It is suggested that factors other than child-bearing play an important part in promoting the weight difference between married and single women.

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THE BETA-LIPOPROTEIN AND CHOLESTEROL CONCENTRATIONS IN SERA OF NIGERIANS*

BY

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The relation of diet to the development of atherosclerosis can be studied in contrasting cultural groups which have different dietary habits. This appears to be one approach to a study of the late effects of diet upon this form of chronic disease.

In a previous study of Central American subjects (Mann, Munoz, and Scrimshaw, in press) we have demonstrated that there are striking differences in the serum lipid patterns among populations with different environmental characteristics. It was shown that the serum cholesterol levels of adults of either sex averaged less than 175 mg. per 100 ml. at all ages, that these levels were not significantly related to age, and that, while the rural Central Americans were thin as judged by relative weight ($\frac{\text{observed weight}}{\text{desirable weight}}$), this thinness could not account for the low serum cholesterol levels observed. In contrast to these findings with cholesterol, the serum beta-lipoprotein levels of the rural Central Americans were similar to those of age- and sex-matched North Americans. There was thus a unique disparity between the lipid patterns of the largely vegetarian Central Americans and the North American comparison group. A comparison group of urban Guatemalans were similar in both dietary and serum lipid patterns to North Americans. These observations promise to illuminate the problem of the relative atherogenicity of the various serum lipid components. They require further study to determine the mechanisms which control these various serum lipid components.

* A portion of this work was completed while Dr. G. V. Mann was an Established Investigator of the American Heart Association.

The studies reported here were undertaken at about the same time to extend these observations to another racial group in an entirely different environment in order to verify the findings described, and to minimize the possibility that genetic factors might be responsible. The serum cholesterol and lipoprotein levels of 46 Nigerian men have been studied, and dietary and cultural evaluations have been made which permit segregation of these into three groups. The resulting data resemble those obtained in Central America, and confirm the belief that primitive peoples, consuming largely vegetarian diets, with a high calorie turnover in relation to body size, show low levels of serum cholesterol coupled with beta-lipoprotein levels similar to those of urban United States citizens.

Methods

Nature of the Sample

The Nigerian subjects were drawn from three different areas of the country. The cultural characteristics and dietary patterns differ considerably between these three areas. Group A consisted of natives of the dry northern part of Nigeria, coming from Moslem tribes whose staple foods are sorghum and millet (*Pennisetum* spp.), who eat little fish or meat, and whose diet contains little fat apart from that present in the vegetables consumed. Group B was composed of natives of the Ibo tribe from the humid Eastern Region, whose staples are yams (*Dioscorea* spp.) and cassava (*Manihot utilissima*), which they supplement with reasonable quantities of green and leafy vegetables, fruit, and a little fish or meat. Their diet contains red palm oil, but the total fat content is low compared with that of North Americans. Group C was drawn from pagan tribes of the Middle Belt of Nigeria, that area which lies along the banks of the rivers Niger and Benue in their westward and eastward passage through the country. These people eat both roots and grain, a little fish, and red palm oil.

The wealthier members of these tribes were excluded from this investigation, and the three groups consisted of peasant farmers and the labouring classes. Such peasants, at all ages, expend large amounts of energy from their limited calorie resources to maintain themselves at a subsistence level, and, in contrast to that of North Americans, their weight decreases with age after the fourth decade (Nicol, 1952).

Apart from a clinical impression that the acute complications of atherosclerosis are rarely seen in these Nigerian peasants, little is known of the incidence of this disease from necropsy material studied in Nigeria. However, Edington (1954) has studied 3,645 reports of necropsies carried out in the Gold Coast, where the cultures are somewhat similar to those of Nigeria, and has recorded a low incidence of hypertension (0.2%), atheroma (0.5%), and coronary disease (0.8%).

Clinical Examinations*

The subjects in all three groups were examined in Kaduna General Hospital while they were convalescent from minor surgical procedures. A general clinical examination was performed, heights, weights, and blood pressures were recorded, and individuals suffering from gross infestations or active disease were excluded. Signs of cardiovascular disease were not observed clinically, and in all but one individual, whose blood pressure was 140/90, the systolic and diastolic tensions were low.

Dietary Methods

The diets of the subjects forming the sample were not measured individually. When in hospital they ate food brought in for them by relatives living in Kaduna, this being the normal custom in Nigeria. Such is the conservatism of these people regarding dietary patterns that their food intake, in whatever part of the country they happen

to be living in, differs little from that typical of their native tribe in its own environment. The characteristics of these typical tribal diets have been established by studies carried out over the past eight years, and are shown in Table I. The methods employed and the data obtained have been fully documented elsewhere (Nicol, 1949, 1952, 1953), and

TABLE I.—*Dietary Characteristics of Three Nigerian Tribes Studied*

Group:	A	B	C (Middle Belt)
Culture	Moslem	Ibo	Pagan
Staples	Sorghum, millet	Yam, cassava	Sorghum, yam
Alcohol	Very little	Palm wine	Sorghum beer, palm wine
Calories	2,800	2,350	2,500
Protein—animal ..	3 g.	20 g.	6 g.
" plant	90 "	26 "	70 "
" total	93 "	46 "	76 "
Fat	26 "	45 "	45 "
" % of calories	8	15	15

confirmed in more recent studies as yet unpublished (Nicol). The nutrient composition of the diets was computed from Platt's (1945) "Tables of Representative Values of Foodstuffs commonly used in Tropical Countries," additional information being obtained from the United States Department of Agriculture's "Composition of Foods used in Far Eastern Countries" (Leung, Pecot, and Watt, 1952) and from the Food and Agriculture Organization's "Food Composition Tables for International Use" (Chatfield, 1949).

Biochemical Methods

All blood samples were taken in the early morning before the first meal, and the sera were separated the same day in the laboratories of the West African Institute for Trypanosomiasis Research. The sera were stored in a refrigerator overnight and dispatched by air the next day in a large flask of ice. The ice was replenished at Lagos and Accra (Gold Coast) when necessary. The sera reached the laboratories in Boston within five days of being separated, having been kept at 0–5° C. while in transit.

The analysis of beta-lipoproteins was done by the ultracentrifugal method of Gofman (Gofman *et al.*, 1950). The total cholesterol was measured by the method of Abell (Abell *et al.*, 1952).

Statistical Methods

The comparison of means of the three Nigerian subgroups was done by conventional "t" tests. For this purpose the subjects were not considered according to age. The comparison of the Nigerian subjects with North American data (Mann, Lawry, O'Connell, Wysocki, and Stare, in preparation) was done by a matched series method in which a Nigerian subject was randomly matched with one or more North American subjects according to age and relative weight. It was possible to match 30 of the Nigerian subjects by this procedure. While the data of 81 North American subjects were used to supply means of those who matched, these means were taken as contributing one estimate each, so that each comparison group contributed 29 degrees of freedom.

Results

A description of the cultural characteristics and diets of the Nigerian subjects is shown in Table I. Those diets

are characterized by their low calorie content in relation to the physical activities required, the low level of animal protein in all groups, the low total protein level of Group B, and, finally, the low fat content of all the diets. The groups differ among themselves principally in the lower protein content of Group B.

The summaries of the serum lipid measurements of the Nigerian subjects are shown in Table II as means and standard deviations. When the differences of these were examined, only the difference of the S_F12–20 level between Group A and B was found to exceed a probability of 5%, that the observed level of Group B exceeded that of Group A by more than would be expected from sampling error.

The similarity of the three subgroups in respect to age, relative weight, and diet, as well as serum lipid levels, seemed to justify their pooling for the matched series comparison with North American subjects, which is summarized in Table III. This comparison revealed a highly significant

TABLE III.—*A Comparison of the Serum Lipid Levels of 30 Nigerian Men with those of an Age- and Weight-matched Group of United States Men*

	S _F 12–20 (mg./100 ml.)		S _F 20–100 (mg./100 ml.)		Total Cholesterol (mg./100 ml.)	
	$\bar{x}$	±S.D.	$\bar{x}$	±S.D.	$\bar{x}$	±S.D.
Nigerian	25.4	±14.5	40.1	±29.8	129.9	±43.1
North American ..	33.6	±17.0	58.3	±45.5	212.6	±42.8
t	2.01		1.83		7.39	
Degrees of freedom ..	58		58		57	
P	0.05		>0.05		>10 ^{–12}	

difference of serum cholesterol levels. The Nigerian subjects showed levels averaging 83 mg. per 100 ml. lower than the North American comparison group. The S_F12–20 levels of the North Americans were higher by an average of 9 mg. per 100 ml., and this difference had a probability of 0.05, but the observed S_F 20–100 difference was not significant (P > 0.05).

Discussion

These data confirm the observations previously made in Central American subjects. They indicate that some environmental factors are at work which prevent the rise of serum cholesterol levels with age which is characteristic of North Americans. The Nigerian subgroups did not appear to differ significantly among themselves.

There are at least three explanations for these phenomena which are suggested by the data. The differences of serum lipid patterns among the comparison groups might result from (a) differences of fat intake, (b) differences of physical activity, (c) differences of protein intake.

The differences of fat intake among the groups represent an attractive explanation, and one which has been exhaustively documented by Keys (1953) and Keys and Keys (1954). There are certain difficulties with this explanation. Why and how does the lower fat intake of the vegetarian groups selectively affect the serum cholesterol concentration without influencing the beta-lipoprotein levels? Why are dietary restrictions of fat in North American subjects not more effective in reducing the serum lipid levels (Keys, 1952), and why, when reduced to the low levels of the Central American and African groups studied here, are they

TABLE II.—*Serum Cholesterol and Beta-lipoprotein Concentrations of 46 Nigerian Men, Grouped According to Cultural Origins*

Group	Age (Years)		No. of Subjects	Relative Weight		S _F 12–20 (mg./100 ml.)		S _F 20–100 (mg./100 ml.)		Total Cholesterol (mg./100 ml.)	
	$\bar{x}$	±S.D.		$\bar{x}$	±S.D.	$\bar{x}$	±S.D.	$\bar{x}$	±S.D.	$\bar{x}$	±S.D.
A	28.8	±8.8	23	0.87	±0.09	21.1	±11.5	36.7	±26.9	118.8	±43.9
B	29.2	±11.9	14	0.91	±0.12	30.2*	±13.5	44.9	±35.2	129.9	±40.9
C	29.9	±13.2	9	0.91	±0.04	22.3	±11.0	43.8	±21.5	119.9	±33.7
Combined groups ..	29.1	±10.5	46	0.89	±0.10	24.1	±12.5	40.6	±28.5	122.5	±40.6

*P < 0.05 in AB comparison.

often found to increase the serum cholesterol levels (Hatch and Kendall, 1952)? We are inclined to doubt that the serum lipid differences observed in the present studies are caused by low fat intakes.

We have proposed in another study (Mann, Teel, Hayes, McNally, and Bruno, submitted for publication) that muscular exercise may exert an effect in reducing or controlling the serum lipid levels in a manner analogous to the favourable effect of exercise on carbohydrate tolerance in diabetic subjects. This hypothesis would attribute the low cholesterol levels found in Guatemalans and Africans not to the limited diet but to the muscular activity and energy expenditure required in those cultures. This proposes that a large muscle mass or a large muscular expenditure is the effective agent, and that this, not the diet, is the important element in preventing hyperlipaemia and perhaps atherosclerosis. Reflection will show that such an explanation is at least as compatible with the available observations as is the dietary fat explanation which has been so widely advocated. When this effect of exercise was examined experimentally in human subjects it was found that doubling the calorie intake of young men who were consuming a very high fat diet did not increase the serum cholesterol or lipoprotein levels so long as the excess energy was expended in physical activity. There was some evidence that physical training actually diminished the serum lipid levels in the presence of large food intakes, and that a large muscle mass tended to delay the hyperlipaemia which results from periods of positive calorie balance (Walker *et al.*, 1953). The same experiment did not confirm the hypothesis that the level of serum beta-lipoprotein is directly correlated with the magnitude of the energy exchange.

There is no evidence in human subjects that the protein intake influences the regulation of the serum lipid levels. We have shown in cebus monkeys (Mann *et al.*, 1953) that a mild deficiency of cysteine and its precursors is necessary for the production of hyperlipaemia and atherosclerosis in that species by cholesterol feeding. The association of low dietary protein intakes with low serum lipid levels, and the dissociation of cholesterol and beta-lipoprotein levels in the human subjects studied here, all detract from the possibility that these dietary situations produce the same results in the two species.

Summary

It has been shown that the serum cholesterol levels of 46 Nigerian men were significantly lower than the levels of an age- and weight-matched group of United States citizens. The serum lipoprotein levels of the Nigerians were only slightly lower than those of the North Americans, and these differences were of doubtful significance.

The Nigerian subjects were found to consume diets of low protein and fat content.

The relation of these dietary characteristics to the serum lipid findings has been discussed.

The data are interpreted as supporting a hypothesis which relates muscle mass and the magnitude of energy expenditure to the control of the serum lipid levels.

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TRUE ENTEROLITHS OF THE SMALL INTESTINE

WITH REPORTS OF THREE CASES

BY

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True enteroliths of the small intestine are of three main types: (1) those consisting mainly of bile acids; (2) those consisting mainly of calcium oxalate; and (3) those consisting mainly of phosphate. Bile acid enteroliths are formed mainly of cholic acid; a small proportion of cholic acid may also be present. Two such stones, formed in duodenal diverticula, were analysed by me a few years ago (Fowweather, 1949), and the clinical features of the case were also described (Armitage, Fowweather, and Johnstone, 1950). Three cases involving the other types of stone have since come to my notice.

Case 1

The first case was that of a woman of 65 who was first seen by Dr. A. Leese, who diagnosed the presence of enteroliths and referred her to Mr. George Armitage for operation. It was then found that a portion of small intestine just proximal to the ileo-caecal valve was the seat of considerable changes. These consisted mainly of a series of constrictions, eight in all, with stones lying in pouches above some of the dilatations. To indicate the position of the stones the constrictions were numbered 1 to 8, from the ileo-caecal valve upwards.

Stones Marked "Above 8."—Two stones were present, both dark brown in colour and oval in shape. (1) Weight 10 g.; length 30 mm. In the centre was a plum stone weighing 0.6 g. The material surrounding it was dark in colour and laminated. There was evidence of loss of part of the outer layer. When broken, the fractured surface was lighter in colour than the unbroken surface, and when powdered was bright orange. (2) Weight 7 g.; length 28 mm. In the centre was a plum stone weighing 0.5 g. The surrounding material was very similar to that of (1) and also showed some loss from the outer layer.

Stones Marked "7 and 8"—Two stones were found between constrictions 7 and 8. (1) This was a plum stone weighing 1 g. and 22 mm. in length. The surface was shiny and smooth as if worn. (2) This was also a plum stone, covered with a thin, hard, yellow-brown coating. Total weight 1.4 g.; length 24 mm. The plum stone itself weighed 1 g.

Stone Marked "5 and 6."—This was of irregular shape (about 12 mm. in diameter) and weighed 0.8 g. It had the same brownish colour as those already mentioned, but no foreign body could be recognized within it. It is conceivable that a portion of the outer layer lost from the first or second stone referred to may have acted as the nucleus for deposition in this case.