

POLYUNSATURATED FATTY ACIDS IN LEPROSY

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INTRODUCTION

Experiments of Burr and Burr (^{2,3}) clearly established that the complete exclusion of fat, and thereby the polyunsaturated fatty acids—unsaturated fatty acids with more than one double band (essential fatty acids, EFA)—from the diet slowly produces a definite disease. Much work has been done on the relation between EFA and human blood of different age groups, of different dietary habits and of different diseases.

Sinclair (¹⁰) found that coronary heart disease may be due to dietary deficiency of the EFA, linoleic and arachidonic acids being most important. There is less absolute proof that EFA are required more by man than in the case of other species. Brown and others (¹) demonstrated a decrease of plasma polyunsaturated fatty acids in adult man during a prolonged low fat diet. Page and associates (⁹) stated that the occurrence of atherosclerosis—civilized man's greatest killer—is focal, and that the presence of EFA (lipids in general) are the essential vital substances to prevent it. Sinclair (¹⁰) reported that deficiency of polyunsaturated fatty acids may increase the susceptibility to noxious agents, including the carcinogenic effects of x-ray irradiation and chemical carcinogens. They also pointed out that in patients with anemia, total fatty acids show higher values than in normal subjects, as is also the case of tuberculosis patients. Hansen (⁷) reported that in infants on a low fat diet there develops a characteristic eczema which rapidly responds when fat is added to the restricted regimen. Faber and Roberts (⁵) and Cornbleet (⁴) correlated the appearance of eczematous symptoms in adult subjects with the reduction in the level of EFA in the blood, and especially with that of linoleate and arachidonate.

Thus it will be seen that there exists a definite relationship between the level of polyunsaturated fatty acids of the blood with different diseases. The present work was therefore undertaken to study the level of polyunsaturated fatty acids in different types of leprosy patients of different age groups.

MATERIAL AND METHODS

The patients were classified into four different classes: (1) lepromatous, (2) tuberculoid major, (3) tuberculoid minor, and (4) maculoanesthetic. Each of these four classes were further divided into four different age groups, ranging from 15 years upward. All the patients were from the inpatient section of Acworth Leprosy Home, Wadala, Bombay, who were all vegetarian and of the low income group.

From each of the subjects under consideration, in the postabsorptive state (8-10 hours), 10 cc. of blood was drawn from the antecubital vein with a dry syringe. The blood was allowed to clot, and the supernatant liquid was centrifuged and the clear serum was used for analysis. Three cc. of serum was used in each case. The fatty acids were extracted from the serum by the method of Wiese and Hansen⁽¹¹⁾, first precipitating the proteins with a 3:1 alcohol-ether mixture, and then extracting the fatty acids from the supernatant liquid by repeated saponifications, acidifying in fatty acids, and dissolving them in petroleum ether (40°-60°). The fatty acids were isomerised by the alkali isomerisation method of Herb and Riemenschneider⁽⁸⁾. The 21 per cent KOH-glycol reagent was prepared by heating 28 gm. of 85 per cent KOH in 100 gm. of ethylene glycol (Merck) to 180°C and cooling to room temperature. The reaction tube was 1 × 10" in size, and the weighing cups 8 mm. in diameter and 6 mm. in height, both made of pyrex glass. The spectroscopic methyl alcohol was prepared first by refluxing the methyl alcohol (Merck) with Zn-dust and KOH, and then distilling it. All dilutions of isomerised mixtures were made with the spectroscopic methyl alcohol, and readings were taken on the Beckman-DU spectrophotometer.

Equations for calculating percentages of acids.—Constants for acids other than hexaenoic were taken from the data of Herb and Riemenschneider⁽⁸⁾ and constants of hexaenoic were taken from Hammond and Lundburg⁽⁶⁾.

RESULTS AND DISCUSSION

Polyenoic fatty acids in the serum of the patients of lepromatous type.—The distribution of dienoic, trienoic, tetraenoic, pentaenoic and hexaenoic acids in the total fatty acids of the serum of the lepromatous patients of various age groups is shown in the first section of Table 1.

Age group 15 to 24 years: The mean value for total fatty acids is 304 ± 3.65 mgm. per 100 cc. of serum. Dienoic acids constitute 22.67 ± 4.52 per cent, trienoic acids 5.54 ± 0.99 per cent and tetraenoic acids 4.96 ± 3.07 per cent. The average percentages of pentaenoic and hexaenoic acids with standard deviations are 1.39 ± 0.17 and 1.18 ± 0.62 , respectively.

Age group 25 to 34 years: The mean value for total fatty acids is 312 ± 17.00 mgm. per 100 cc. of serum, which is found to be very high as compared to normal subjects (293 mgm.). The percentages of constituting polyenoic fatty acids also indicate change from normal subjects. Except the trienoic acids, all other percentages of polyenoic fatty acids are lower than those of normal subjects. In the case of trienoic acids, it is nearly double the normal titer. The mean values of percentages of polyenoic fatty acids are (values in parentheses are of normal subjects): dienoic, 21.06 ± 1.66 (21.43); trienoic, 4.47 ± 0.89 (2.23); tetraenoic, 2.83 ± 0.47 (6.80); pentaenoic, 0.53 ± 0.11 (1.06); and hexaenoic, 0.44 ± 0.29 (2.55).

Age group 35 to 44 years: The mean value of total fatty acids is

TABLE 1.—*Polyenoic fatty acids in the serum of patients of various types.*

Age group (years)	No. of samples	Total fatty acids ^a	Polyenoic fatty acids in per cent of total fatty acids				
			Dienoic	Trienoic	Tetraenoic	Pentaenoic	Hexaenoic
<i>1. Lepromatous type</i>							
15-24	4	304± 3.68 ^b	22.67±4.53	5.54±0.99	4.96±3.08	1.39±0.17	1.18±0.62
25-34	4	312±17.00	21.06±1.66	4.47±0.89	2.83±0.47	0.52±0.11	0.44±0.29
35-44	4	321±17.00	20.35±0.38	6.81±1.25	2.61±0.23	0.34±0.07	0.33±0.13
45+	4	332± 9.87	18.41±1.23	3.75±0.14	2.45±0.46	0.38±0.16	0.56±0.25
<i>2. Major tuberculoid group</i>							
15-24	4	289± 9.76 ^b	24.30±0.85	5.59±2.08	4.30±0.27	1.92±0.69	2.25±0.48
25-34	5	307± 6.75	21.95±1.21	4.88±0.97	4.53±0.55	1.26±0.28	2.04±0.44
35-44	5	320± 7.70	20.39±0.47	5.72±2.46	2.77±1.26	0.73±0.37	1.84±0.28
45+	7	344± 7.01	20.15±1.52	4.30±1.45	3.68±0.97	0.92±0.65	1.98±0.40
<i>3. Minor tuberculoid group</i>							
15-24	6	283±11.66 ^b	22.77±2.09	4.27±1.33	4.87±1.41	0.98±0.25	1.98±0.35
25-34	5	310± 8.88	20.67±1.35	3.19±1.11	4.05±1.46	0.71±0.36	1.65±0.35
35-44	6	321±10.74	20.39±1.59	6.63±1.47	5.29±1.15	1.20±0.20	2.05±0.34
45+	5	346± 8.87	19.75±0.95	4.47±1.05	4.53±1.23	1.15±0.47	1.20±0.36
<i>4. Maculoanesthetic group</i>							
15-24	4	293±13.81 ^b	22.96±1.62	4.31±1.55	5.34±1.56	1.49±0.84	1.95±0.34
25-34	5	310± 9.37	22.26±1.93	5.58±1.29	4.75±0.92	1.93±1.23	2.00±0.27
35-44	4	326± 5.15	23.79±0.63	6.43±2.16	5.06±1.80	1.59±0.57	2.26±0.56
45+	5	344±12.00	20.54±0.93	5.12±1.23	4.96±1.02	1.29±0.54	1.82±0.34

^a Milligrams in 100 cc. of serum.^b Standard deviations.

328±17.00 mgm. per 100 cc. of serum, which also is high as compared to that of normal subjects (309 mgm.). In this group also, similar to the preceding group, except for trienoic acids the percentage values of polyenoic fatty acids are lower than those of normal subjects. In case of trienoic acids, the value is found to be about 3 times the normal. The values are: dienoic, 20.35±0.38 (21.83); trienoic, 7.33±1.25 (2.18); tetraenoic, 2.58±0.23 (6.23); pentaenoic, 0.39±0.07 (1.10); and hexaenoic, 0.11±0.01 (2.58).

Age group 45 years onwards: The mean value for total fatty acids in this group is 332±9.88 mgm. per 100 cc. of serum, higher than that in normal subjects (324 mgm.). In this group the same trend is seen as in above two groups in case of the polyenoic fatty acids, i.e., except trienoic acids, the percentage values of the other polyenoic fatty acids are lower than those of normal subjects. They are: dienoic, 18.41±1.22 (19.99); trienoic 3.75±0.14 (2.17); tetraenoic, 2.45±0.46 (5.53); pentaenoic, 0.38±0.15 (0.97); and hexaenoic, 0.56±0.25 (2.06).

In general, in this class of the disease, it can be seen that the values of total fatty acids in the serum increases with the increase in age. The same is the case with the values of the dienoic, tetraenoic and pentaenoic acids. There is no such trend with the trienoic and hexaenoic acids. The values of total fatty acids are higher in this class, as compared to normal subjects, but the values of percentages of polyenoic acids are lower than normal, the only exception being trienoic acids. In all age groups, as normal subjects, the values decrease from dienoic to hexaenoic.

Major tuberculoid cases.—The values of total fatty acids in 100 cc. of serum of the major tuberculoid cases, with the percentages of their constituting polyenoic fatty acids, are shown in the second section of Table 1. In this class the values of total fatty acids increase and the percentage values of dienoic acids decrease with the increase of age, thus showing similarity to the trend in normal subjects. That of the percentage values of trienoic acids is somewhat similar to that in the lepromatous type. In the case of the pentaenoic and hexaenoic acids, there is a decrease with the increase in age up to the third age group, and then again increases. Similar to the lepromatous type, the values of total fatty acids are higher than in normal subjects, but the percentage values of the constituting polyenoic acids, other than trienoic, are lower than in normals. Another characteristic point of this form of the disease is that the percentage values of hexaenoic acids are higher than those of pentaenoic acids.

Minor tuberculoid cases.—The third section of Table 1 represents the values of total fatty acids in 100 cc. of serum in minor tuberculoid cases, with the percentage values of their constituting polyenoic fatty acids. In this class, also, the value of total fatty acids increase with age, and the percentage values of dienoic acids decrease, showing a similarity to the findings in the major tuberculoid class. In case of trienoic acids, there is a similarity to the major tuberculoid class as well as to the lepromatous type. There is a similar trend as regards the percentage values of tetraenoic, pentaenoic and hexaenoic acids, i.e., they first decrease from the 15-24 years age group to the 25-34 years group, then they increase in the next age group and decrease in the last one. Similar to the above two forms of the disease, values of total fatty acids are higher than those of normal subjects, and lower with the polyenoic acids, except that of trienoic acids. The percentage values of hexaenoic acids are higher than those of pentaenoic acids.

Maculoanesthetic cases.—The values of total fatty acids in 100 cc. of serum of the patients of maculoanesthetic class, with the percentage values of their constituting polyenoic fatty acids, are given in the fourth section of Table 1. In this case also the values of total fatty acids and percentage value of dienoic acids show a trend similar to

that in the lepromatous and tuberculoid types. In case of trienoic and hexaenoic acids, their values increase with the increase in age up to the 35-44-years age group, and again decrease in the last age group. The percentage values of hexaenoic acids are higher than those of pentaenoic acids. In this form of the disease, also, the values of total fatty acids are higher and those of the polyenoic acids are lower than those in normal subjects, the only exception being of trienoic acids.

SUMMARY

The polyenoic fatty acids of the serum of the various types of leprosy patients of different ages have been determined by the alkali isomerisation method of Herb and Riemenschneider. In all the forms of the disease studied, the values of total fatty acids are higher, and the percentage values of polyenoic fatty acids (other than trienoic acids) are lower, than those in normal subjects. The values of total fatty acids increase and the percentage values of dienoic acids decrease with increase in age. Except for the lepromatous type, the percentage values of hexaenoic acids are higher than those of pentaenoic acids in other types. The percentage values of polyenoic acids go on decreasing from dienoic to hexaenoic acids.

RESUMEN

Por el método de la isomerización alcalina de Herb y Riemenschneider se han determinado los ácidos grasos poliénoicos del suero de las varias formas de leproso de distintas edades. En todas las formas de la enfermedad estudiadas, los valores de ácidos grasos totales son más altos, y los valores porcentarios de los ácidos grasos poliénoicos (aparte de los ácidos triénoicos) más bajos que en los sujetos normales. Los valores de los ácidos grasos totales aumentan y los valores porcentarios de los ácidos diénoicos disminuyen al subir la edad. Exceptuada la forma lepromatosa, los valores porcentarios de los ácidos hexaénoicos son más altos que los de los ácidos pentaénoicos. Los valores porcentarios de los ácidos poliénoicos siguen disminuyendo de los ácidos diénoicos al hexaénoicos.

RESUMÉ

Les acides gras polyénoïques présents dans le sérum de malades atteints de différents types de lèpre et d'âges divers ont été déterminés par la méthode d'isomérisation basique de Herb et Riemenschneider. Dans toutes les formes de la maladie qui ont été considérées, les valeurs absolues d'acides gras sont plus hautes et les proportions d'acides gras polyénoïques autres que l'acides triénoïques sont plus basses que chez les sujets normaux. Les valeurs absolues d'acides gras augmentent avec l'âge alors que la proportion d'acides diénoïques décroît. Sauf dans le type lépromateux de l'affection, la proportion d'acides hexaénoïques est plus élevée que celle d'acides pentaénoïques. Les pourcentages d'acides polyénoïques décroissent progressivement de l'acides diénoïques à l'acides hexaénoïques.

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