.9 ± 40.0	127.9 ± 31.6	Not significant P = 0.31
	Male	145.6 ± 37.8	132.8 ± 33.9	Not significant P = 0.14	123.8 ± 38.4	131.2 ± 29.3	Not significant P = 0.25
LDL Cholesterol mg/dl	Female	154.5 ± 37.9	126.6 ± 39.0	Significant P = 0.01	142.2 ± 37.2	131.1 ± 38.2	Not significant P = 0.15
	Male	155.2 ± 49.0	139.8 ± 30.4	Not significant P = 0.15	143.0 ± 36.8	137.5 ± 27.6	Not significant P = 0.30
Apolipo-protein mg/dl	Female	186.8 ± 13.0	190.8 ± 16.0	Not significant P = 0.18	190.4 ± 12.3	190.0 ± 14.3	Not significant P = 0.46
	Male	185.1 ± 12.1	186.7 ± 10.7	Not significant P = 0.33	189.6 ± 10.7	187.9 ± 10.4	Not significant P = 0.30

* mean ± S.D.

P 0.075 is considered not significant

Correlation of HDL-cholesterol data with the total-cholesterol, triglycerides, LDL-cholesterol and Apolipoprotein-A

The correlation coefficients between the HDL-cholesterol values and other lipid variables are presented in Table III and Figure VI - IX.

A negative correlation was observed between the changes in HDL-cholesterol and the changes in triglycerides, total-cholesterol and LDL-cholesterol.

A positive correlation was noted between the changes in HDL-cholesterol and apolipoprotein-A.

Influence of age on the results of the plasma lipid parameters during the exercise programme

In order to demonstrate the influence of age in our study, we arbitrarily separated the individuals into two age groups i.e. < 40 years and ≥ 40 years.

Tables IV-A,B show the effect of age on the lipid parameters at the initial and final weeks of the study. Among the control males, there was no demonstrated age effect on any parameters at any stage of the study.

TABLE III: CORRELATION COEFFICIENT (r) OF HIGH DENSITY-LIPOPROTEIN-CHOLESTEROL DATA WITH OTHER LIPID VARIABLES

EXERCISE GROUP

Δ = difference between Sixteenth week and baseline

	r	p
Δ HDL-cholesterol vs Δ Total-cholesterol	-0.66	significant $p < 0.01$
Δ HDL-cholesterol vs Δ Triglycerides	-0.67	significant $p < 0.01$
Δ HDL-cholesterol vs Δ LDL-cholesterol	-0.70	significant $p < 0.01$
Δ HDL-cholesterol vs Δ Apolipoprotein-A	+0.39	significant $p < 0.01$

$p < 0.075$ is considered significant

FIGURE VI: CORRELATION BETWEEN THE CHANGES† IN HDL-CHOLESTEROL AND TOTAL-CHOLESTEROL

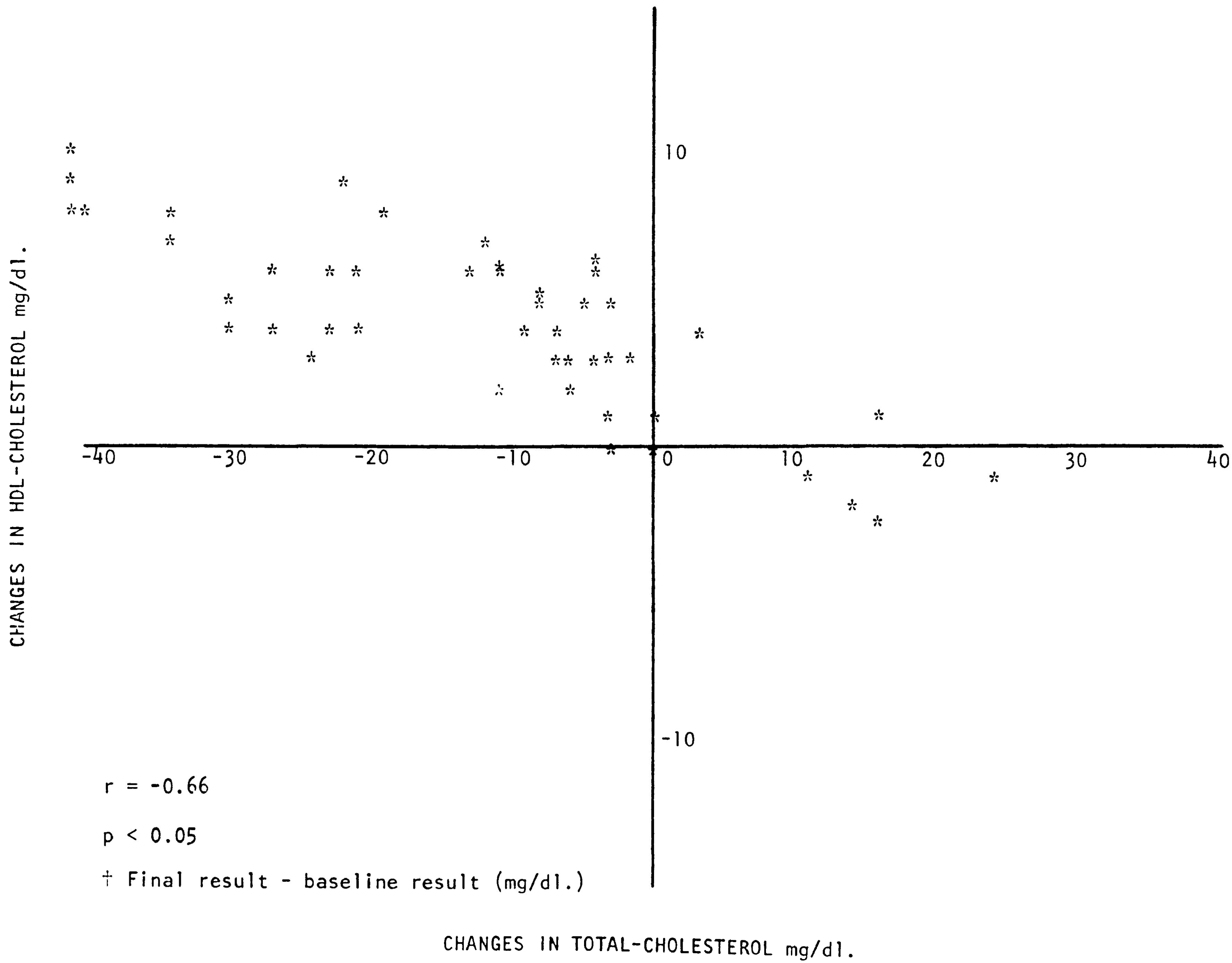
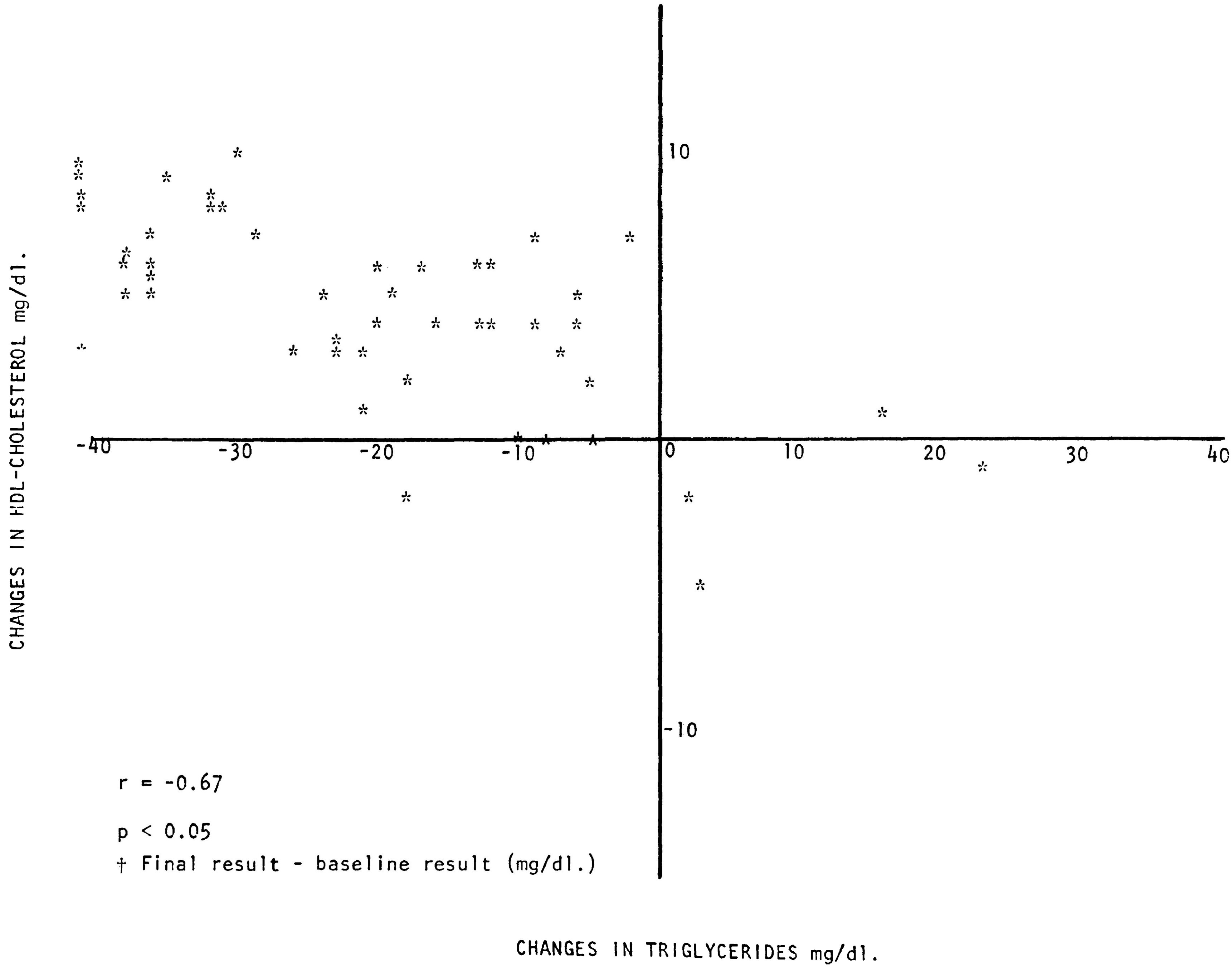


FIGURE VII: CORRELATION BETWEEN THE CHANGES† IN HDL-CHOLESTEROL AND TRIGLYCERIDES

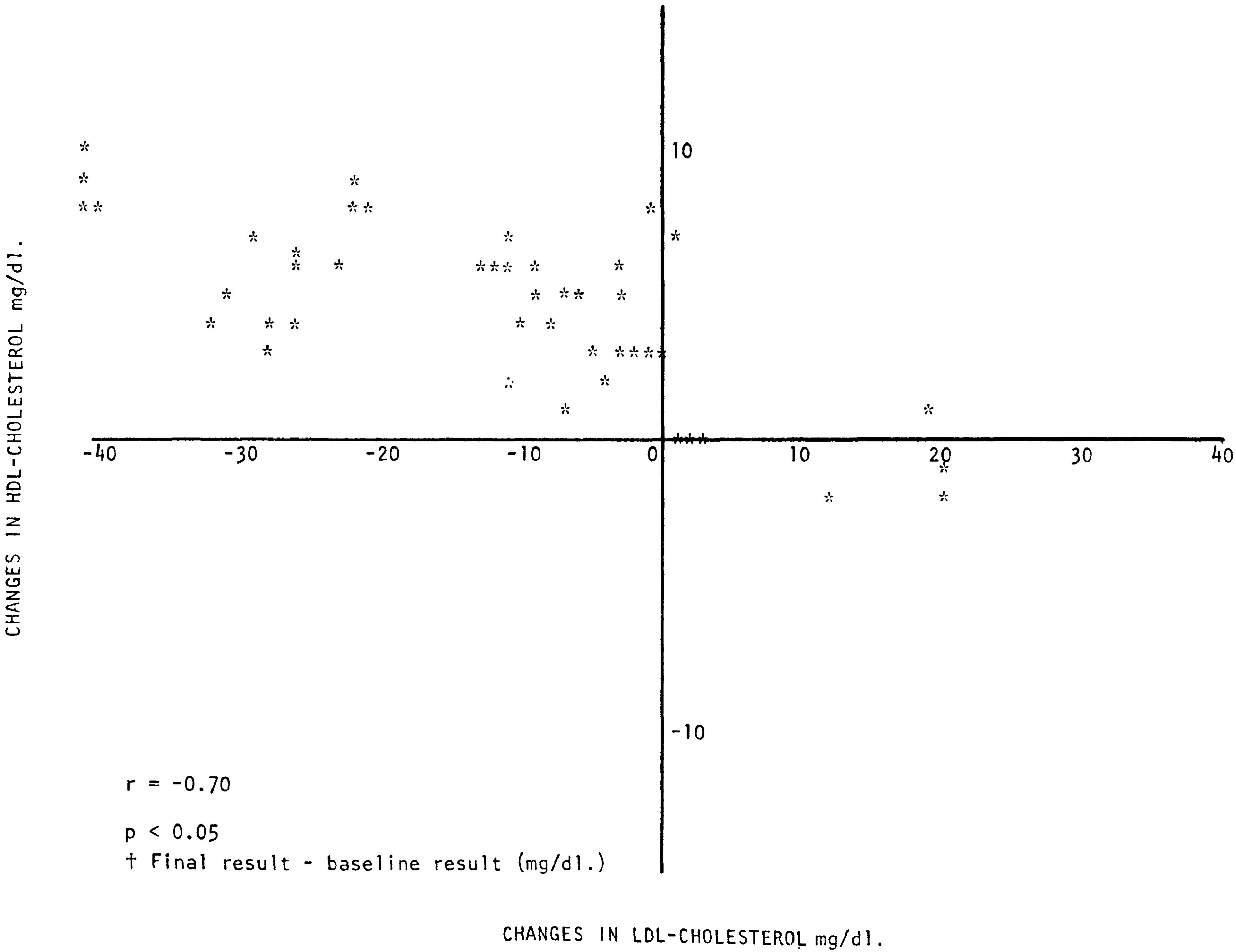


$r = -0.67$

$p < 0.05$

† Final result - baseline result (mg/dl.)

FIGURE VIII: CORRELATION BETWEEN THE CHANGES† IN HDL-CHOLESTEROL AND LDL-CHOLESTEROL

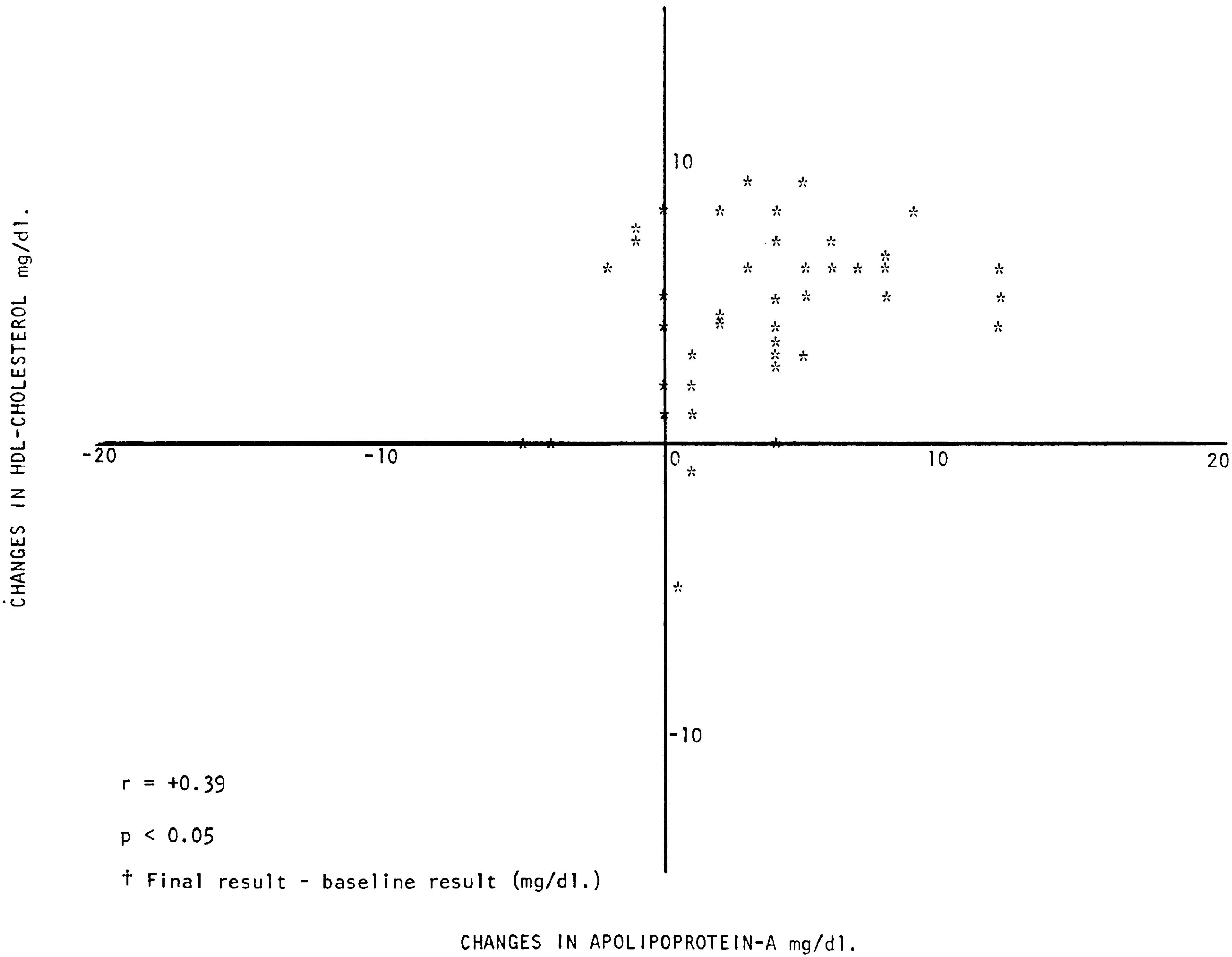


r = -0.70

$p < 0.05$

† Final result - baseline result (mg/dl.)

FIGURE IX: CORRELATION BETWEEN THE CHANGES† IN HDL-CHOLESTEROL AND APOLIPOPROTEIN-A



$r = +0.39$

$p < 0.05$

† Final result - baseline result (mg/dl.)

TABLE IV-A

COMPARISON OF PLASMA LIPID PARAMETERS† BETWEEN AGE GROUPS WITHIN THE MALE AND FEMALE NON-PARTICIPATING CONTROL GROUPS AT THE INITIAL AND FINAL STAGES OF THE STUDY PERIOD

		INITIAL			FINAL			
		< 40 years	≥ 40 years	Difference-t	< 40 years	≥ 40 years	Difference-t	v
HDL Cholesterol mg/dl	Males	49.1 ± 6.5	49.3 ± 4.6	Not significant 0.051	51.0 ± 5.2	49.6 ± 6.2	Not significant 0.496	15
	Females	57.6 ± 6.3	50.3 ± 5.8	Significant 2.604***	57.7 ± 7.1	50.1 ± 6.0	Significant 2.441***	17
Total Cholesterol mg/dl	Males	218.6 ± 30.1	212.3 ± 33.4	Not significant 0.337	214.3 ± 22.7	212.8 ± 32.9	Not significant 0.116	15
	Females	191.0 ± 45.4	223.5 ± 46.1	Significant 1.531*	204.0 ± 36.8	236.4 ± 42.0	Significant 1.785**	17
Tri-glycerides mg/dl	Males	138.3 ± 42.6	125.4 ± 20.3	Not significant 0.783	138.3 ± 35.4	123.3 ± 19.7	Not significant 1.064	15
	Females	112.9 ± 37.1	136.4 ± 28.1	Significant 1.914**	116.7 ± 31.1	135.6 ± 23.1	Not significant 1.448	17
LDL Cholesterol mg/dl	Males	141.8 ± 27.5	137.6 ± 35.2	Not significant 0.273	136.6 ± 20.8	138.5 ± 35.3	Not significant 0.149	15
	Females	110.9 ± 42.6	145.6 ± 40.3	Significant 1.712**	123.0 ± 36.6	159.8 ± 38.0	Significant 1.956**	17
Apolipo-protein mg/dl	Males	188.0 ± 13.0	185.3 ± 8.2	Not significant 0.315	190.3 ± 12.1	186.0 ± 7.9	Not significant 0.863	15
	Females	197.7 ± 15.9	184.0 ± 19.0	Significant 1.793**	196.1 ± 13.2	183.3 ± 15.4	Significant 2.127**	17

v total degrees of freedom

† mean ± S.D.

Significance *** less than 1.0%
** less than 5.0%
* less than 7.5%

TABLE IV-B

COMPARISON OF PLASMA LIPID PARAMETERS† BETWEEN AGE GROUPS WITHIN THE MALE AND FEMALE EXERCISE GROUPS AT THE INITIAL AND FINAL STAGES OF THE STUDY PERIOD

		INITIAL			FINAL			
		< 40 years	≥ 40 years	Difference-t	< 40 years	≥ 40 years	Difference-t	v
HDL Cholesterol mg/dl	Males	49.1 ± 13.5	39.9 ± 8.5	Significant 2.225**	51.3 ± 12.9	45.8 ± 8.0	Not significant 1.385	26
	Females	54.4 ± 8.8	46.7 ± 9.4	Significant 1.928**	59.5 ± 9.2	50.7 ± 8.1	Significant 2.325**	19
Total-Cholesterol mg/dl	Males	198.8 ± 36.5	246.7 ± 44.8	Significant 2.964***	195.1 ± 30.6	229.2 ± 36.0	Significant 2.591***	26
	Females	209.2 ± 33.0	255.2 ± 36.6	Significant 3.012***	198.2 ± 27.4	241.8 ± 39.9	Significant 2.891***	19
Tri-glycerides mg/dl	Males	135.5 ± 44.8	152.2 ± 32.3	Not significant 1.154	121.7 ± 46.7	125.2 ± 33.4	Not significant 0.228	26
	Females	127.1 ± 51.8	156.2 ± 38.5	Not significant 1.469	105.5 ± 46.9	138.8 ± 25.4	Significant 2.052**	19
LDL Cholesterol mg/dl	Males	122.4 ± 31.9	176.5 ± 48.8	Significant 3.350***	119.6 ± 22.6	158.2 ± 36.7	Significant 3.112***	26
	Females	129.5 ± 24.5	177.3 ± 33.7	Significant 3.678***	117.9 ± 15.6	164.3 ± 37.8	Significant 3.603***	19
Apolipo-protein mg/dl	Males	190.9 ± 16.7	181.4 ± 6.0	Significant 2.154**	191.7 ± 14.8	188.2 ± 7.1	Not significant 0.855	26
	Females	191.8 ± 12.8	182.2 ± 11.9	Significant 1.785**	194.8 ± 13.1	186.6 ± 10.8	Significant 1.586*	19

v total degrees of freedom

† mean ± S.D.

Significance *** less than 1.0%
** less than 5.0%
* less than 7.5%

Within the control females, there were significant age dependent differences in all parameters at both the initial and final stages of the study, except the final triglycerides level.

Among the exercise males, the younger age group exhibited higher HDL-cholesterol and apolipoprotein-A levels at the initial stage. By the end of the programme this age effect had disappeared. Total-cholesterol and LDL-cholesterol levels were lower at the start in the younger age group and remained lower at the final stage. There was no age-dependency for triglyceride levels at either initial or final periods of the study.

Among the exercise females, the under 40 years age group exhibited higher HDL-cholesterol and apolipoprotein-A levels at the initial stage and maintained this difference to the final week. They also had lower triglycerides, total-cholesterol and LDL-cholesterol levels than the over 40 years age group at both initial and final periods of the exercise programme.

Tables V-A,B show the influence of age on the changes of various parameters during the 16-week study.

Only the exercise males, 40 years and over, showed any significant change in plasma parameter levels between the initial and final weeks

TABLE V-A

COMPARISON OF PLASMA LIPID PARAMETERS† BETWEEN INITIAL AND FINAL WEEKS OF THE STUDY PERIOD WITHIN AGE GROUPS OF NON-PARTICIPATING CONTROL MALES AND FEMALES

		< 40 years				≥ 40 years			
		Initial	Final	Difference-t	v	Initial	Final	Difference-t	v
HDL Cholesterol mg/dl	Males	49.1 ± 6.5	51.0 ± 5.2	Not significant 0.681	16	49.3 ± 4.6	49.6 ± 6.2	Not significant 0.140	14
	Females	57.6 ± 6.3	57.7 ± 7.1	Not significant 0.039	20	50.3 ± 5.8	50.1 ± 6.0	Not significant 0.044	14
Total-Cholesterol mg/dl	Males	218.6 ± 30.1	214.3 ± 22.7	Not significant 0.330	16	212.3 ± 33.4	212.8 ± 32.9	Not significant 0.030	14
	Females	191.0 ± 45.4	204.0 ± 36.8	Not significant 0.738	20	223.5 ± 46.1	230.4 ± 42.0	Not significant 0.585	14
Tri-glycerides mg/dl	Males	138.3 ± 42.6	138.3 ± 35.4	Not significant 0.000	16	125.4 ± 20.3	123.3 ± 19.7	Not significant 0.213	14
	Females	112.9 ± 37.1	116.7 ± 31.1	Not significant 0.262	20	136.4 ± 28.0	135.6 ± 23.1	Not significant 0.067	14
LDL Cholesterol mg/dl	Males	141.8 ± 27.5	136.6 ± 20.8	Not significant 0.454	16	137.6 ± 35.2	138.6 ± 35.3	Not significant 0.049	14
	Females	110.9 ± 42.6	123.0 ± 36.6	Not significant 0.263	20	145.6 ± 40.3	159.8 ± 38.0	Not significant 0.087	14
Apolipo-protein mg/dl	Males	188.0 ± 13.0	190.3 ± 12.1	Not significant 0.395	16	185.3 ± 8.2	186.0 ± 7.9	Not significant 0.187	14
	Females	197.7 ± 15.9	196.1 ± 13.2	Not significant 0.713	20	184.0 ± 19.0	183.3 ± 15.4	Not significant 0.722	14

v total degrees of freedom

† mean ± S.D.

TABLE V-B

COMPARISON OF PLASMA LIPID PARAMETERS† BETWEEN INITIAL AND FINAL WEEKS OF THE STUDY PERIOD WITHIN AGE GROUPS OF EXERCISE MALES AND FEMALES

		< 40 years				≥ 40 years			
		Initial	Final	Difference-t	v	Initial	Final	Difference-t	v
HDL Cholesterol mg/dl	Males	49.1 ± 13.5	51.3 ± 12.9	Not significant 0.388	20	39.9 ± 8.5	45.8 ± 8.0	Significant 2.095**	32
	Females	54.4 ± 8.8	59.5 ± 9.2	Not significant 1.268	18	46.7 ± 9.4	50.7 ± 8.1	Not significant 1.071	20
Total Cholesterol mg/dl	Males	198.8 ± 36.5	195.1 ± 30.6	Not significant 0.260	20	246.7 ± 44.8	229.2 ± 36.0	Not significant 1.255	32
	Females	209.2 ± 33.0	198.2 ± 27.4	Not significant 0.812	18	255.2 ± 36.6	241.8 ± 39.9	Not significant 0.818	20
Tri-glycerides mg/dl	Males	135.5 ± 44.8	121.7 ± 46.7	Not significant 0.703	20	152.2 ± 32.3	125.2 ± 33.4	Significant 2.401**	32
	Females	127.1 ± 51.8	105.5 ± 46.9	Not significant 0.978	18	156.2 ± 38.5	138.8 ± 25.4	Not significant 1.248	20
LDL Cholesterol mg/dl	Males	122.4 ± 31.9	119.6 ± 22.6	Not significant 0.230	20	176.5 ± 48.8	158.2 ± 36.7	Not significant 1.268	32
	Females	129.5 ± 24.5	117.9 ± 15.6	Not significant 1.262	18	177.3 ± 33.7	164.3 ± 37.8	Not significant 0.851	20
Apolipo-protein mg/dl	Males	190.9 ± 16.7	191.7 ± 14.8	Not significant 0.122	20	181.4 ± 6.0	188.2 ± 7.1	Significant 2.990***	32
	Females	191.8 ± 12.8	194.8 ± 13.1	Not significant 0.518	18	182.2 ± 11.9	186.6 ± 10.8	Not significant 0.904	20

Significance *** less than 1.0%
** less than 5.0%
* less than 7.5%

v total degrees of freedom

† mean ± S.D.

of the study. Their mean HDL-cholesterol and apolipoprotein-A levels increased significantly and their triglycerides level decreased significantly in this group.

Tables VI-A,B show the comparison of the exercise and control groups at the initial and final stage of the programme. The exercise males, under 40 years of age, showed no significant difference from the control males of the same age group at either the initial or final stage for HDL-cholesterol, triglycerides and apolipoprotein-A levels and relatively low degree of significances for total-cholesterol and LDL-cholesterol.

Initially, the exercise males 40 years and over showed a significantly lower level of HDL-cholesterol and significantly higher level of total-cholesterol, triglycerides and LDL-cholesterol than the control males of the same age group. These differences had disappeared by the final stage of the exercise programme.

The exercise and control females under 40 years showed no significant differences in all parameters at either the initial or final periods.

The exercise females 40 years and over showed no significant differences from the control females of the same age group at any time during the 16-week period for HDL-cholesterol, triglycerides

TABLE VI-A

COMPARISON OF PLASMA LIPID PARAMETERS† BETWEEN EXERCISE AND NON-PARTICIPATING CONTROL WITHIN MALE AGE GROUPS

		INITIAL			FINAL			
		Exercise Group	Non-Participating Group	Difference-t	Exercise Group	Non-Participating Group	Difference-t	v
HDL Cholesterol mg/dl	< 40 years	49.1 ± 13.5	49.1 ± 6.5	Not significant 0.004	51.3 ± 12.9	51.0 ± 5.2	Not significant 0.059	18
	≥ 40 years	39.9 ± 8.5	49.3 ± 4.6	Significant 2.892***	45.8 ± 8.0	49.6 ± 6.2	Not significant 1.187	23
Total Cholesterol	< 40 years	198.8 ± 36.5	218.6 ± 30.1	Not significant 1.299	195.1 ± 30.6	214.3 ± 22.7	Significant 1.563*	18
	≥ 40 years	246.7 ± 44.8	212.3 ± 33.4	Significant 1.931**	229.2 ± 36.0	212.8 ± 32.9	Not significant 1.097	23
Tri-glycerides	< 40 years	135.5 ± 44.8	138.3 ± 42.6	Not significant 0.146	121.7 ± 46.7	138.3 ± 35.4	Not significant 0.878	18
	≥ 40 years	152.2 ± 32.3	125.4 ± 20.3	Significant 2.147**	125.2 ± 33.4	123.3 ± 19.7	Not significant 0.150	23
LDL Cholesterol mg/dl	< 40 years	122.4 ± 31.9	141.8 ± 27.5	Not significant 1.438	119.6 ± 22.6	136.6 ± 20.8	Significant 1.723*	18
	≥ 40 years	176.5 ± 48.8	137.6 ± 35.2	Significant 2.077**	158.2 ± 36.7	138.6 ± 35.3	Not significant 1.266	23
Apolipo-protein mg/dl	< 40 years	190.9 ± 16.7	188.0 ± 13.0	Not significant 0.427	191.7 ± 14.8	190.3 ± 12.1	Not significant 0.229	18
	≥ 40 years	181.4 ± 6.0	185.3 ± 8.2	Not significant 1.328	188.2 ± 7.1	186.0 ± 7.9	Not significant 0.690	23

v total degrees of freedom

† mean ± S.D.

Significance *** less than 1.0%
** less than 5.0%
* less than 7.5%

TABLE VI-B

COMPARISON OF PLASMA LIPID PARAMETERS† BETWEEN EXERCISE AND NON-PARTICIPATING CONTROL WITHIN FEMALE AGE GROUPS

		INITIAL			FINAL			
		Exercise Group	Non-Participating Group	Difference-t	Exercise Group	Non-Participating Group	Difference-t	v
HDL Cholesterol mg/dl	< 40 years	54.4 ± 8.8	57.6 ± 6.3	Not significant 0.980	59.5 ± 9.2	57.7 ± 7.1	Not significant 0.494	19
	≥ 40 years	46.7 ± 9.4	50.3 ± 5.8	Not significant 0.933	50.7 ± 8.1	50.1 ± 6.0	Not significant 0.180	17
Total-Cholesterol mg/dl	< 40 years	209.2 ± 33.0	191.0 ± 45.4	Not significant 1.041	198.2 ± 27.4	204.0 ± 36.8	Not significant 0.406	19
	≥ 40 years	255.2 ± 36.6	223.5 ± 46.1	Significant 1.672*	241.8 ± 39.9	236.4 ± 42.0	Not significant 0.287	17
Tri-glycerides mg/dl	< 40 years	127.1 ± 51.8	112.9 ± 37.1	Not significant 0.727	105.5 ± 46.9	116.7 ± 31.1	Not significant 0.652	19
	≥ 40 years	156.2 ± 38.5	136.4 ± 28.0	Not significant 1.231	138.8 ± 25.4	135.6 ± 23.1	Not significant 0.281	17
LDL Cholesterol mg/dl	< 40 years	129.5 ± 24.5	110.9 ± 42.6	Not significant 1.208	117.9 ± 15.6	123.0 ± 36.6	Not significant 0.407	19
	≥ 40 years	177.3 ± 33.7	145.6 ± 40.3	Significant 1.862**	164.3 ± 37.8	159.8 ± 38.0	Not significant 0.257	17
Apolipo-protein mg/dl	< 40 years	191.8 ± 12.8	197.7 ± 15.9	Not significant 0.932	194.8 ± 13.1	196.1 ± 13.2	Not significant 0.225	19
	≥ 40 years	182.2 ± 11.9	184.0 ± 19.0	Not significant 0.758	186.6 ± 10.8	183.3 ± 15.4	Not significant 0.552	17

v total degrees of freedom

† mean ± S.D.

Significance *** less than 1.0%
** less than 5.0%
* less than 7.5%

and apolipoprotein-A levels. However, significant differences were observed in the initial stage for total-cholesterol and LDL-cholesterol levels. By 16 weeks such changes had disappeared.

<u>Comparison of initial and final stage data with respect to the HDL-cholesterol Range Subgroup</u>

In keeping with standard clinical interpretation, HDL-cholesterol normal ranges of 40-54 mg/dl for males and 50-68 mg/dl for females were used to establish Range Subgroups (RS); i.e. for males RS_1 is $\geq$ 40 mg/dl, RS_2 < 40 mg/dl; for females RS_1 is $\geq$ 50 mg/dl, RS_2 < 50 mg/dl. This subgrouping enabled us to observe the response of individual levels to the exercise programme.

Table VII shows that there were significant differences between initial and final weeks for the exercise males RS_2 only in HDL-cholesterol, triglycerides and apolipoprotein-A levels. Among the exercise females there was a significant difference in HDL-cholesterol in RS_2.

TABLE VII

COMPARISON OF PLASMA LIPID PARAMETERS BETWEEN THE INITIAL AND FINAL WEEKS OF THE STUDY PERIOD WITHIN HDL-CHOLESTEROL RANGE SUBGROUP OF THE EXERCISE AND NON-PARTICIPATING CONTROL GROUPS

(SUBGROUPS SELECTED BY INITIAL PLASMA HDL-CHOLESTEROL LEVELS)

	HDL-CHOLESTEROL			TOTAL CHOLESTEROL			TRIGLYCERIDES			LDL-CHOLESTEROL			APOLIPOPROTEIN		
	(1)	(2)	t	(1)	(2)	t	(1)	(2)	t	(1)	(2)	t	(1)	(2)	t
♂ CONTROL RS_1	49.2	50.3	0.150	215.6	213.6	0.201	132.2	131.2	0.092	139.8	137.5	0.236	187.0	187.9	0.256
♀ CONTROL RS_1	58.9	59.9	0.411	210.4	214.7	0.495	119.4	118.6	0.039	128.1	131.0	0.171	198.6	198.9	0.054
♀ CONTROL RS_2	47.2	47.4	0.164	213.2	223.9	0.694	138.8	139.6	0.058	137.9	149.3	0.797	176.0	175.2	0.245
♂ EXERCISE RS_1	50.1	52.6	0.749	218.5	213.5	0.336	141.7	125.9	1.558 *	140.2	135.7	0.318	189.6	193.3	0.847
♂ EXERCISE RS_2	33.7	40.8	3.026 ***	242.4	219.3	1.635 *	151.7	120.6	2.348 ***	178.4	154.4	1.343	178.2	183.8	2.368 **
♀ EXERCISE RS_1	59.0	62.7	1.277	234.6	229.5	0.266	136.5	119.0	0.848	148.4	143.3	0.288	197.6	199.7	0.697
♀ EXERCISE RS_2	42.5	47.8	2.881 ***	232.1	213.4	1.092	147.6	126.5	1.163	160.1	141.2	1.195	176.9	182.1	1.302

HDL-CHOLESTEROL RANGE SUBGROUP (RS):

♂ RS_1 ≥ 40 mg/dl, RS_2 < 40 mg/dl

♀ RS_1 ≥ 50 mg/dl, RS_2 < 50 mg/dl

(1) mean value at initial stage

(2) mean value at final stage

Significance *** less than 0.1%
** less than 5.0%
* less than 7.5%

DISCUSSION

In 1951, Barr[4] and his associates at Cornell Medical Centre noted that thirty-three patients with coronary artery disease had relatively low levels of High Density Lipoproteins in their plasma. They proposed that HDL appeared to have a protective function against coronary heart disease. This study has now been amply substantiated by the work of Miller[30, 31] and Castelli.[9]

In 1977, the Framingham Heart Study[18] identified a number of risk factors associated with heart disease. This study showed that the risk of an individual developing clinical coronary artery heart disease is inversely related to the plasma HDL-cholesterol level.

Our experiment was designed to study the effect of "physical fitness" exercise on the plasma levels of HDL-cholesterol and other lipid fractions. We were most interested in studying the HDL-cholesterol levels since this fraction is, as noted above, a measure of an individual's risk to develop coronary artery disease.

Review of lipid biochemistry   There are three major classes of lipoproteins:[39] very low density lipoprotein (VLDL); low density lipoprotein (LDL) and high density lipoprotein (HDL). A fourth class, the chylomicron, which transports absorbed lipid from the small intestine to the liver, is normally absent in the fasting state. The different classes of lipoproteins are distinguished by particle size, density, lipid and apolipoprotein[15] composition.

High density lipoprotein is the smallest of the plasma lipoproteins with relatively high density (1.063 - 1.210 g/ml). The major lipid is phospholipid. The major apolipoprotein (apo) is A (both AI and AII) with a minor fraction apolipoprotein-C. Its ultrastructure is that of a micelle,[6] composed of an outer coat of phospholipid, unesterified cholesterol and specific protein, and an inner core of cholesterol ester and triglyceride.

The very low density lipoprotein (VLDL) is the vehicle for the transport of endogenous lipids to peripheral cells. The triglyceride-rich VLDL contains mainly apolipoprotein-C with minor fractions of apo A and E. In peripheral tissues, such as muscle and adipose tissue, VLDL triglyceride is hydrolysed by the action of lipoprotein lipase.[31, 39] As the VLDL is catabolized, the apolipoprotein-C is recaptured by HDL in exchange for the cholesterol esters. This process of delipidation converts the VLDL to cholesterol-rich LDL containing mainly apolipoprotein-B with minor fractions of C and E.

The catabolism of LDL[34] appears to occur largely in extra-hepatic tissue by a receptor mediated process. The binding of LDL to the cell surface results in endocytosis of the lipoprotein and its eventual degradation in the lysosomes. The unesterified cholesterol is then free to be used by the cell for membrane synthesis and other metabolic processes requiring the steroid nucleus[6]. The excess cholesterol from tissue cells is most likely transported to the liver for excretion by HDL.[6]

It is through this reverse transportation by HDL in carrying cholesterol from tissue and arterial walls to the liver for excretion that HDL is thought to prevent atherogenesis.[3] Tissue culture studies have also shown that HDL inhibits the uptake of the cholesterol-rich LDL by arterial wall smooth muscle. Thus, a high plasma HDL level appears to function as a protective factor against atherosclerosis.[9]

It is known that HDL-cholesterol is synthesized in the liver and small intestine.[39] It is also recognized that HDL-cholesterol level can be altered by several physiological and pathological conditions. Lower HDL-cholesterol concentrations tend to occur in individuals who are on a diet rich in carbohydrate and in those who are sedentary, obese,[36] cigarette smokers,[10] diabetic or uremic.[39] Higher levels of

HDL-cholesterol are found in individuals with moderate alcohol consumption.[16] Women on estrogen and/or contraceptive therapy tend to have elevated HDL-cholesterol levels. In the males, high levels of HDL-cholesterol in the pre-adolescent begin to drop in adolescence[39] to reach a lower level in adultcy.[39] The impact of sex hormones on HDL-cholesterol metabolism is well-demonstrated in our data (Table 1 and Figure 1) showing significant differences between male and female exercise groups. This result agrees with the recent report by Campione.[8] Furthermore, significant age effect is evident in both exercise and control females (Table IV-A-B). This finding reiterates the importance of hormonal influence on the lipid levels.

HDL-cholesterol is assumed to be a protective factor against atherosclerosis,[9] a correlation study (Table III) between HDL and the other lipid fractions could provide an insight into these metabolic relationships. Our results (Table III and Figures VI-IX) show a negative correlation between HDL-cholesterol and total-cholesterol which reflects a significant rise in HDL-cholesterol while the total cholesterol remains relatively constant throughout the study. A negative correlation was observed between HDL-cholesterol and LDL-cholesterol, and between HDL-cholesterol and triglycerides. These results show that a shift towards a more beneficial distribution of the total lipid is brought on by the exercise programme.

Can a 16-week exercise programme of moderate intensity be beneficial to sedentary individuals as far as lipid distribution is concerned? Our data provides some evidence to suggest yes, if that individual is over 40 years old, or has a low HDL-cholesterol level.

After 16 weeks of exercise, both gender groups did show marginal significant increase in HDL-cholesterol (Table 1). When age groups were considered (Table V-B) the increase in HDL-cholesterol was restricted to the over 40 year old males only. When the study subjects were divided by HDL-cholesterol Range (Table VII) the subnormal individuals (RS_2) in both genders also showed a marked significant increase in HDL-cholesterol.

Most reports[21, 25, 28, 43, 44] suggest that such an increase in HDL-cholesterol level is the result of strenuous physical training. Our study demonstrates that the efficacy of an exercise programme of moderate intensity (average 15 kilometers of jogging per week) is sufficient to elevate the HDL-cholesterol level in certain individuals.

Our study (Tables 1 - 11) showed minimal changes in apolipoprotein-A throughout the course of the exercise programme. When the groups were age-separated (Table V-B), the over 40 year males did show lower levels at the initial stage and this difference disappeared at the 16th week. When the groups were separated by HDL-cholesterol Range (Table VII) the male subnormal group (RS_2) showed a significant increase in apolipoprotein-A. In liver and intestinal perfusion studies of rats,[39] both High Density Lipoprotein (HDL) and apolipoprotein-A are found to be synthesized in these organs. Approximately 90% of the total apolipoprotein-A is found in the High Density Lipoprotein fraction. Measurement of apolipoprotein-A is a measurement of HDL. This explains the positive correlation between these two parameters.

Our data (Tables I and V-A,B) did not show any significant change in total-cholesterol and LDL-cholesterol levels during the exercise programme. However, when the groups were separated by age, those males and females over 40 years initially showed higher levels than their corresponding controls (Table VI-A,B). By the final week, no significant differences were observed. Does this imply that exercise activity lowered the total-cholesterol and LDL-cholesterol in this age group? In our judgement, exercise has minimal effect on total-cholesterol. This is in agreement with the recent literature.[7, 22, 26, 29, 42] Moreover, one should not expect any significant change in LDL-cholesterol since there were reciprocal changes in HDL-cholesterol and triglycerides in the presence of relatively constant total-cholesterol levels.

Lewis[26] and Haskell[20] reported that exercise decreased the triglyceride levels of hypertriglyceridemic individuals. In the present study, only the male group over 40 years showed a significant decrease in triglycerides. In the HDL-cholesterol subnormal group (RS_2) the same observation was noted. However, 90% of all exercise participants experienced a numerical decrease in plasma triglyceride levels.

The process of exercise-induced reduction in triglycerides is complicated. It may be explained by the combination of increased tissue uptake and increase of lipoprotein lipase activity.[31, 32] The latter is responsible for the catabolism of triglyceride-rich Very Low Density Lipoprotein in peripheral tissues.[23] Physical exercise can influence the activity of lipoprotein lipase in both human and other animals.[33, 5] In the present study, a negative correlation coefficient between HDL-cholesterol and triglycerides is observed (Table III and Figure VI). These reciprocal changes in triglycerides and HDL-cholesterol may be due to the activation of lipoprotein lipase activity. Tamir[39] shows that the hydrolysis of triglycerides results in the liberation of apolipoprotein-C which is transferred to the High Density Lipoprotein. This direct transfer of apolipoprotein to High Density Lipoprotein may explain the negative correlation of these two parameters.

In conclusion, within the limitation of sample size, our results suggest that participation in a moderate physical fitness programme designed for the sedentary individuals can have a beneficial effect on plasma lipid parameters particularly in males over 40 years of age. However, we cannot assume that exercise alone was responsible for these changes since we are not certain that other aspects of the lifestyle of the participants remained unchanged. Since higher levels of High Density Lipoprotein cholesterol are associated with a lower incidence of coronary heart disease,[3, 15, 24, 27, 31, 35] our study supports the view that voluntary and moderate exercise has not only a recreational effect, but at the same time could reduce the incidence of morbidity due to coronary heart disease.

SAMPLE DATA - EXERCISE GROUP (FEMALE)

HDL-Cholesterol mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	27	45	50	54	49	53
2	51	45	48	49	47	49
3	50	37	38	42	42	45
4	36	63	63	64	63	67
5	26	57	54	63	63	64
6	49	65	66	69	69	67
7	35	56	59	62	59	62
8	26	52	50	51	54	53
9	48	58	57	56	54	60
10	51	41	42	42	45	45
11	28	42	42	44	44	45
12	50	50	49	48	50	55
13	38	69	72	73	70	76
14	43	57	50	54	51	56
15	51	39	42	40	41	44
16	30	48	53	55	54	55
17	49	43	50	52	51	51
18	50	39	37	36	39	40
19	50	40	42	45	44	46
20	28	49	50	51	51	53
21	29	63	62	64	62	67
Mean ± S.D.		50.3 ± 9.7	51.2 ± 9.3	53.0 ± 10	52.5 ± 9.0	54.9 ± 9.5

SAMPLE DATA - EXERCISE GROUP (MALE)

HDL-Cholesterol mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	50	38	43	40	45	44
2	34	54	55	52	61	60
3	35	39	46	44	48	46
4	48	27	30	30	34	33
5	26	51	53	50	52	49
6	48	38	43	44	45	44
7	51	34	39	39	41	43
8	27	43	45	46	45	43
9	51	25	30	30	33	33
10	32	49	50	47	50	49
11	26	41	43	43	44	39
12	49	46	49	52	52	49
13	51	35	43	40	44	45
14	38	59	60	56	62	63
15	29	75	76	74	76	75
16	47	61	65	60	66	66
17	48	42	49	50	53	51
18	49	32	34	34	35	35
19	45	42	43	44	46	45
20	40	37	42	41	44	45
21	42	43	44	40	45	46
22	41	50	55	53	54	55
23	45	40	44	42	45	46
24	43	45	52	52	53	51
25	26	26	32	31	34	35
26	28	63	64	54	64	66
27	30	44	43	39	44	39
28	46	43	48	50	53	48
Mean ± S.D.		43.6 ± 11.4	47.1 ± 10.6	45.6 ± 9.6	48.9 ± 10.2	48.0 ± 10.3
Male & female Combined						
Mean ± S.D.		46.5 ± 11.1	48.9 ± 10.2	48.8 ± 10.4	50.4 ± 9.8	50.9 ± 10.4

SAMPLE DATA - EXERCISE GROUP (FEMALE)

Total-Cholesterol mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	27	219	220	201	190	179
2	51	288	276	279	258	261
3	50	229	210	201	200	182
4	36	246	240	223	210	216
5	26	219	210	234	207	231
6	49	313	340	350	310	302
7	35	210	199	191	200	197
8	26	148	161	161	157	164
9	48	247	259	236	257	241
10	51	289	264	267	323	282
11	28	188	172	181	186	181
12	50	236	232	220	220	228
13	38	266	258	261	260	254
14	43	255	285	277	270	279
15	51	253	250	256	250	248
16	30	181	189	186	179	189
17	49	272	266	265	260	238
18	50	175	180	187	178	172
19	50	250	215	228	258	227
20	28	209	197	195	217	188
21	29	206	210	189	190	183
Mean ± S.D.		233.3±41.4	230.1±43.4	228.0±44.9	227.6±44.6	221.0±40.4

SAMPLE DATA - EXERCISE GROUP (MALE)

Total-Cholesterol mg/dl.

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	50	278	260	284	265	257
2	34	176	180	181	180	165
3	35	238	240	245	250	214
4	48	271	240	236	241	260
5	26	190	176	207	199	201
6	48	210	201	213	237	206
7	51	328	289	266	278	258
8	27	222	220	222	200	219
9	51	171	170	161	160	152
10	32	143	190	172	150	144
11	26	141	160	135	140	155
12	49	252	265	298	249	250
13	51	301	308	277	289	252
14	38	221	228	235	230	212
15	29	249	244	240	260	249
16	47	253	233	241	231	250
17	48	253	240	240	235	231
18	49	227	230	240	240	225
19	45	315	298	290	289	291
20	40	201	210	208	200	204
21	42	240	230	199	190	236
22	41	260	260	261	240	230
23	45	219	240	248	230	192
24	43	243	240	236	230	239
25	26	222	220	213	194	192
26	28	209	196	150	224	203
27	30	176	184	164	187	192
28	46	172	181	197	227	164
Mean±S.D.		227.9±47.4	226.2±38.6	223.5±42.8	223.0±38.8	215.8±37.4
Male & Female Combined						
Mean S.D.		230.2±44.6	227.8±40.3	225.5±43.3	225.0±41.0	218.0±38.4

SAMPLE DATA - EXERCISE GROUP (FEMALE)

Triglycerides mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	27	121	101	75	81	80
2	51	124	100	120	100	108
3	50	207	187	178	161	175
4	36	110	141	109	80	101
5	26	231	210	221	200	195
6	49	107	110	98	118	102
7	35	87	33	61	48	51
8	26	92	78	86	76	71
9	48	144	141	139	140	126
10	51	188	170	166	150	168
11	28	129	120	121	110	106
12	50	162	129	126	131	124
13	38	205	190	188	180	176
14	43	102	120	126	130	125
15	51	162	150	141	130	126
16	30	68	51	40	49	66
17	49	217	188	176	172	165
18	50	131	100	136	120	147
19	50	174	149	168	169	161
20	28	103	95	86	79	90
21	29	125	110	128	131	119
Mean ± S.D.		142.3±46.6	127.3±45.9	128.0±44.9	121.7±42.4	122.9±40.0

SAMPLE DATA - EXERCISE GROUP (MALE)

Triglycerides mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	50	106	105	99	80	86
2	34	109	100	91	80	71
3	35	121	115	110	105	112
4	48	158	147	136	184	141
5	26	186	195	166	155	188
6	48	121	101	75	81	83
7	51	140	110	102	100	86
8	27	107	100	98	100	102
9	51	131	130	102	110	100
10	32	66	39	49	75	56
11	26	99	100	101	100	81
12	49	174	120	140	136	151
13	51	157	144	140	118	127
14	38	199	189	190	181	187
15	29	183	162	159	144	175
16	47	196	183	172	183	190
17	48	158	160	122	100	113
18	49	177	165	151	150	136
19	45	86	77	91	72	79
20	40	210	186	199	191	179
21	42	148	150	118	120	122
22	41	191	178	170	160	167
23	45	162	155	141	130	126
24	43	142	130	134	125	130
25	26	186	160	144	149	151
26	28	124	109	111	120	103
27	30	110	90	101	131	113
28	46	131	104	120	118	112
Mean ± S.D.		145.6±37.8	132.3±38.5	126.1±35.2	124.9±34.8	123.8±38.4
Male & Female Combined						
Mean ± S.D.		144.2±41.3	130.1±41.5	127.0±39.2	123.5±37.9	123.4±38.7

SAMPLE DATA - EXERCISE GROUP (FEMALE)

LDL-Cholesterol mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	27	150	150	132	125	110
2	51	218	208	206	191	190
3	50	150	135	123	126	102
4	36	161	149	137	131	129
5	26	116	114	127	104	128
6	49	226	252	260	217	215
7	35	137	133	117	131	125
8	26	78	94	93	88	97
9	48	160	174	152	175	156
10	51	211	188	192	248	203
11	28	120	106	113	122	115
12	50	154	157	147	144	148
13	38	156	148	151	154	145
14	43	178	211	198	193	198
15	51	182	178	188	183	179
16	30	120	126	123	115	121
17	49	186	179	178	175	164
18	50	110	123	124	116	103
19	50	175	143	149	180	149
20	28	139	128	127	150	117
21	29	118	126	100	102	92
Mean ± S.D.		154.5±37.9	153.4±38.9	149.4±40.9	150.9±41.4	142.2±37.2

SAMPLE DATA - EXERCISE GROUP (MALE)

LDL-Cholesterol mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	50	219	196	224	204	196
2	34	100	105	111	103	91
3	35	175	171	179	181	146
4	48	212	181	179	172	199
5	26	102	84	124	116	114
6	48	148	138	154	174	145
7	51	266	228	207	217	198
8	27	158	155	156	135	156
9	51	120	114	111	105	99
10	32	81	132	115	85	84
11	26	80	97	72	76	100
12	49	171	192	218	170	171
13	51	235	236	209	221	182
14	38	122	130	141	132	112
15	29	138	136	134	155	139
16	47	153	132	147	129	146
17	48	179	159	166	162	157
18	49	160	161	176	175	162
19	45	256	240	228	229	230
20	40	122	131	127	118	123
21	42	167	156	136	121	166
22	41	172	170	174	154	141
23	45	147	165	178	159	121
24	43	170	162	157	152	159
25	26	159	156	153	130	127
26	28	121	110	74	136	117
27	30	110	123	105	117	130
28	46	103	112	123	150	94
Mean ± S.D.		155.2±49.0	152.6±40.3	152.8±41.9	149.2±39.3	143.0±36.8

SAMPLE DATA - EXERCISE GROUP (FEMALE)

Apolipoprotein-A mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	27	178	175	186	185	178
2	51	180	175	182	183	180
3	50	174	170	180	180	178
4	36	207	210	215	210	211
5	26	200	200	197	200	199
6	49	199	205	202	198	200
7	35	201	195	202	191	209
8	26	187	190	191	194	187
9	48	198	200	195	190	198
10	51	171	172	170	175	183
11	28	168	164	170	180	178
12	50	198	193	205	200	198
13	38	195	195	195	205	201
14	43	189	191	194	189	190
15	51	170	167	166	170	174
16	30	180	180	183	175	179
17	49	180	180	188	191	197
18	50	168	165	165	168	169
19	50	177	175	176	178	185
20	28	200	199	206	200	202
21	29	202	210	202	200	204
Mean ± S.D.		186.8±13.0	186.2±15.0	189.0±14.2	188.7±11.9	190.4±12.3

SAMPLE DATA - EXERCISE GROUP (MALE)

Apolipoprotein-A mg/dl

No.	Age	Baseline	4th Week	8th Week	12th Week	16th Week
1	50	176	175	181	185	182
2	34	197	200	203	200	204
3	35	183	180	181	190	187
4	48	183	185	187	190	188
5	26	208	200	201	200	205
6	48	174	175	181	185	186
7	51	175	170	175	172	181
8	27	180	177	181	184	176
9	51	176	180	184	186	185
10	32	181	174	179	180	185
11	26	169	171	173	178	175
12	49	182	180	188	190	187
13	51	176	171	179	184	184
14	38	201	200	210	205	196
14	29	218	218	212	209	213
16	47	192	190	195	200	200
17	48	182	180	191	185	187
18	49	183	185	185	184	187
19	45	178	174	183	183	182
20	40	179	175	178	180	181
21	42	177	184	181	180	181
22	41	195	199	196	195	200
23	45	187	190	192	190	190
24	43	186	190	193	195	203
25	26	168	170	174	175	171
26	28	209	211	215	205	210
27	30	186	181	178	185	187
28	46	183	190	183	190	195
Mean ± S.D.		185.1±12.1	184.7±12.6	187.8±11.6	188.8±9.4	189.6±10.7
Male and Female Combined						
Mean ± S.D.		185.8±12.4	185.3±13.6	188.3±12.7	188.7±10.4	189.9±11.3

SAMPLE DATA (mg/dl) - NON-PARTICIPATING GROUP (FEMALE)

		HDL-Cholesterol		Total-Cholesterol		Triglycerides		LDL-Cholesterol		Apolipo-Protein	
No.	Age	Base	16 Wk	Base	16 Wk	Base	16 Wk	Base	16 Wk	Base	16 Wk
1	49	49	46	232	241	161	148	150	165	178	173
2	25	66	66	201	239	55	74	125	158	211	214
3	32	58	57	251	248	210	196	151	152	197	195
4	27	58	59	156	172	78	88	82	95	203	210
5	27	54	59	187	168	89	77	119	94	192	206
6	25	51	56	186	204	104	110	114	126	190	199
7	34	48	55	196	222	150	129	118	141	191	180
8	41	51	52	199	210	121	127	124	133	168	164
9	39	55	51	193	184	146	141	109	105	196	189
10	53	47	51	182	204	101	121	115	129	169	178
11	52	47	46	241	279	188	175	156	198	177	180
12	38	47	46	248	226	186	166	163	147	162	168
13	33	49	50	167	179	109	128	96	106	179	178
14	50	46	44	182	176	120	135	112	105	179	179
15	28	57	61	248	229	187	181	155	132	202	211
16	42	59	56	201	221	119	95	118	146	216	203
17	24	56	59	150	179	112	88	72	102	207	203
18	27	64	63	164	154	160	144	71	62	194	186
19	31	55	58	159	190	140	154	76	101	189	190
20	30	65	64	193	188	75	102	113	103	191	192
21	36	51	46	248	228	191	178	159	146	166	173
22	26	69	72	211	204	106	144	121	103	194	201
23	32	66	70	201	222	90	103	117	131	201	192
24	35	48	44	243	231	102	111	175	165	169	169
25	48	44	45	228	257	132	143	156	188	173	179
26	30	66	64	146	161	91	106	62	76	221	199
27	29	62	68	206	185	88	122	126	92	209	205
28	28	61	56	294	286	126	114	208	207	199	187
29	30	55	54	180	183	111	101	103	112	188	186
30	54	59	61	323	303	149	141	234	214	212	210
Mean		55.4	56.0	206.8	211.4	126.6	127.9	126.6	131.1	190.8	190.0
S.D.		±7.2	±8.0	±42.9	±39.7	±37.1	±31.6	±39.0	±38.2	±16.0	±14.3
p		0.39 (N.S.)*		0.34 (N.S.)*		0.44 (N.S.)*		0.33 (N.S.)*		0.42 (N.S.)*	

* no significant difference

SAMPLE DATA (mg/dl) - NON-PARTICIPATING GROUP (MALE)

		HDL-Cholesterol		Total-Cholesterol		Triglycerides		LDL-Cholesterol		Apolipo-Protein	
No.	Age	Base	16 Wk	Base	16 Wk	Base	16 Wk	Base	16 Wk	Base	16 Wk
1	45	44	42	249	238	141	149	174	166	175	180
2	36	42	45	201	197	149	141	129	124	176	180
3	47	48	46	249	268	129	115	175	199	187	179
4	51	44	45	207	217	130	122	137	148	173	180
5	48	51	58	236	226	116	133	162	141	188	192
6	43	46	44	197	187	107	89	130	125	181	177
7	27	61	56	197	191	125	131	111	109	192	194
8	27	44	48	240	232	191	178	158	148	186	184
9	28	46	47	161	186	102	110	95	117	177	175
10	50	50	52	152	160	141	143	74	80	190	194
11	42	56	53	220	209	89	106	146	135	196	189
12	30	49	51	231	210	125	120	157	135	192	201
13	20	42	44	268	251	192	179	188	177	166	172
14	37	54	56	229	242	158	173	143	153	206	203
15	31	55	58	219	212	57	72	152	140	196	192
16	48	55	57	188	197	150	129	103	114	192	197
17	39	49	54	221	208	146	141	143	126	201	206
Mean		49.2	50.4	215.6	213.6	132.8	131.2	139.8	137.5	186.7	187.9
S.D.		±5.5	±5.6	±30.9	±27.1	±33.9	±29.3	±30.4	±37.6	±10.7	±10.4
p		0.27	(N.S.)*	0.42	(N.S.)*	0.44	(N.S.)*	0.42	(N.S.)*	0.37	(N.S.)*

* no significant difference

APPENDIX 11

TABLE V111-A

NON-PARTICIPATING FEMALE CONTROL DATA REDUCTION

AGE SUBGROUPS

SUBGROUPS OVER 40 YEARS			UNDER 40 YEARS			UNDER 40 YEARS RESIDUAL		
NO.	AGE	INITIAL HDL-CHOL. LEVEL	NO.	AGE	INITIAL HDL-CHOL. LEVEL	NO.	AGE	INITIAL HDL-CHOL. LEVEL
1	49	49	2	25	66	4	27	58
8	41	51	3	32	58	5	27	54
10	53	47	6	25	51	7	34	48
11	52	47	9	39	55	12	38	47
14	50	46	13	33	49	15	28	57
16	42	59	17	24	56	18	27	64
25	48	44	19	31	55	20	30	65
30	54	59	21	36	51	22	26	69
			23	32	66	24	35	48
			26	30	66	27	29	62
			28	28	61	29	30	55
N		8			11			11
MEAN		50.3			57.6			57.0
S.D.		±5.8			±6.3			± 7.5

APPENDIX 11 TABLE V111-B

NON-PARTICIPATING FEMALE CONTROL DATA REDUCTION

INITIAL HDL-CHOLESTEROL RANGE SUBGROUPS

(RS_1 OVER 50 mg/dl, RS_2 UNDER 50 mg/dl)

SUBGROUPS

RS_2			RS_1			RS_1 RESIDUAL		
NO.	AGE	HDL-CHOL.	NO.	AGE	HDL-CHOL.	NO.	AGE	HDL-CHOL.
1	49	49	2	25	66	4	27	58
7	34	48	3	32	58	8	41	51
10	53	47	5	27	54	16	42	59
11	52	47	6	25	51	18	27	64
12	38	47	9	39	55	19	31	55
13	33	49	15	28	57	21	36	51
14	50	46	17	24	56	22	26	69
24	35	48	20	30	65	26	30	66
25	48	44	23	32	66	27	29	62
			28	28	61	29	30	55
						30	54	59
N		9			10			11
MEAN		47.2			58.9			59.0
S.D.		±1.6			±5.3			±5.9

The effects of age and the initial HDL-cholesterol level on all plasma parameter changes, over the duration of the study period, were investigated. It was found that the female sample numbers in the control group under 40 years and RS_1 Subgroups were 2.75 and 2.33 times larger than the over 40 years and RS_2 Subgroups respectively. To avoid a violation of the two-sample t-test assumption of equal variances, it was decided to reduce these sample data by 50% through random selection of the participant number codes. The two sets of under 40 year age and RS_1 Subgroups were then tested for homogeneity by the t-test. The null hypothesis was supported in each case by t-values of under 1.0. Therefore it was concluded that the sample data reduction did not bias these comparisons.

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