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POPULATION STUDY ON BLOOD COAGULATION AND SERUM LIPIDS

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Two population groups differing in dietary habits and physical activity were examined on blood lipids and blood coagulability. The results showed a higher lipid concentration and shorter clotting time in the physically less active group, having a higher fat and caloric intake.

A great interest is shown in recent times to survey the field of cholesterol metabolism and the possible relationship of its derangement to the patogenesis of atherosclerosis and coronary heart disease. There is no more doubt that diet, particularly fat has a potent influence on the blood cholesterol and lipoproteins and there is more and more evidence that fats in the diet influence the coagulability of blood. Although the basic causes of atherosclerosis and thrombosis still remain obscure, there is a good deal of evidence from epidemiological surveys and clinical observations indicating that whenever the concentration of cholesterol and cholesterol bearing lipoproteins in the blood is maintained at a high level, there is a marked tendency toward the development of atherosclerosis and coronary heart disease.

In the paper presented at the Conference on atherosclerosis and coronary heart disease which took place in New-York in January 1956, Prof. A. Keys concluded among others that both the serum cholesterol concentration and the frequency of the ishaemic heart disease are closely correlated with the proportion of fats in the habitual diet, and that the influence of dietary fat on the frequency of coronary heart disease probably operates through the effects on both atherosclerosis and thrombosis.

In a previous study organized by Prof. Keys in 1954 in Naples, a statistically significant difference was found in the serum cholesterol

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level between 46 men of the wealthy class and 356 heavy workers (1). This difference corresponded to the proportion of fat in their diet. In this paper we present the preliminary data of a further study which was carried out in Naples in spring 1957 and in which besides the blood lipids the determination of the blood coagulability was performed in two population groups differing in diet and physical activity.

Subjects and Methods

The subjects were clinically healthy men from two social classes in Naples at the age of 51 year on an average. In one group there were the members of the wealthy class (bankers, directors of various companies and the members of the local Rotary club) and the second group was chosen from the heavy workers in the harbour of Naples. The judgment of clinical health was made after a physical examination including E.C.G. and history taking by an internist. On medical examination a few subjects were rejected as being not completely clinically healthy. The study included 30 men of the wealthy class and 32 workers.

All blood samples were drawn from veins in the antecubital fossa. The tourniquet applied was as short as possible in order to avoid a prolonged stasis of the blood. The subjects reported for the examination in the morning and were in fasting condition.

For the whole blood clotting time the blood was withdrawn with siliconized syringes and needles coated with Arquad 2C. If there was any difficulty in obtaining blood, a new venepuncture was made with a new needle and syringe. A stopwatch was started as soon as the blood entered the syringe. One ml. of blood was added to each of 4 siliconized tubes in water bath of 37° C and 1.5 ml. of blood were added to 0.5 ml. of 0.1 M sodium oxalate, the latter mixture being centrifuged to provide oxalated plasma. The prothrombin time was estimated at 37°C in a 100 per cent, and then in a 60, 40, 30 and 12,5 per cent plasma, diluted with 0.85 per cent NaCl. To 0.1 ml. of this diluted plasma 0.1 ml. of thrombokinaze »Geigy« was added and then, after a few seconds 0.1 ml. of 0.02 M calcium chloride was added, and the stopwatch was started.

The blood lipids were determined as follows: Total lipids were determined by the method of Sperry and Brand (2), the lipid phosphorus by the method of Fisk and Subberow (3) on the total lipids extract as above obtained. Phosphatidyl ethanolamine was determined by the method of Axelrod and Brodie (4). Total Cholesterol was analysed by the Anderson and Keys version of the method of Abell and al. (5), and the Cholesterol distribution between alpha and beta lipoproteins was measured separately by the method of Anderson and Keys (6). The

Phenol+Heparin test was performed according to the personal suggestion of Dr. Scanu and Dr. Lewis from Dr. Page Laboratory. All the analyses were carried out in duplicate.

Body weight and height were measured in socks and underwear. For comparison purposes relative body weights were calculated as a percentage of the average United States Standards for the corresponding height and age as given in the Medico-Actuarial Mortality Investigation (1912), sometimes referred to as Davenport's standards (7).

The subcutaneous fat thickness was estimated with calipers having a constant tension at all openings, and was recorded for the double skinfold under a constant pressure of 10 gm. per square milimeter of jaw face. Measurements were made over the triceps muscle at a point halfway between the elbow and the shoulder tip of the scapula. The scapula fold was measured over the tip of the scapula.

Two additional anthropometrical measurements were taken with the purpose of obtaining better information about the difference in the subjects' physique. The biacromal diameter was determined as the distance between the most lateral margins of the acromion process of the scapulae, and the bicristal diameter was obtained as the greatest distance between the lateral margins of the iliac crest.

Results

Table 1. shows the results of the anthropometrical examination. The average relative body weight compared with the American standards is higher among the wealthy class subjects than in the workers whose relative body weight is in accord with the values given as normal in the standard. The sum of skinfold measurements as a measure of leannes–fatness shows a striking difference between these two groups. The higher values for the wealthy class subjects indicate the contribution of the adipose tissue to the differences in relative body weight.

Table 1. presents some indices which can be useful for determinig the difference in body size and composition. The sitting height over the standing height as a measure of the relative lenght of legs versus the trunk does not show any difference. The index biacromial diameter over the bicristal diameter which can be an index of masculinity versus feminility shows higher values for harbour workers. The upper arm diameter minus thickness of the skinfold taken in the same place is a good measure of muscularity. The difference shown in table 1, indicates that workers have a better developed muscular tissue. The last index, showing the biacromial plus bicristal diameter over the height is an index of skeletal width. It shows only slightly higher values in workers.

Table 1.

Anthropometrical measurements

Category	Age		Relative body weight (%)			
	Mean	Range	Mean	Range	S. D.	S. E.
Industrialists	50.7	43—60	109.3	83.3—143.5	12.98	2.41
Workers	51.7	42—60	100.8	81.4—133	15.03	2.74

Sitting height *Biacromial Diam.*

	Standing height				Bicristal Diam.			
	Mean	Range	S. D.	S. E.	Mean	Range	S. D.	S. E.
Industrialists	.53	.50—.55	.014	.003	1.31	1.06—1.56	.091	.017
Workers	.53	.50—.56	.014	.003	1.34	1.20— .49	.075	.014

	Skinfolds				Upper Arm Diam.-Skinfold Thickness			
	Mean	Range	S. D.	S. E.	Mean	Range	S. D.	S. E.
Industrialists	41.1	25—69	13.34	2.48	5.26	2.97—7.51	1.156	.215
Workers	25.4	10—45	10.31	1.88	6.36	3.05—8.30	1.095	.200

Biacromial Diam. + Bicristal Diam.

	Standing height			
	Mean	Range	S. D.	S. E.
Industrialists	.41	.37—.44	.021	.004
Workers	.42	.38—.45	.017	.003

From these indices we can conclude that the subjects from the wealthy class in Naples have a greater value for relative body weight than the workers in the harbour of the same age, and that the difference can be accounted for the larger amount of adipose tissue, since there was no difference in the skeletal frame. At the same time the workers have a better developed muscular tissue.

Table 2.

Blood coagulability and blood lipids in two different population groups in Naples.
(I = Industrialists; W = Workers)

		mean	S. E.	t
Whole blood clotting time (min)	I.	27.96	0.89	—4.77
	W.	34.11	0.88	
Prothrombin time (sec)	I.	21.3	0.21	—7.85
	W.	23.6	0.21	
Total lipids (mg/100 ml)	I.	837.0	33.2	5.66
	W.	637.0	14.3	
Lipid Phosphorus (mg/100 ml)	I.	11.22	0.67	4.16
	W.	8.41	0.18	
Phosphatidyl ethanolamine (mg/100 ml)	I.	0.566	0.039	0.65
	W.	0.534	0.027	
Total cholesterol (mg/100 ml)	I.	213.0	6.89	3.88
	W.	180.0	4.79	
Cholesterol in beta-lipoproteins (mg/100 ml)	I.	172.0	6.83	3.92
	W.	139.0	5.03	
% beta-lipoproteins	I.	81	0.87	3.69
	W.	77	1.05	
Phenol Test (opt. dens.)	I.	0.518	0.023	3.64
	W.	0.325	0.020	

The results of blood examinations concerning the coagulability of blood lipids are summarized in Table 2. The blood coagulability measured as the whole blood clotting time and prothrombin time was shorter in the subjects of the wealthy class than in the workers and all values for blood lipids as well as phenol + heparin test were higher in the subjects of the wealthy class. The statistical analysis of the data shows that the differences between these two groups were significant

for all the values except for the difference in phosphatidyl–ethanolamine. Phosphatidyl–ethanolamine was considered as a factor responsible for the shortening of blood coagulability after a fatty meal (8).

Discussion

Evidence is presented that there is a difference in blood lipids concentration and blood coagulability measured as a whole blood clotting time and prothrombin time in the population groups differing in physical activity, relative obesity and diet. These differences are in agreement with vital statistical data showing that coronary heart disease is relatively rare among the general population of Naples, but that it is fairly common among the upper class of Naples.

From the study of Dr. Keys (9) in the Japanese in Japan, Hawai and Los Angeles it can be concluded that cholesterol differences do not depend on differences in relative obesity measured as skinfold thickness obtained by the use of calipers with constant pressure, or in physical activity. This confirms the previous work done in Minnesota, Sweden and South Africa, which have shown that the large differences in serum cholesterol concentration observed when the populations of different dietary habits are compared, are not the results of a difference in physical activity (10). This does not imply, however, that physical activity has no effect on cholesterol metabolism.

The third variable to be discussed is the diet. Unfortunately, we were not able to perform a dietary survey among the subjects examined, but as a source of information about the diet of our subjects we used the data of a nutritional survey carried out in the Quarter of Stella which is one of the largest and most populated quarter of Naples.

Table 3.

Average daily consumption of proteins, fats, carbohydrates and total calories in the quarter of Stella in Naples.

Category	Proteins gr.	Fats gr.	% vegetable Fats	Carbo-hydrates gr.	Calories	% calories from fat.
Industrialists and Owners	80	120	50.8	489	3425	31.9
Workers	62	67	65.5	428	2619	23.2

In Table 3. the results of the nutritional survey are summarized for the wealthy class as well as for the workers. The difference in the ca-

lorie intake which exists between these two groups is related mostly to the difference in the fat intake. The workers consumed half as much fat as the wealthy class subjects. The amount of fat expressed as the percentage of the total calories is 32 per cent for the wealthy class subjects and 23 per cent for the workers. The intake of vegetable fat versus animal fat shows a difference in these two groups. The subjects of the wealthy class cover 50.8 per cent of their daily fat intake from vegetable origin against 65.6% in the working class.

Conclusions

These results offer additional evidence that the difference in diet influences the level of blood lipids in man. New evidence is presented that the population groups which differ in their dietary habits and in their mode of life show a significant difference in the blood coagulability measured as the whole blood clotting time and the prothrombin time. This supports our previous, unpublished observations made in two population groups differing in blood lipids concentration and in the incidence of ishaemic heart disease in Minnesota.

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Sadržaj

KOAGULACIJA KRVI I LIPOIDI U SERUMU

(Studija stanovništva)

Dvije populacione grupe, koje se razlikuju po svojoj prehrani, a osobito po povećanoj potrošnji masnoća i smanjenom fizičkom aktivitetu, ispitivane su s obzirom na nivo krvnih lipida i na koagulabilitet krvi. Rezultati pokazuju, da prehrana, osobito povećana potrošnja animalnih masnoća, te smanjena psihička aktivnost utječu na povećanje nivoa krvnih lipida i bržeg koagulabiliteta krvi mjerenog kao vrijeme zgrušavanja i protrombinsko vrijeme. Ti rezultati potvrđuju već ranija zapažanja autora o povećanom koagulabilitetu krvi u populacionim grupama, koje se istovremeno razlikuju i u koncentraciji krvnih lipida i u učestalosti koronarnih bolesti.

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