

P35

LIST OF PROCEDURES DONE
PROJECT REPORT

TITLE OF THE PROJECT:

RISK FACTORS WITH SPECIAL REFERENCE TO
LIPOPROTEIN PROFILES IN SURVIVORS OF
MYOCARDIAL INFARCTION BELOW THE AGE OF
40 YEARS.

NAME.....K. K. HARIDAS.....

PROGRAMME:.....DM CARDIOLOGY.....

MONTH & YEAR
OF SUBMISSION:.....NOVEMBER 1988.....

Forwarded
K. K. Haridas
25/11/88

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Date	

- Note:—
- (i) In the case compilation of procedures done, the contents and the subsequent pages should be made into different sections (a) Procedures done (b) Procedures assisted (c) Procedures participated (d) Procedures attended/participated etc in Other Centres. Each section should be preceded by a leaf carrying the name of the section that is succeeding.
 - (ii) The Contents page will carry into, as per model given under

PROCEDURES DONE

Closed Mitral valvotomy.....124 (say)
 Patent ductus arteriosus-ligation.....10
 Atrial septal defects.....20

PROCEDURES ASSISTED

Closed Mitral valvotomy.....100 (say)

- (iii) In the subsequent pages details of each procedure done/assisted should be given in the format given below:—

Heading: **Closed mitral valvotomy**

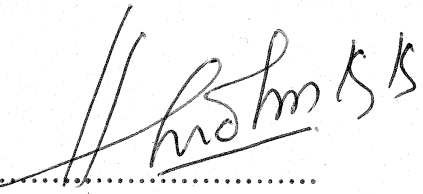
Date	Name of the patient	Age	Sex	Patient No.
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- (iv) In the case of Project Report in the page immediately following the Certificate page the under-mentioned details should be given:—

- (a) Title
- (b) Duration
- (c) Aim and scope
- (d) 50 word summary of work done

CERTIFICATE

I, Dr.....K. K. HARIDAS.....hereby declare that I have actually performed all the procedures listed/carried out the project under report.

Signature.....

Place: TRIVANDRUM
Name in.....K. K. HARIDAS
Date: 1/11/88 capital letters

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INTRODUCTION

Less than one percent of all myocardial infarctions occur in subjects below the age of 40 years¹. Several reports in the recent years have addressed to the problem of acute myocardial infarction in younger subjects^{2,3,4,5,6,7,8}. Reports from the developed countries show a male predominance^{2,3,4,5,6,7}. Atherosclerotic coronary occlusive disease forms the major cause of myocardial infarction in males below 40 years of age^{2,3,9}. In contrast female subjects in the same age group show a higher proportion of nonatheromatous causes like coronary thrombosis, spasm, embolism, and coronary arteritis⁸. The less common causes of myocardial infarction in the young include, anomalies of coronary arterial origin, myocardial bridges, ostial narrowing of the coronary arteries nonspecific aortoarteritis, or due to aortic dissection involving the coronary ostium. Coronary arterial dissection or thrombosis may be the result of trauma. Situations where the coronary arterial system is normal but there is an imbalance between the myocardial oxygen supply and demand like anaemia, hypertrophied ventricles and thyrotoxicosis may rarely result in myocardial infarction in the young.

A less well understood yet a definite entity, is the occurrence of myocardial infarction with normal coronary arteries (Syndrome X)^{10,11}. This has been observed in all age groups, although in a greater proportion in younger subjects. Many etiological possibilities have been postulated. Some of them

already described like spasm, or myocardial bridges. Spontaneous lysis of an intra coronary thrombus may result in normal coronary angiogram.

Many studies from developed countries have focused on various risk factors in young subjects. Smoking seems to be the most common risk factor (>80% of subjects)^{2,3,4,5,6,7}. History of coronary artery disease in family members before the age of 60 years is a strong risk factor in those patients with atherosclerotic coronary disease, while it is not so with other causes^{2,3}. The prevalence of hypertension, obesity, sedantry habits, diabetes and hyperuricemia has varied considerably among various reports from western literature^{5,6,7}. Abnormalities in serum lipids have been shown to be an important risk factor in most of western subjects^{2,3,5,6,7}. Hypercholesterolemia (total cholesterol > 250 mg/dl) has been noted in 20-46% of patients with atherosclerotic coronary disease^{2,3,6,7}. 5% of young subjects with normal coronary arteries have also been shown to have serum cholesterol levels more than 250 mg/dl². Hypertriglyceridemia has been observed in about one third of these subjects⁴. Abnormalities in various lipoproteins have also been described.

Until lately there were few reports which dealt with the pattern of coronary arterial abnormalities and the risk factors in young subjects with myocardial infarction from India and developing countries. Kaul et al. have recently reported the findings of coronary angiography and risk factor prevalence in young survivors of myocardial infarction¹¹. They noted that it is

a disease of young males, majority of them having atherosclerotic coronary lesions. Smoking was the main contributory risk factor, followed by hypercholesterolemia hypertension and a positive family history. Multiple risk factors were present in less than half the total number of their patients. In another report from Bombay, smoking and diabetes were noted to be the main risks for coronary disease in young⁷.

Although the course of the disease, prognosis and management would be chiefly determined by the presence or absence and severity of atherosclerotic coronary disease, and nature of therapy ; modulation of the risk factors would help in modifying the progression or even inducing probable regression of the atherosclerotic process.

The aim of this study was to ;

1. Assess various risk factors in survivors of myocardial infarction below the age of 40 years.
2. Study the lipoprotein profiles in these patients and compare them with healthy controls in the same age group and sex.

MATERIALS AND METHODS

34 subjects who were diagnosed to have myocardial infarction by history, typical electrocardiographic criteria and serial enzyme changes and were on follow up in our institution were studied. All the subjects were males. Their ages ranged between 28 and 40 years (mean 35.5 ± 3.9 years) at the time of present

evaluation. The age at which myocardial infarction occurred ranged from 23-38 years (mean 32.5 ± 4.3 years). A detailed interrogation for risk factors was done during the present visit. Additional information regarding the clinical details, age, weight and height at the time of hospital admission were noted from the case records. The questionnaire included smoking habits (recorded as number of cigarette/day X years) past history of diabetes, hypertension, gout, activity status at work and at leisure and family history of coronary artery disease / myocardial infarction and sudden cardiac death. Height and weight at the time of present examination were recorded. Details of drug intake was noted.

Obesity was defined as a relative body weight (RBW) of ≥ 0.28 from Quetlett's formula⁶

$$RBW = \frac{Wt. \text{ in kg}}{(Ht. \text{ in Cm})^2} \times 100$$

Physical activity was evaluated to scale, of 1-4 as described by Elinfeldt et al⁶.

Scale	At Work	During leisure
I	Office x work, sitting $\frac{1}{2}$ the day,	reading, watching TV
II	Light work \bar{e} much walking throught eggs. (teacher, foreman, etc)	Walking/cycling 4 hr/wk.
III	Most of factory work/or these involving moderate activity. eg. Postman, etc.	Gardening regular jogging swimming 1-2 hr/wk
IV	Heavy manual labour	Hard regular physical training.

Blood samples were collected for lipoprotein analysis after 12-14 hours fasting over night; 8-10 ml of whole blood was kept at room temperature for 60 minutes and centrifuged at 2500 rpm. for 10 minutes to obtain the serum for chemical analysis.

Serum total cholesterol was estimated by CHOD-PAP method and Triglycerides were estimated by peridochrom - triglyceride GPO-PAP method using assay kits from Boehringer - Mannheim Gm 6H Diagnostica Germany . Colorimetric readings were taken in UV visible spectrophotometer (Beckmann) at 500 nM wavelength .

High density lipoprotein cholesterol (HDLC) was estimated by polyionic precipitation using polyethylene glycol-6000 (200g/L in 200 m Mo/L Glycine sodium hydroxide buffer at P H 10)¹².

The LDL and VLDL are precipitated and the supernatant was used for HDL - cholesterol estimation by the cholesterol estimation method described above.

Serum lipoproteins were separated by quantitative electrophoresis using polyacrylamide disc gel using previously described methods^{13,14,15}. 20 μ L of serum and 200 μ L of sample gel were prestained with Sudan Black dye and separated by electrophoresis in 3.5% polyacrylamide gel disc. Electrophoresis was run for 35 minutes at 5 mA current strength per disc (tube)^(Fig 1). After electrophoresis the gels were stored at 4^oC temperature shielded from light to avoid colour fading. The electrophoretograms were obtained by scanning using a soft laser scanning densitometer (model S1-TRS Biomed instruments California) with 610 nm interference filter. Integrated peak areas for the α (HDL-c) β (LDL-c) and pre β (VLDL-c) were obtained by/ digitised integrator incorporated in the scanning densitometer (Fig.1). From the integrated peak areas the percentage fraction of each band, was calculated and the actual values were computed from the total cholesterol (TC). For meaningful results the base line at the point with lowest absorbance between HDL and LDL fraction was set at zero.

Statistical analysis of the values were obtained for mean values of. Total cholesterol, Triglycerides HDL, LDL and VLDL cholesterol, ratio of Total cholesterol/ HDL-c and HDL-c/LDL-c for tests and male controls in the same age group using Students-T test (two tailed). A P value <0.05 was considered significant.

Abnormalities in Total cholesterol, Triglycerides, or any of the ratios were also assessed for their significance by Chi Square test.

RESULTS

All our subjects were males, thirty two patients (94%) were moderate to heavy smokers (>10 beedies/ cigarettes/day) prior to myocardial infarction. Twelve patients continued to smoke at the time of the study. A family history of coronary artery disease was present in 3 patients (8%). Three were obese prior to MI (8%) one patient each had diabetes, hypertension and gout. Physical activity scales prior to MI were 3-4 in 6 patients 17.6% rest of them were in scales 2 less. Multiple risk factors were noted in 6 patients (17.6%) and none in one patient (2.9%) (Table.1). None of the patients were on any lipid lowering drugs when studied.

Lipoprotein distribution : (Table.II) - The mean total cholesterol was 209 ± 77.37 mg/dl in patients and 198.93 ± 34.86 in controls. There was no statistically significant difference between the two groups. However, 6 patients had serum total cholesterol levels more than 250 mg/dl compared to only one in the control group.

Abnormalities in serum triglycerides were present in eight patients (23.5%) while it was seen only in one control (3.2%) ($P = <0.05$). Nevertheless, the mean triglyceride levels in both group were not statistically different. There were also no significant differences between the mean LDL and VLDL values

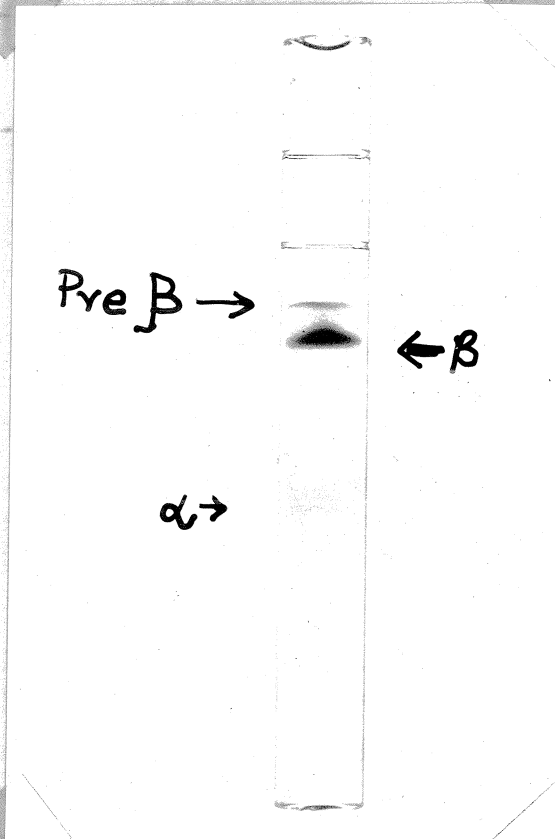
TABLE. I.

RISK FACTORS IN YOUNG MI - n = 34

	<u>n</u>	<u>%</u>
Smoking	32	94
Family History	3	8
Obesity	3	8
Diabetes	1	2.9
Hypertension	1	,,
Gout	1	,,
Sedantry Habits	28	82.4
Multiple risk factors	6	17.6
No. risk factor	1	2.9

n = Number

Fig I



Electrophoretic tube showing α , β and pre β bands.

TABLE. II.

Comparison of lipoprotein fraction in patients and controls

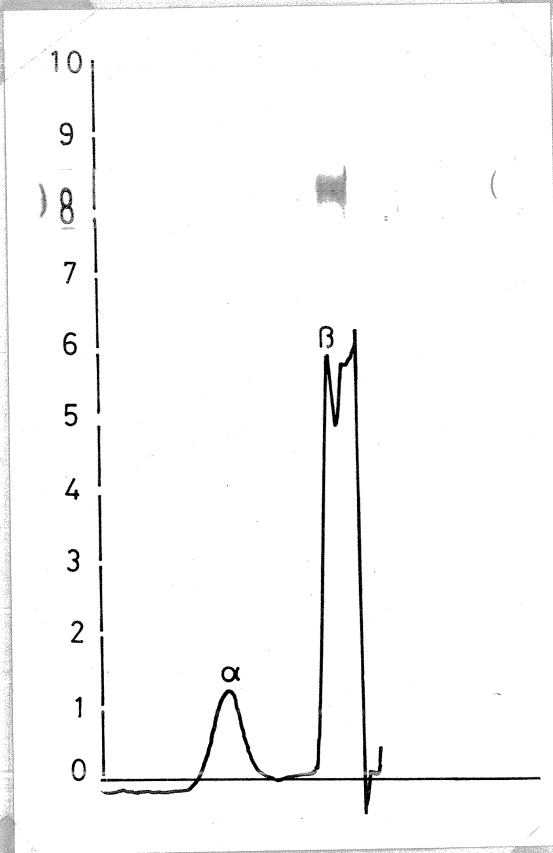
	PATIENTS	CONTROLS	P VALUE
Number	34	31	
Total Cholesterol			
>250	6* (17.6%)	1 (3.2%)	NS
Mean \pm SD	209 \pm 77.37	198.93 \pm 34.86	NS
Triglycerides			
>200	8* (23.5%)	1 (3.2%)	< 0.025 0.0128
Mean \pm SD	165.67 \pm 90.8	135.8 \pm 38.02	NS
HDLc			
Mean \pm SD	30.17 \pm 6.56	41.64 \pm 7.53	< 0.001
LDLc			
Mean \pm SD	148.2 \pm 70.67	131.54 \pm 35.58	NS
VLDLc			
Mean \pm SD	30.94 \pm 29.06	26.7 \pm 13.26	NS
TC/HDL ratio	7.05 \pm 2.24	4.87 \pm 0.97	< 0.001
HDL/LDL ratio	0.225 \pm 0.071	0.333 \pm 0.095	< 0.001

NS = Not significant ,

All absolute values for lipoproteins in mg/dl .

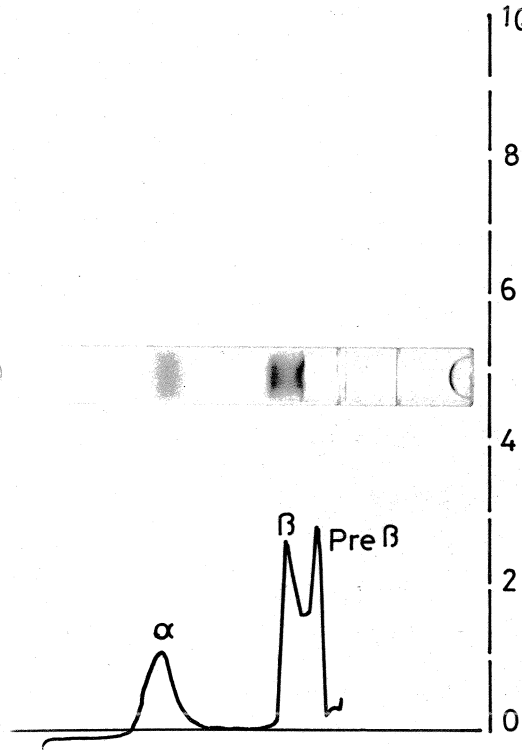
* number of subjects (% of total)

Fig II



LIPOPROTEIN ELECTROPHORETIC PATTERN IN YOUNG MI WITH LOW RATIO OF HDLc/LDLc

Fig III



ELECTROPHORETOGRAM OF CONTROL WITH NORMAL HDLc/LDLc Ratio

between patients and the controls which would incriminate them as risk factors in this study.

The serum HDL-c levels were significantly lower in the patients (mean = 30.17 ± 6.56) compared to healthy controls (mean = 41.64 ± 7.53) $P = < 0.001$. Highly significant differences were also observed when the ratios of Total Cholesterol/ HDL-c and HDL-c/LDL-c were compared between both groups ($P < 0.001$). 85% of patients belonged to the "low" HDL-c subset (ratios > 5 ie. equivalent to HDL-c/TC % of < 20) while only 45% of controls belonged to this subset ($P < 0.005$); similarly HDL-c/LDL-c ratios showed that 61.2% of controls compared to 17.6% of patients belonged to the "high" subset.

DISCUSSION

Myocardial infarction in the young is a strikingly male disease in this study. Other reports from Indian patients show a similar male predilection^{4,7,11}. Observation from western literature shows a slightly higher incidence of myocardial infarction in young females although majority of the subjects are still males^{2,3,5,6}. As has been reported in earlier works, smoking is the single most common risk factor implicated^{2,3,4,5,6,7,11}. The higher prevalence of smoking among males may explain the ~~xxxxxx~~ "male^Dness" of our patient population. Incidence of all the other risk factors have been low, a point of similarity with studies from elsewhere in Indian subjects^{4,11}.

There have been paucity of reports on abnormalities of serum lipids and lipoprotein fractions in young Indian subjects with coronary artery disease. Kaul et al have noted angiographically discernable coronary lesions in 80 of their 104 patients while hypercholesterolemia was noted only in 29 of these¹¹. Our data suggests that if one were to estimate only Total Cholesterol and Triglycerides, many more with subtle and yet significant lipid abnormalities would be overlooked. TC/HDL ratio was noted to be abnormal in about 85% of patients (compare Fig. II & III).

Prospective epidemiologic studies have established a striking independent association between HDL levels, coronary vascular events and degree of coronary arterial lesions^{26,19,20,30}. Burnner et al have recently reported that low HDL-c/TC ratio (<0.20%) is associated with higher risk of coronary events in a 20 year follow up study³⁰. Irrespective of the Total Cholesterol levels an HDL cholesterol/TC ratio of less than 21% in males would indicate a 2-4 times higher risk for future coronary events^{19,20}. The congruence of this observation gains strength when the ratio of HDL-c/LDL-c of less than 0.3 was true in most of our young subjects with myocardial infarction.

Glomset et al were the first ones to postulate that HDL-c transports cholesterol from the peripheral tissues to the liver a process involving the conversion of free cholesterol in HDL-c to cholesteryl ester by the action of enzyme lecithin cholesterol acyl transferase (LCAT)²⁴. It may thus play a role in the efflux of

cholesterol from arterial endothelium. However it remains to be established whether this reverse transport of cholesterol diminishes atherosclerosis. Animal experiments suggests that this may be possible²¹.

Alternatively Carew et al. suggested that HDL-c may bind with the LDL-c receptor on the endothelial surface thereby blocking uptake of latter by the cell²⁵. It is now known that the affinity of the LDL-c receptor is different from those that bind to HDL. This raises the possibility that an HDL subfraction may have the property of binding to the LDL receptor. For this reason recent studies have suggested that HDL-c subfractions or Apolipoprotein subclasses are better indicators of the risk for atherosclerotic coronary diseases²⁹. However Schmidt et al. while evaluating the values of various lipoprotein subfractions and lipoproteins in patients with coronary artery disease, have concluded that the ratio of HDL-c/Total Cholesterol was the only independant predictor of coronary heart disease¹⁷, thus fortifying the role for conventional lipoprotein assays as done in this study.

The scavenger role of HDL-c during intravascular lipolysis may result in the transfer of some constituents of VLDL catabolism to HDL-c. This may determine the nature of debris deposited in the endothelial macrophages, thereby governing atherogenesis²². Exercise induced lipolysis increases HDL levels in humans²².

In light of the role of HDL-c; this study has indicated that it may be an important negative contributory factor causing myocardial infarction at an earlier age. Interpreting the results of this study needs a careful appraisal of the situation. Being survivors of a myocardial infarction majority were used to a different life style with respect to dietary pattern, smoking and physical activity compared to the pre-infarction status. Naturally, one would expect these changes in the post infarction period to alter the plasma lipids towards the better. If that assumption be it, then it is likely that the results of the study gains more in importance when extrapolated to a preinfarction risk assessment.

CONCLUSION

1. Myocardial infarction prior to the age of 40 is a disease of males.
2. Smoking is the single most common risk factor.
3. Lipoprotein analysis reveals low HDL-c levels in majority of the young patients who have survived a myocardial infarction.
4. The significance of these lipid abnormalities on further coronary events in these patients needs long term studies.

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REFERENCES

1. Gupta, S.P., Kherapaul, N.K. Incidence of Acute myocardial infarction and sudden coronary death in Rohtak City.
J. Asso. Phy. India. 1981; 29 : 365 .
2. Glover. M.U., Kubes, M.T. Warren S.E., Wieweg W.V.R. Myocardial infarction before the age of 36. Risk factors and arteriographic analysis.
Am. J. Card. 1982 ; 49 : 1600 .
3. Underwood. D.A, Proudfit Lim J, McMillan. J.P. Symptomatic coronary artery disease in patients aged 21 to 30 years.
Am. J. Card. 1985 ; 55 : 631 .
4. Al Yusuf, Bhtnagar.S.K, Hashmi.J, Kolar J,.
Myocardial infarction in the young - A long term follow up study.
I'nd Heart J. - 1985; 37 (5) : 275 .
5. Nixon. J.V, Lewis.H.R, Smitherman T.C, Shapiro, W. Myocardial infarction in men in the third decade of life. Annals of Int. Med. 1976 ; 85 : 759 .
6. Bergstrand. R, Vedin. A, Wilhelmss, C. Wallin, J , Wedel H, Wilhemsen, L. Myocardial infarction in men below the age of 40. Brit. Heart. J. 1978; 40 : 783 .
7. Dolder. M.A, Oliver. M.F,. Myocardial infarction in young men, study of risk factor in 9 countries.
Brit. Heart J. 1975 ; 37 : 493 .

8. Wei. J.Y., Buckley.B.H. Myocardial infarction before 36 years of age in women, predominance of apparant non-atherosclerotic events. Am. Heart J. 1982; 104 (3): 561.
9. Savran S.V, Bryson A.L, Welch T.G. Zaret B.L., Mc Gowan R.L., Flamm M.D. jr,. Clinical correlates of coronary ciniangiography in young males with myocardial infarction. Amer. Heart J. 1976 ; 91 : 551 .
10. Khan A.H. Haywood L.J. Myocardial infarction in nine Patients with radiologically patient coronary arteries. New Eng. J. Med., 1974 ; 291 : & 427 .
11. Kaul U, Dogra B, Manchanda S.C, Wasir H.S., Rajani. M, Bhatia M.L. Myocardial infarction in young Indian patients; risk factors and coronary arteriographic profile. Am. Heart J. 1986 ; 112 : 71 .
12. Dc Macker P.N, et al. A study of the use of polyethylene glycol in estimating cholesterol in HDL.Clin Chemistry 1980, 13 : 1775 .
13. Herbert L.N, Misto Wada, Ehrnart L.A, Lewis L.A. Polyacrylamide gel-disc electrophoresis as a screening procedure for serum lipoprotein abnormalities . Clin. Chemistry 1973 ; 19 (2) : 228 .
14. Wada M, Naito H.K, Ehrhart L.A. Lewis L.A. Polyacrylamide gel block electrophoresis of plasma lipoproteins. Clin. Chemistry, 1973 ; 19 (2) : 235 .
15. Muniz N. Measurement of plasma lipoproteins by electro-phoresis on polyacfylamide gel . Clin. Chemistry. 1977 ; 23 (10) : 1826 .

16. Berge K.G, Canner P?L, Hannline A. High density lipoprotein cholesterol and prognosis after myocardial infarction. ~~xxx~~ Circulation, 1982 ; 66 (6) : 1176 .
17. Schmidt S.B, Wasserman A.G, Muesing R.A, Schlesselman SE, Larosa J.C, Ross A.M. Amer. J. Card. 1985 ; 55 : 1459.
18. Padmani.E, Motlag DB. Lipoprotein profile changes among chronic smokers and smokers with acute myocardial infarction. Ind. Heart J. 1987, 39 (1) : 38 .
19. Kannel W.B, Castelli W.P, Gordan T. Cholesterol in the prediction of atherosclerotic disease; new perspectives based on Framingham study. Ann. Int. Med. 1979 ; 90 : 85 .
20. Miller G.J, Miller N.E. Plasma HDL concentration and development of ischaemic heart disease. Lancet , 1975 ; 1 : 16 .
21. Wesselino witch D, Wissler R.H, Huges R, Borensztajn.T. Reversal of advanced atherosclerosis in rhesus monkeys, light microscopic studies. Atherosclerosis, 1976 ; 23: 155
22. Levy RI, Ref kind BM ; Structure, function and metabolism of high density lipoprotein; A status report . Circulation; 1980 , 62 Suppl IV : 4 .
23. Gordon T, Castelli W.P, H or hand M.C, Kannel W.D, Dawber T.R. High density lipoprotein as a protective factor against coronary heart disease. Am. J. Med. 1977 ; 62 : 707 .
24. Glomset J.A. The plasma lecithin , cholesterol acyl transferase reactions. J Lipid Res. 1968; 9 : 155 .

25. Carew T.E, Koschinsky T, Heys S.B. Steinberg D.
A mechanism by which HDL may slow the atherogenic process. Lancet , 1976 ; 1 : 1315 .
26. Davis CE, Gordan D. La Rosa J, Wood P.D.S, Halperin M.
Correlations of plasma HDL-c levels with other plasma lipid and lipoprotein concentrations .
Circulation, 1980 ; 62 (Suppl IV) .
27. Kristensen B.O,. Effect of long term treatment with betablocking drugs on plasma lipids a lipoproteins.
Br. Med. J. (Clin Res), 1981; 283 : 191.
28. Taskai Ital, Lipid profile in patients with myocardial infarction from the city of Bombay.
Ind. Heart. J. 1983 ; 35 (3) : 169.
29. Mahley R.W, Innerarity, T.L. Bersot T.P. Lipson A, Margolis, S. Alterations in human HDL with or without increased plasma cholesterol induced by diet high in cholesterol. Lancet, 1978 ; 2 : 807.
30. Brunner D. et al. Relation of serum total cholesterol and high density lipoprotein cholesterol percentage to the incidence of definite coronary events: Twenty years follow up of the Donolo- Tel Aviv prospective coronary artery disease study.
Am. J. Card., 1987 ; 59 : 1271 .

LIPOPROTEIN DATA OF PATIENTS

Sl. No.	Age	Total Chol	Triglyceride	HDL	LDL	VLDL
1.	31	148	71	22	126	--
2.	33	163	92	29	134	--
3.	39	215	103	30	147	38
4.	31	156	90	26	112	18
5.	39	332	162	33	299	--
6.	34	218	158	40	149	39
7.	30	334	339	40	212	82
8.	39	260	102	39	221	--
9.	40	171	211	22	92	57
10.	34	185	176	25	160	--
11.	32	315	320	28	230	57
12.	40	254	273	26	105	123
13.	32	128	142	23	75	30
14.	40	217	435	28	92	97
15.	39	187	70	34	153	--
16.	38	160	88	32	102	26
17.	40	535	183	38	457	40
18.	34	219	197	19	153	47
19.	38	196	145	44	121	31
20.	38	212	250	26	132	54
21.	38	150	115	28	123	29
22.	28	156	76	39	117	--
23.	31	212	105	33	159	20
24.	28	156	215	27	103	26
25.	36	126	159	20	106 (Broad β type)	
26.	28	180	85	38	132	10
27.	40	214	174	28	147	39
28.	35	154	351	23	109	23
29.	37	210	142	25	148	37
30.	39	224	173	29	150	45
31.	39	162	100	41	101	20
32.	36	195	163	30	123	42
33.	36	191	125	29	140	22
34.	37	141	43	32	109	--

nb: all values in mg/dl

LIPOPROTEIN DATA OF CONTROLS

Sl. No.	Age	Total Chol.	Triglyceride	HDL	LDL	VLDL
1.	36	231	98	46	173	12
2.	31	246	122	48	192	6
3.	33	169	105	48	103	18
4.	36	230	136	58	144	28
5.	27	245	129	34	181	29
6.	31	260	106	52	198	10
7.	35	155	104	38	117	29
8.	31	169	148	27	101	47
9.	36	175	121	36	95	14
10.	36	161	109	33	97	31
11.	33	171	188	45	90	36
12.	33	136	70	49	81	6
13.	32	204	175	40	134	30
14.	34	151	129	28	90	24
15.	31	225	166	36	178	11
16.	33	203	177	41	124	38
17.	39	184	244	44	89	51
18.	31	246	122	48	192	6
19.	38	197	140	50	118	29
20.	34	174	174	35	93	46
21.	29	177	142	38	117	22
22.	38	222	167	41	141	40
23.	39	247	167	40	156	51
24.	34	171	125	33	99	39
25.	32	146	79	44	142	--
26.	32	235	162	44	168	23
27.	40	211	141	53	119	39
28.	38	163	90	42	110	11
29.	37	201	105	44	129	28
30.	35	242	189	46	173	23
31.	38	190	172	30	134	27

nb: all values in mg/dl.