LIPID STUDIES IN PATIENTS WITH ADVANCED DIABETIC ATHEROSCLEROSIS *

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In recent years, interest in the problem of human atherosclerosis has been great. Since the early observations on atheromatous lesions in cholesterol-fed rabbits,[1, 2] there have been many studies on the possible rôle of elevated blood cholesterol in the genesis of atherosclerosis. The work of Gofman and his co-workers [3] has been of interest in this connection through their demonstration of a technic for measuring cholesterol-bearing lipoproteins in serum. The group of molecules known as the Sf_{10-20} group, according to Svedberg's designation of flotation densities, has been reported to be elevated in individuals with coronary artery disease and is thought by the above investigators to be directly concerned in the atherosclerotic process. Elevation of other serum lipid fractions in atherosclerotic patients has been a controversial subject, as has also the effect of "lipotropic substances" on these lipids.[4-10]

The present study was designed to determine the levels of a number of lipid entities in the blood of a group of patients with advanced atherosclerosis in association with diabetes, and to determine the effects of certain pharmacologic agents upon these lipids.

Procedure

The patients selected consisted of 24 elderly hospitalized diabetics (seven men and 17 women with diabetes of varying duration), several of whom had had leg amputations for diabetic gangrene, myocardial infarctions and cerebral accidents (table 1).

Patients were maintained on relatively constant "diabetic diets." After two control specimens on consecutive days, bloods were drawn in the fasting state at weekly intervals while the patients were on one of three medication programs. The serum was analyzed for free and total cholesterol, for lipid-phosphorus and for lipoprotein (Sf_{10-20}). Insulin requirement and average semiquantitative urinary glucose excretion were recorded. Medications

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TABLE I

Patient	Sex	Age	Insulin	Initial Cholesterol	Initial Lipoprotein	Dorsalis Pedis Pulsation	Amputation	Current Gangrene	Known Myocardial Infarction	CVA
P. A.	M	62	No	211.5 ±0.24%	72.0 ±9.72%	0	1 accidental	0	0	0
E. C.	F	86	Yes	222.5 ±1.12%	77.5 ±13.55%	0	2	0	0	0
L. F.	F	62	Yes	144.5 ±0.35%	55.0 ±14.55%	2	0	0	0	0
M. G.	F	82	Yes	208.5 ±0.72%	60.0 ±13.33%	0	0	0	0	Yes
C. L.	F	73	Yes	272.5 ±2.75%	176 (one only)	0	0	0	0	Yes
G. M.	F	82	Yes	267.0 ±5.99%	95.0 ±5.26%	0	0	0	0	0
R. P.	F	70	Yes	134.0 ±2.99%	33.5 ±1.49%	0	0	0	Yes	0
T. V.	M	71	Yes	207.0 ±3.86%	97.0 ±11.34%	0	1 osteo	0	0	0
J. B.	M	71	No	202.0 ±0.99%	29.0 ±0%	2	0	0	0	Yes
J. Co.	F	81	Yes	226.5 ±1.55%	43.0 ±20.93%	0	0	0	0	0
P. F.	F	75	Yes	183.0 ±2.73%	57.0 ±17.54%	2	0	0	0	Yes
A. F.	M	72	Yes	259.5 ±0.58%	92.5 ±4.86%	1	0	0	0	0
J. L.	F	60	Yes	232.5 ±5.38%	66.0 ±1.52%	0	1	0	0	0
T. N.	F	81	No	214.5 ±3.03%	42.5 ±24.71%	2	0	0	0	0
M. S.	M	69	Yes	172.5 ±0.29%	29.0 ±10.34%	0	1 later	Yes	0	0
E. S.	F	72	No	218.5 ±1.60%	33.5 ±31.34%	0	2	0	0	0
J. Ca.	M	69	Yes	176 (one only)	43 (one only)	0	2	0	0	0
M. D.	F	68	Yes	271.5 ±2.03%	142.5 ±15.79%	1	0	0	0	0
A. G.	F	71	Yes	268.0 ±1.12%	125.0 ±2.40%	0	0	Yes	0	0
J. M.	F	61	Yes	224.0 ±2.68%	81.0 ±3.70%	2	0	0	0	Yes
A. M.	F	60	Yes	220.5 ±4.31%	84.5 ±1.78%	2	0	0	0	Yes
P. M.	M	84	Yes	193.0 ±1.04%	44.0 ±18.18%	0	2	0	0	0
V. P.	F	80	Yes	190.0 ±4.74%	45.5 ±16.48%	2	0	0	0	Yes
M. P.	F	88	No	198.5 ±1.76%	60.0 ±1.67%	0	0	0	0	0

administered were choline, inositol and testosterone propionate, respectively, because of their known or supposed effects on serum lipids and/or on the atherosclerotic process.

Eight patients received 6 gm. of choline citrate daily for 31 days. Eight patients received 9 gm. of inositol daily for 27 days, and eight patients received 25 mg. of testosterone propionate daily intramuscularly, for varying periods.

METHODS

Cholesterol was determined by the method of Michaels et al.,[11] in which the cholesterol digitonide is determined turbidimetrically. The normal range is essentially the same by this method as that reported by Schoenheimer and Sperry.[12] Lipid phosphorus was determined by the method of Youngburg and Youngburg.[13] Lipoprotein Sf_{10-20} was determined by an ultracentrifugation technic.*

RESULTS

A study of the effects of the three medications on these blood constituents failed to reveal consistent alterations which could be ascribed to the medications in the case of choline and inositol. In the case of testosterone, three patients appeared to have a decrease in the levels of cholesterol and lipoprotein during therapy (figure 1). Consistent changes in the diabetic state were not observed during or following the periods of treatment. A decrease in insulin requirement, however, was noted in four patients on testosterone.

* Grateful acknowledgment is made to Dr. John Gofman for performance of these determinations.

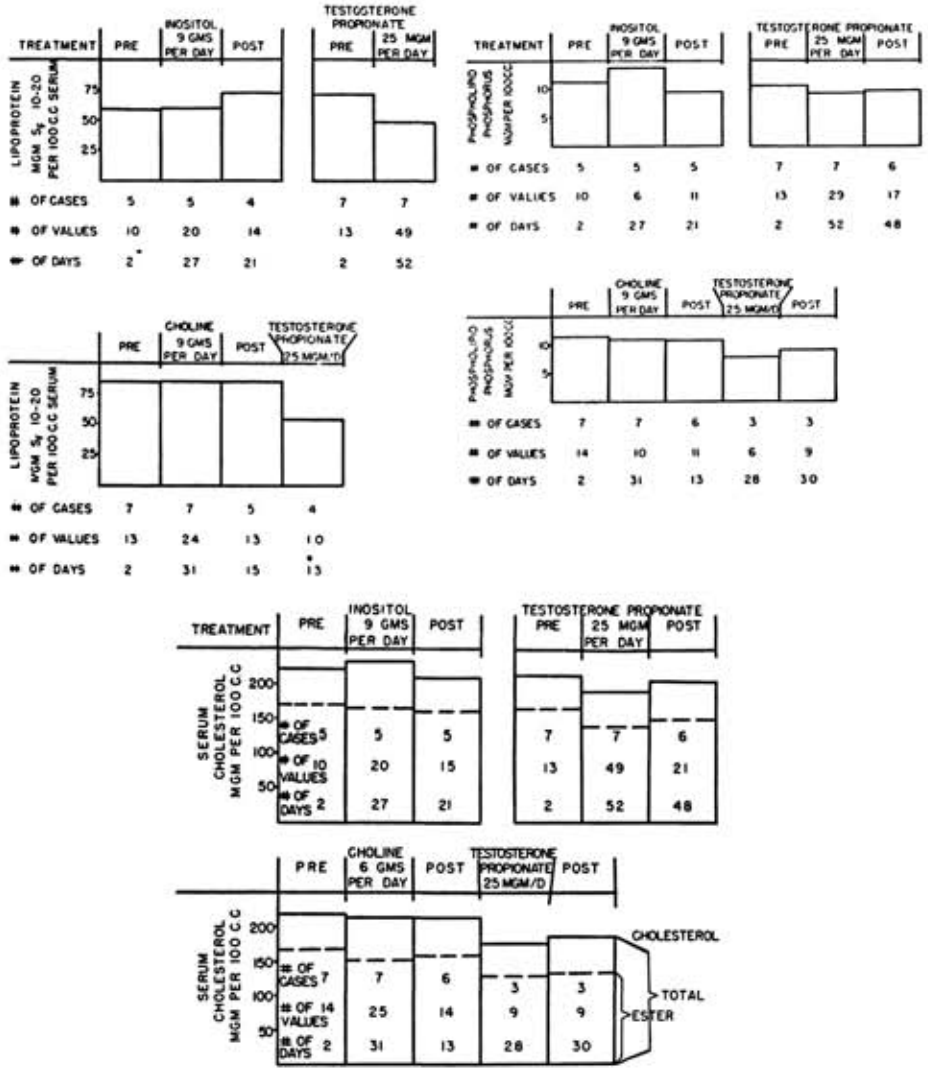


FIG. 1. Examination of the data suggests that inositol and choline had no effect on any of the lipid entities, but that testosterone propionate may have decreased the serum concentration of all of the lipid fractions studied. Further work will be required before one will know whether these observations are statistically significant.

Taking the group as a whole, a considerable spread in the values of the various lipid components was observed (table 2). This spread was particularly great in the case of the lipoproteins, with a coefficient of variation equal to 48 per cent, compared to coefficients of 20 per cent for total cholesterol, 21 per cent for cholesterol esters and 22 per cent for lipid phosphorus. The difference between two daily control values in individual patients was ± 10.93 per cent in the case of lipoprotein and ± 2.25 per cent in the case of total cholesterol. The mean cholesterol level was 203.1 mg. per cent in

the seven males and 217.4 mg. per cent in the 17 females. In a control group of hospitalized elderly patients, none of whom had had myocardial infarctions, strokes, amputations or diabetes, the mean level of cholesterol was found to be 169.5 mg. per cent in 10 males and 158 mg. per cent in 13 females.

TABLE II

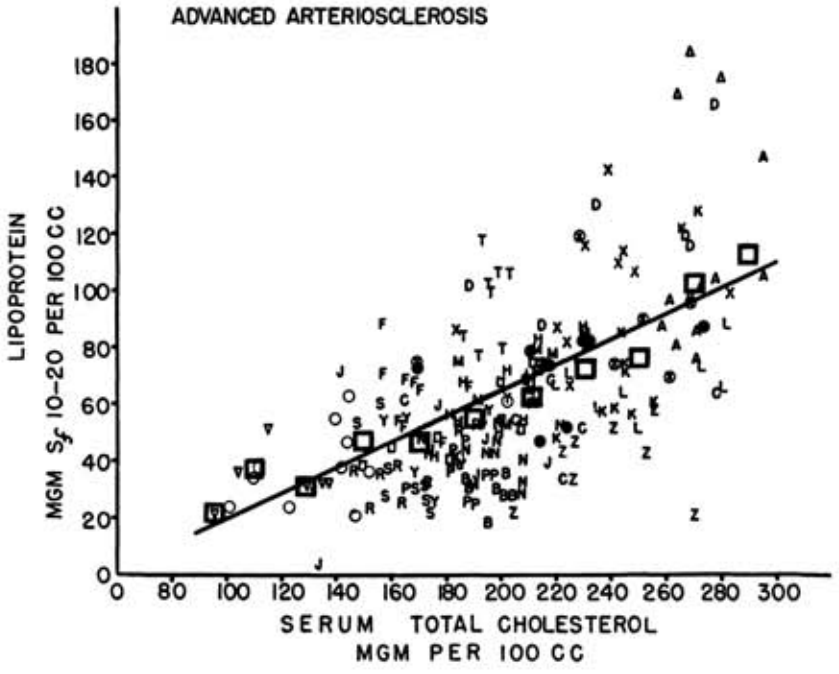
Values of Blood Lipid Components in Elderly Diabetics

	Number of Determinations	Mean	S. D.
Total Group			
Cholesterol—Total	208	202.8	41.1
Esters	206	148.6	31.0
Lipid phosphorus	166	10.09	2.18
Lipoprotein	208	63.0	30.5
Choline			
Cholesterol—Total	39	206.1	45.2
Esters	39	149.3	33.8
Lipid phosphorus	34	10.56	2.24
Lipoprotein	39	86.8	37.4
Inositol			
Cholesterol—Total	70	212.0	40.1
Esters	69	156.1	31.0
Lipid phosphorus	47	10.66	2.37
Lipoprotein	70	56.0	24.4
Testosterone Propionate			
Cholesterol—Total	71	199.5	32.2
Esters	70	146.0	24.8
Lipid phosphorus	61	9.72	1.73
Lipoprotein	71	58.7	27.4

An analysis of our data reveals a significant correlation between the levels of many of the lipid components studied (figure 2). The correlation coefficient between total cholesterol and lipoprotein is 0.613; between esterified cholesterol and lipoprotein, 0.530; between lipid phosphorus and lipoprotein, 0.453, and between esterified cholesterol and lipid phosphorus, 0.671.

COMMENT

Under the conditions of this study, it appears that large doses of inositol and choline produced little or no alteration in the diabetic state or the levels of blood lipid components in these patients. The interpretation of the fall in lipid levels during and for a short period after testosterone therapy in some patients is not apparent at the present time. Further studies are in progress.



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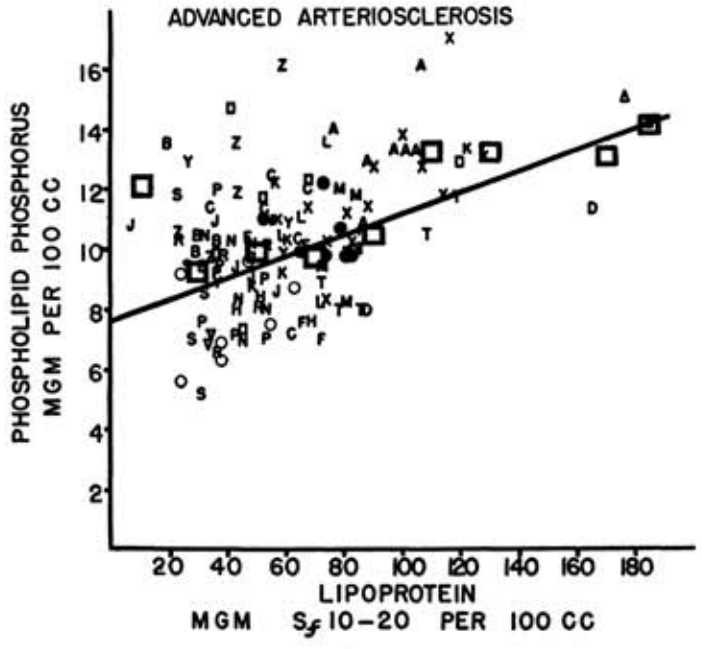

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FIG. 2.

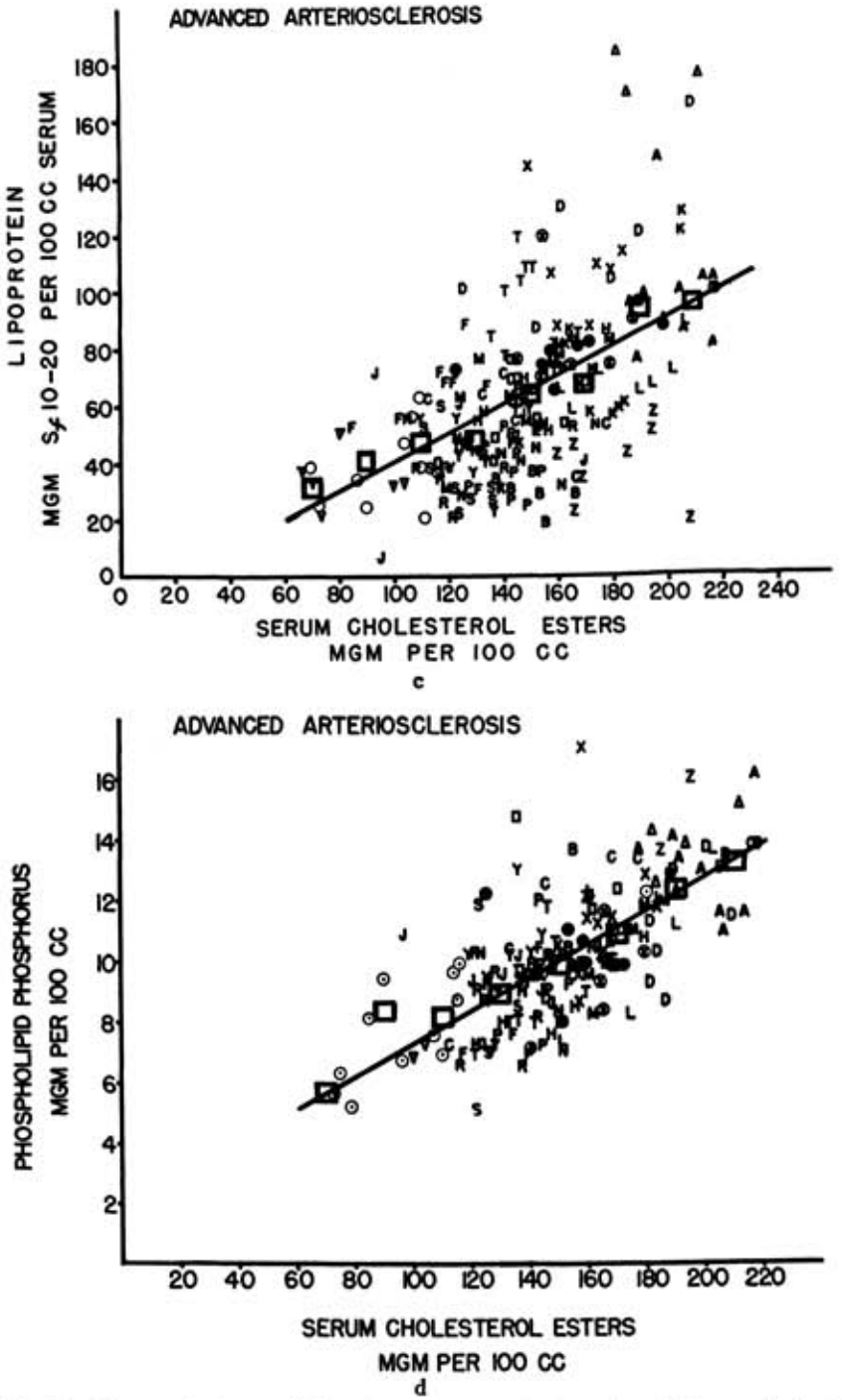

FIG. 2. The correlation coefficient between total cholesterol and lipoprotein is 0.613; between esterified cholesterol and lipoprotein, 0.531; between lipid phosphorus and lipoprotein, 0.453, and between esterified cholesterol and lipid phosphorus, 0.671. The first and last figures have high statistical significance in terms of "linear correlation" of the respective entities.

The same statement applies to the changes in the diabetic state observed in an occasional patient receiving testosterone.

The data indicate that when cholesterol levels are determined by a reliable method, a highly significant degree of correlation is found between cholesterol and its esterified fraction on the one hand, and lipoproteins of the Sf_{10-20} group on the other. This contrasts with the conclusion of Gofman et al.[3] that only a very general correlation exists, and of Jones et al.[14] that no correlation exists. Re-analysis of Gofman's own data by Keys revealed a highly significant correlation between blood cholesterol and lipoprotein levels.[15] It appears, then, that the measurement of total cholesterol gives essentially the same information in patients with atherosclerosis as does the measurement of lipoproteins, Sf_{10-20}.

It can be seen from table 1 that the patients with the highest levels of cholesterol or of lipoprotein are not always the ones with the greatest clinical evidence of atherosclerosis. Indeed, it is interesting to note that the patient (R. P.), with the lowest level of cholesterol and one of the lowest levels of lipoprotein, died during the course of the study as the result of a myocardial infarction (proved at autopsy). It is not obvious that any of the lipid components studied can be correlated with the presence or degree of atherosclerosis in any individual patient. Whether the lower mean blood cholesterol levels in the group of elderly nondiabetic "controls" is related to the absence of the diabetic state, or to some other factor, is unknown at this time.

Summary and Conclusions

A group of 24 diabetic patients with advanced atherosclerosis, several of them having lost extremities as the result of atherosclerotic involvement, was studied from the following standpoints:

1. To evaluate the effect of various "lipotropic agents" upon:
   a. Their diabetes as evidenced by insulin requirement.
   b. The levels of certain blood lipid components.
2. To determine whether any correlation existed between certain lipid components of the blood, and between these lipids and the atherosclerotic process.

In this group, the following observations were made:

1. Serum total cholesterol, lipoprotein, cholesterol ester and phospholipid values varied over a wide range.
2. No obvious correlation between any of the lipid fractions and the atherosclerotic process was found, although the mean serum cholesterol values were significantly higher than in a group of nondiabetic elderly "controls."

3. Linear mathematical correlation was noted between the lipoprotein and total cholesterol, lipoprotein and cholesterol esters, and cholesterol esters and serum phospholipids.

4. Under the conditions of this study, no significant changes in blood lipids were observed in patients receiving relatively large amounts of inositol or choline. No significant change in the diabetic state was observed in response to the same agents.

5. Depression of serum cholesterol esters, of serum phospholipids and of serum lipoprotein was observed in several patients receiving testosterone propionate in a dose of 25 mg. daily. Some changes in insulin requirement were also noted. These changes were not constant.

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