Diet and Atherosclerosis

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SUMMARY

Among the various factors affecting the development of atherosclerosis and its complications, the diet emerges as an important influence. This article reviews the evidence linking diet and atherosclerosis; the relation between serum cholesterol concentration and incidence of coronary heart disease, and the effect of various dietary components on the serum lipids of man. The role of diet in the prevention of coronary heart disease is briefly discussed.

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The term atherosclerosis, introduced by Marchand in 1904, designates the form of arteriosclerosis characterised by accumulation of soft, amorphous, lipid material in the vascular intima. The presence of cholesterol among the lipids of the atheromatous plaque has been known for many years, and more recent studies have shown that the cholesterol content of the human aorta increases with the severity of the lesion.[11]

The origin of the lipids deposited in the atheromatous lesion is still a controversial subject, but it appears that they are mainly derived from the lipids circulating as lipoproteins in the blood plasma, in agreement with Virchow's filtration theory.

Atherosclerosis has been considered as a consequence of the ageing process, but it is currently well established that, although atherosclerosis is statistically related to age, the rate of its development varies considerably from individual to individual and among different populations. It is presently believed that atherosclerosis is a complex process influenced by a number of factors, environmental as well as genetic. Among the former, the diet, and more specifically its content in saturated fatty acid glycerides and cholesterol, has emerged as an important influence, and, since the diet can be modified, there is much interest at present in the possibility of preventing the development of atherosclerosis and its clinical complications by suitable modifications of the diet.

It hardly needs to be said that current interest in atherosclerosis is due to the fact that coronary heart disease (CHD), one of the complications of the atherosclerotic process, is the leading cause of death in many Western countries.

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INFLUENCE OF THE DIET IN THE DEVELOPMENT OF ATHEROSCLEROSIS AND ITS COMPLICATIONS

The evidence relating diet and atherosclerosis may be considered under 3 headings.

Experiments in Animals

Animal experiments have demonstrated the possibility of producing lesions resembling those found in human atherosclerosis, by feeding certain diets. Following the early observations of Anitschkow and other Russian authors,[5,6] numerous workers have demonstrated the production of lipid deposits in the arteries of various animal species, by feeding diets containing high proportions of fat and cholesterol. There are considerable differences between animal species in regard to their susceptibility to develop experimental atherosclerosis, as well as differences between the experimental lesions and those found in man. The relevance of experimental atherosclerosis for understanding the development of atherosclerosis in man has been described by Katz and Stamler[24] as follows: 'Even the most severe critics of the cholesterol concept of atherosclerosis cannot fail to note the conspicuous similarities between cholesterol-induced lesions, whether in rabbit, common dog, or chick, and the lesions seen in man. Taking into account the differences among the various species in architecture of the great vessels, the similarities between experimental cholesterol-induced and human lesions are indeed remarkable.'

More recently Wigand[60] and others have demonstrated the production of atherosclerosis by feeding diets rich in saturated fats, without the addition of cholesterol. It is now well established that atherosclerosis can be produced experimentally in a variety of animal species by feeding diets which produce hypercholesterolaemia, whether or not they contain added cholesterol.[50,52]

Observations in Populations Subjected to Changes in their Habitual Diets

The German pathologist Aschoff noted during the latter part of World War I a decline in the frequency of finding severe atherosclerotic lesions at autopsy, which he attributed to the decrease in fat intake among the civilian population.

Similar, but more extensive observations were made during World War II, particularly in the Scandinavian countries. Mortality statistics, hospital records and necropsy reports in Finland, Sweden and Norway indicate a reduction during the war years in the intensity of atherosclerosis and the frequency of its clinical complications.[41] Comparable data have been reported in other countries.

Needless to say, the change in the diet was not the only change in the mode of life of these populations, and the reduction of fat intake was not the only dietary change. However, as noted by Keys,[25] there is little doubt that in every population for which acceptable data are available, a decrease in fat consumption was associated with a decrease in the incidence of atherosclerotic heart disease, followed by an increase upon return to a diet of higher fat content.

Migration of populations to a different geographical area often involves changes in dietary habits, giving an opportunity to study possible associations between diet and disease. Among these studies the observations of different groups of Jews migrating to Israel are widely quoted. The relative incidence of myocardial infarction among men recently migrated from Yemen was considerably lower than among earlier Yemenite immigrants, and this difference has been related to the difference in fat content of the diets consumed by these two groups.

Epidemiological Studies

Shortly after World War II a number of publications demonstrated that, in contrast with common experience in Western countries, autopsies in other parts of the world revealed a strikingly low incidence of advanced atherosclerotic lesions. It is a pleasure to acknowledge here the important contribution of the South African workers to these studies.[22] More recent epidemiological studies have given strong support to the relation between diet and the manifestations of the atherosclerotic process, in particular CHD. Keys *et al.*[26] have reported the results of their International Co-operative Study, describing the findings on 12 770 men aged 40 - 59 years at the outset of the study. The subjects include 11 cohorts, each of 500 to 1 000 men, in Finland (2 cohorts), rural Yugoslavia (3 cohorts), Italy (2 cohorts), Greece (2 cohorts) and Japan (2 cohorts). Over 95% of all the men in that age group, residing in the corresponding geographical areas, were examined. In addition, two other cohorts in Yugoslavia, one in Holland, and a cohort of 2 571 railroad workers in the United States, were also studied. The report[26] describes the findings of the 5th-year re-examination, which covered 94% of all survivors. There were marked differences in the intake of saturated fatty acid glycerides, ranging from 22% of the total calorie intake in East Finland to 3% in Japan, and a high correlation between the age standardised rate of CHD deaths and infarctions, and the proportion of calories supplied by saturated fatty acid glycerides ($r = +0,84$). On the other hand, the correlation between incidence of coronary infarction and total fat intake was low ($r = +0,40$). It is of interest to note that some of the populations studied, in particular those of the Greek islands, have a high intake of fat, mainly in the form of olive oil. They have, therefore, a high intake of monoene fatty acid glycerides (about 29% of the total calorie intake in the case of the island of Crete), but a low intake of saturated fatty acid glycerides. The mortality due to CHD, as well as the total mortality, was extremely low in these populations. The total number of deaths for the Greek cohorts in 5 years was 21. The expected number of deaths for men of this age, estimated from the *USA Vital Statistics for 1962*, for an equal period of time, is 70,5. The observed mortality is therefore only

30% of that expected. The intake of monoene fatty acids showed a negative correlation with the incidence of CHD ($r = -0,42$), but this correlation did not reach statistical significance ($P>0,05$). Furthermore, this negative correlation could be accounted for by the negative correlation between the intakes of saturated and monoene fatty acid glycerides ($r = -0,36$).

The data of the International Co-operative Study indicate therefore a close association between the intake of saturated fatty acid glycerides and the incidence of CHD. As noted by Keys,[26] these data do not prove that saturated fatty acids in the diet play a major role in the development of CHD, but the results are consistent with that hypothesis.

It is currently believed that the effect of the diet on the development of the atherosclerotic process and its complications, in particular CHD, is mediated through the effect of the diet on the serum lipid levels. This view is represented by the so-called dietary fat, cholesterol, theory of atherosclerosis. In its simplest form this theory postulates that the diet, and, more specifically, its content of fat and cholesterol, has an influence on the serum cholesterol level which, in turn, has an influence on the development of the atherosclerotic process. Without considering more recent sophistication in the statement of the theory, it must be noted that this view is not incompatible with the currently accepted idea of the multifactorial causation of atherosclerosis. Recognition of the diet's role does not imply disregard of other factors. There is no longer any question about the significance of the diet in the development of atherosclerosis; the question is rather, as Keys has pointed out,[27] to determine the magnitude of its role and the extent to which dietary management might contribute to the control of CHD.

Since the effect of the diet is believed to be related to its effect on the blood lipids, particularly cholesterol, an analysis of the effect of the diet on atherosclerosis demands a consideration of the relation between serum cholesterol and the development of atherosclerosis and its complications, as well as a consideration of the effect of the diet on the concentration of serum cholesterol and other lipids.

Serum Lipids and Coronary Heart Disease

As already noted, there is much evidence that elevated serum lipid levels, in particular high levels of serum cholesterol, are associated with the development of atherosclerotic lesions. The evidence relating serum cholesterol levels and CHD in man has been obtained in three types of studies:

Clinical observations. An extensive literature shows that patients suffering from CHD have, as a group, higher serum cholesterol levels than comparable individuals free of the disease. Numerous reports, which I cannot attempt to analyse in detail, support the view that high serum cholesterol levels and CHD are indeed associated. They do not prove, however, that elevated serum cholesterol is the actual cause of CHD. Furthermore, since in many of the studies the cholesterol analysis was done after the coronary event, it is not possible to decide whether the high serum cholesterol level actually preceded the clinical manifestations of the disease. There are many reports, however, demonstrating a high incidence of CHD among individuals suffering from conditions associated with high serum cholesterol levels.

A recent report[34] describes the development of severe atherosclerosis in the heart transplanted into Dr. Blaiberg. Since it is assumed that the heart was healthy when the transplant was made, it appears that the severe coronary atheromatosis developed in the course of the 19 months that the transplanted heart was irrigated by Dr. Blaiberg's hypercholesterolaemic blood (315 mg/100 ml). This observation seems to provide striking evidence in support of the role of elevated cholesterol in the development of coronary atheroma. However, the possibility that the rejection process might have influenced the formation of the lipid deposits should not be ignored.

Epidemiological studies. There is substantial evidence showing that the differences in CHD mortality among different countries are associated with differences in the serum cholesterol levels. When groups of individuals of the same age and sex are compared, it is generally found that those living in countries with high coronary mortality have high serum cholesterol levels as well. Similar results have been obtained when comparing different areas within certain countries, and when comparing in the same country groups of individuals belonging to different socio-economic groups. The relation between serum cholesterol concentration and risk of developing CHD is supported by the results of the International Study previously mentioned.[26] The correlation between the serum cholesterol medians for 13 cohorts and the age-standardised 5-year incidence rate of CHD death and hard criteria infarcts is $r = +0,76$. That between serum cholesterol median and incidence of all CHD is $r = +0,81$.

Follow-up studies. Strong evidence in support of an association between serum cholesterol level and development of CHD has been obtained in follow-up studies of groups of individuals clinically free of CHD at the outset. These studies have consistently shown a marked tendency for the new cases of CHD developing during the course of the study to occur among those men with the highest levels of serum cholesterol. The data of the Co-operative Study published in 1956[32] have been confirmed and extended by other studies such as the Framingham study[23] and others, including the study in our laboratory in Minneapolis. The analysis of these data by Keys *et al.*[36] includes a total of 31 197 man-years during which 251 cases of CHD developed. The relative incidence of CHD (as a percentage of the incidence for the whole group) is strongly associated with the serum cholesterol concentration at the beginning of the study. On the average, men with initial serum cholesterol values of 260 mg/100 ml and more had an incidence of CHD 4,3 times greater than men of the same age living in the same community, whose serum cholesterol level was below 200 mg/100 ml. In a study of 4 300 men followed for 3 years, Westlund[39] reported no new cases of CHD among men with initial cholesterol

below 200 mg/100 ml, and incidence of 4,1% among men with initial cholesterol 350 - 399 mg/100 ml.

The close association between serum cholesterol level and incidence of CHD makes it possible to predict the risk of suffering a coronary event from the serum cholesterol level. The risk appears to be proportional to the third power of the serum cholesterol concentration. A new analysis of the results obtained in the group studied in the Laboratory of Physiological Hygiene for 23 years shows that major CHD incidence was proportional to cholesterol to the 3,4 power.[37] It seems clear that the risk of developing CHD increases continuously with increasing serum cholesterol level. Similar results have been obtained by more elaborated mathematical analysis of the relationship between serum cholesterol and risk.[56]

A recent analysis of CHD risk, using multiple logistic equations, shows that predictions based on a number of variables, including the serum cholesterol level, give useful estimates of the relative risk of CHD for individuals. However, although predictions based on European data are highly correlated with American data, and vice versa, the application of predictions based on European data tends to underestimate the absolute values observed among American men. This important study suggests the contribution of other unidentified and unrelated variables to risk of American men.[22]

The statistical association between serum cholesterol level and risk of developing CHD seems to be established beyond any reasonable doubt, but resistance to accept the evidence supporting this association is still occasionally found in the literature. Thus, in a recent review on nutrition and ischaemic heart disease[63] it is stated that 'excellent studies have shown that blood cholesterol levels constitute a poor index as to impending heart attacks'. It turns out that the references quoted by the author of the review do not include any publication of the last 10 years and no mention is made of any of the follow-up studies considered here.

EFFECT OF THE DIET ON THE SERUM LIPIDS

For many years clinicians have denied the possibility of influencing serum cholesterol concentration in man by dietary means. One of the first observations showing an effect of the diet is that of Schoenheimer who in 1933[49] noted a marked decrease of serum cholesterol in a hyperlipaemic patient when her usual diet was replaced by a pure vegetarian diet.

With the introduction of the rice-fruit diet for the treatment of hypertension it was noted that this diet produces marked decreases of serum cholesterol in a few days. Keys *et al.*[35] reported that the cholesterol-depressing effect of the rice-fruit diet could be prevented or reversed by adding a hydrogenated vegetable fat to the diet. This observation indicates that the effect of the rice-fruit diet on serum cholesterol depends on its low fat content, rather than on its lack of cholesterol.

Effect of Dietary Fat

The early studies by Keys and his associates on different populations demonstrated a relationship between fat intake and serum cholesterol levels. When the average serum cholesterol for groups of 40 - 49-year-old men living in different countries was plotted against the average fat intake (as a percentage of the total calorie intake), a straight-line relationship was obtained.[33] The data demonstrated also that groups of individuals of the same racial stock, but with different dietary habits, show differences in serum cholesterol levels which are related to the fat intake. These early studies did not take into consideration the differences in the nature of the dietary fat. It will be shown later that this relationship reflects the fact that for most usual diets the serum cholesterol level is mainly determined by the amount of saturated fatty acid glycerides in the diet, and that an increase in the total fat intake generally means an increase in the intake of saturated fatty acids.

The influence of the amount and kind of fat in the diet on man's serum cholesterol has been studied in numerous experiments over the past 20 years. For obvious reasons I should like to discuss, as an example, the experiments that Drs Anderson, Keys and myself have been doing for almost 20 years at the Laboratory of Physiological Hygiene. These experiments were made under carefully controlled conditions in middle-aged men free of metabolic diseases, residents in a mental hospital and in an institution for the mentally retarded in Minnesota.

The fats were incorporated into diets as close as possible to habitual American diets in isocaloric exchange for carbohydrates or other fat of different composition. Calorie intake was individually adjusted in order to maintain constant body weight throughout the experiments. All of the experiments followed a reversal design pattern in order to avoid the effect of time trends. The duration of the dietary periods was generally 2 - 3 weeks. Cholesterol intake was maintained constant.

In the analysis of the results it was assumed that the effect of dietary fat on serum cholesterol concentration depends on the fatty acid composition of the glycerides, and that each of the three main kinds of fatty acids (saturated, monoene, and polyene) has a specific effect on serum cholesterol which is independent of the effect of the others.

Statistical analysis of the results of a first series of experiments showed that an increase in the saturated fatty acid content of the diet equal to 1% of the total calorie intake (with corresponding decrease of dietary carbohydrate), was associated with an average increase in serum cholesterol of 2,7 mg/100 ml. Similarly, an increase in the diet content of polyunsaturated fatty acid glycerides corresponding to 1% of the total calorie intake, in exchange for an isocaloric amount of carbohydrates, was associated with an average decrease of serum cholesterol of 1,3 mg/100 ml. No significant change of serum cholesterol was produced by changing the proportion of monounsaturated fatty acids in the diet. Accordingly, the change in serum cholesterol concentration expected when chang-

ing from a given diet to another of different fatty acid composition, other things being equal, is expressed by the equation:

$$\triangle \text{ Chol.} = 2{,}7 \triangle \text{S} - 1{,}3 \triangle \text{P} \qquad (1)$$

where △ Chol. represents the serum cholesterol change in mg/100 ml and △ S and △ P the change in saturated and polyunsaturated fatty acid content of the diet expressed as a percentage of the total calorie intake.[28]

Further experiments have given comparable results and have consistently shown that the cholesterol-rising effect of the saturated fatty acids is about twice the cholesterol-depressing effect of the polyunsaturated fatty acids.[29,30] It follows from this relationship that the effect of a given fat on serum cholesterol concentration depends on the value of the expression 2S − P, that is to say the difference between twice the proportion of saturated fatty acid glycerides and the proportion of polyunsaturated glycerides, both expressed as a percentage of the total calorie intake. It also follows that fats containing an amount of polyunsaturated fatty acids twice the amount of saturated fatty acids have no effect on serum cholesterol because 2S − P = 0.[20]

Application of equation 1 to the data obtained by other workers results in predictions which are in satisfactory agreement with the serum cholesterol changes reported.[30]

It is of interest to compare the results of these short-term experiments with the data from populations subsisting habitually on diets of different fat content. The correlation coefficient between saturated fatty acid glycerides intake (as a percentage of the total calorie intake) and serum cholesterol for 11 cohorts of the Co-operative International Study (27) was $r = +0{,}942$, and $r = +0{,}89$ for 14 cohorts.[26] The latter calculation included one cohort whose dietary intake was calculated from dietary records rather than from the analysis of the diets, as was done with the other cohorts. Using these data Keys has calculated that the relationship between serum cholesterol and 2S − P for these populations is described by the equation:

$$\text{Chol.} = 2{,}85\ (2\text{S} - \text{P}) + 152 \qquad (2)$$

The coefficient 2,85 is greater than the coefficient 1,35 derived from the data of the controlled experiments. Keys has suggested that this difference may be explained in part by an inverse relationship between the intake of saturated fat and the intake of other dietary components that tend to reduce serum cholesterol levels, and by a positive correlation between the intake of saturated fat and the intake of cholesterol. He has also suggested the possibility that life-long subsistence on a given diet produces greater effects on the blood lipids than those noted in short-term experiments.

The changes of serum cholesterol concentration produced by changing the fatty acid composition of the diet are accounted for mainly by changes in the beta lipoprotein cholesterol, and the serum phospholipid changes observed are, in general, parallel to those of cholesterol.

The serum triglycerides are also affected by the amount and composition of the fat in the diet. Fats rich in polyunsaturated fatty acids tend to produce lower serum triglyceride levels than diets containing equal amounts of monoene or saturated fats and of the same carbohydrate content.[4]

In spite of the considerable evidence showing that the blood lipid levels in man can be modified by changing the fatty acid composition of the diet, the validity of the data is denied by some authors. The paper published by Reiser[47] should be mentioned in this respect, as an example of unqualified and misleading criticism.

It is well known that within a given population individuals eating comparable diets may have different cholesterol levels, and that no statistical association can be demonstrated between dietary intake and serum cholesterol concentration of persons in populations eating a relatively uniform diet. Under these conditions, the variability in serum cholesterol attributable to individual factors outweighs that from differences in food selection.

Effect of Dietary Cholesterol

Compared with the rabbit and the chicken, man is less sensitive to dietary cholesterol, but controlled experiments have shown that serum cholesterol concentration in man is influenced by the cholesterol content of the diet. Several authors have shown consistent elevations of serum cholesterol by adding cholesterol to diets of low cholesterol content. In our experiments we used a diet containing about 50 mg of cholesterol/day, which was added with the amounts of cholesterol needed to make the daily intake equal to 380, 520 and 1 460 mg/day respectively. Cholesterol was incorporated into the oil used in the preparation of the diets, and the diets were fed following a reversal design similar to that previously described. It was observed in these experiments that serum cholesterol concentration increased with the cholesterol content of the diet.[19,31] Using our results and the data from 4 other laboratories, it was calculated that the change in serum cholesterol produced by changing the cholesterol content of the diet over the range of 0 to 3 441 mg/day is described by the equation:

$$\triangle \text{ Chol.} = 1{,}5\ (Z_2 - Z_1) \qquad (3)$$

where △ Chol. represents the change in serum cholesterol concentration in mg/100 ml, and Z_2 and Z_1 represent the square roots of the amounts of cholesterol in the diets being compared, expressed as mg/1 000 kJ.

More recently Mattson *et al.*[48] have found a linear relationship between dietary cholesterol and serum cholesterol concentration. Each 100 mg of cholesterol in 4 000 kJ of diet resulted in approximately a 12 mg/100 ml increase in serum cholesterol concentration. It appears, therefore, that according to these data, our square root formula tends to overestimate the effect of diets with low cholesterol content and to underestimate the effect of high cholesterol diets. For levels of dietary cholesterol of the order of 150 mg/4 000 kJ, both equations give comparable results. This level of dietary cholesterol is somewhat lower than that found in the usual American diets, which is considered to be of the order of 250 mg/4 000 kJ.

Effect of Dietary Carbohydrates

Since carbohydrates have been taken as the reference point to estimate the effects of dietary fat, it follows from

the preceding discussion that carbohydrates have an effect on serum cholesterol intermediate between that of the saturated and the polyunsaturated glycerides. Isocaloric substitution of mixed dietary carbohydrates for saturated glycerides causes a decrease of serum cholesterol, whereas substitution of mixed carbohydrates for polyunsaturated glycerides causes an elevation of serum cholesterol. No change of serum cholesterol is observed when the carbohydrates are substituted for monoene fatty acid glycerides or for fats having a value of $2S - P = 0$.[17,20]

The concept that replacement of sucrose by complex carbohydrates (mainly starch) causes a decrease of serum cholesterol in man, has found wide acceptance, but critical examination of the data available reveals that the changes in serum cholesterol produced in normolipaemic individuals by such dietary exchanges are small, and do not always reach statistical significance. It has been found that when starch is supplied in the form of certain natural foods, the serum cholesterol levels are lower than those observed with a similar diet containing an isocaloric amount of sucrose. Thus, several years ago we observed a decrease of serum cholesterol level when starch in the form of leguminous seeds was substituted for sucrose, but we have been unable to duplicate this result in more recent experiments. We have, however, observed that when starch is supplied as a mixture of various vegetables, the serum cholesterol levels are lower than those observed in the same individuals with diets containing either sucrose, wheat flour, or leguminous seeds. I have recently reviewed the literature[18] and will abstain from discussing here the numerous experiments reported. It appears clear that the changes in serum cholesterol produced by changing the nature of the carbohydrates in the diet of normolipaemic individuals are of limited importance. The following quotation from Macdonald[40] aptly summarised our present knowledge regarding the effect of dietary carbohydrates on the serum cholesterol concentration of normolipaemic individuals: 'The serum cholesterol level is very sensitive to the amount and type of lipid in the diet, and if it is affected by the amount and type of dietary carbohydrate *per se*, then this influence is small compared with that of the dietary lipid'.

Because some populations characterised by low serum cholesterol levels subsist on diets rich in cellulose, it has been suggested[9,22,58] that this complex carbohydrate might have a cholesterol-depressing effect in man, and more recently Trowell[55] has reviewed the relation between dietary fibre and ischaemic heart disease. However, experiments in our laboratory and by others, have shown that daily supplements of 15 g of cellulose have no effect on serum cholesterol concentration in man.[34] Eastwood[15] observed that increasing the cereal fibre content of the diet did not reduce serum cholesterol, whereas removing cereal fibre from lacto-ovo vegetarian diet was associated with a decrease in serum cholesterol.

Several years ago it was observed in our laboratory that the addition of pectin (15 g/day) to the diet produces a decrease of serum cholesterol in normal subjects.[34] The decrease is of the order of 5% of the level observed when feeding the same diet without pectin supplement. This effect has been confirmed by other authors in man and animals. However, the amounts of pectin needed to produce this effect are, by far, much larger than those found in usual diets.

Under unusual dietary conditions it is possible to observe marked changes of serum cholesterol concentration in man when sucrose is substituted for glucose. Thus, Winitz *et al.*[42] using a chemical diet containing amino-acids, vitamins, minerals, 2 g/day of linoleic acid as the only source of fat, and glucose providing about 90% of the calorie intake, observed a marked decrease of serum cholesterol when this diet was substituted by the usual diet of the subjects. When the chemical diet was replaced by a similar diet containing 75% of glucose and 25% of sucrose, instead of glucose alone, the serum cholesterol concentration rose by 48 mg/100 ml in 3 weeks. The diet containing glucose alone was fed again and the serum cholesterol once more decreased. This experiment is of considerable importance in showing that the effect on the serum lipids of exchanging dietary carbohydrates can be drastically affected by the nature of the diet used.

In contrast with the minimal serum cholesterol changes observed in normal individuals when exchanging sucrose and starch in the diet, substitution of sucrose for starch produces marked elevations of serum cholesterol in hyperlipaemic patients, well documented in the literature.[28]

It is well established that when normolipaemic persons, eating a high-fat diet, are changed to a low-fat, high-carbohydrate diet of the same caloric value, almost all of them respond in a few days with an increase in fasting serum triglyceride concentration. The elevation is of the order of 50% of the initial triglyceride level.[2] This effect is much more marked in patients whose serum is visibly hyperlipaemic when eating their ordinary diet, as described by Ahrens *et al.*[3] and confirmed by many other workers.

In normal individuals the serum triglycerides tend to return to the initial level when the carbohydrate diet is maintained for several months, as shown by the original work of Antonis and Bersohn.[7] Habitual subsistence with high carbohydrate diets, however, is usually not associated with high levels of serum triglycerides as indicated by numerous reports. Furthermore, an increase in the carbohydrate content of the diet has no effect on serum triglyceride levels when it is accompanied by an increase in the proportion of polyunsaturated to saturated fatty acid glycerides, as shown by the data of the National Diet-Heart Study. In this study the change from the usual diet, with about 40% of fat calories ($S = 21$, $P = 6$) to a diet with about 30% of fat calories ($S = 7$, $P = 11$) and corresponding increase in carbohydrates, produced no triglyceride rise, but rather some tendency in the opposite direction.[45]

We have recently tested the effect of two cholesterol-lowering diets on serum triglyceride and cholesterol concentration in university students. The diets were prepared by reducing the amounts of total fat, saturated glycerides and cholesterol, and by increasing the amounts of polyunsaturated glycerides and carbohydrates of the diet used at the Hastings State Hospital in Minnesota. Both cholesterol-lowering diets caused marked decreases of serum

cholesterol concentration without significant elevation of serum triglycerides.[4]

As compared with starch, sucrose has been considered to produce greater increases of serum triglycerides. However, our experiments comparing sucrose with starch supplied in the form of different natural products failed to show any difference in serum triglyceride concentration in young university students.[18]

Dietary Protein

There are some reports in the literature describing changes in the serum lipids associated with changes in the amount and composition of the dietary proteins, but most of the evidence indicates that, within the range of variation of the diets usually eaten in the Western countries, protein intake has no significant effect on the serum lipids of man. Drastic reductions of protein intake, however, are associated with low serum lipid levels and with deposit of lipid in the liver.

The results of our experiments and our recent review of the literature concerning the effect of dietary proteins on the serum lipids of normal individuals, indicate that changes in the protein content of the diet are of no particular value in designing diets for the reduction of the serum lipid levels in man.[2]

THE DIET IN THE PREVENTION OF CORONARY HEART DISEASE

Because the development of the atherosclerotic process and its complication of CHD is related to the level of serum cholesterol, and because the level of serum cholesterol can be modified by changing the composition of the diet, there is much hope that a modification of the diet producing a decrease of serum cholesterol concentration can afford a certain degree of protection against the development of atherosclerosis and its clinical complications. Most current work on the dietary prevention of CHD is based on the concept that the serum cholesterol concentration can be significantly reduced by diets low in saturated glycerides and cholesterol and high in polyunsaturated glycerides.

The feasibility of maintaining men with these modified diets has been tested in the National Diet-Heart Study.[45] The results of this study show that it is possible to produce, in a free-living population, significant and sustained changes in serum cholesterol which are in reasonable agreement with the predictions derived from the controlled dietary studies previously discussed.

The trials of dietary prevention of CHD fall into two categories: primary prevention and secondary prevention. In the primary prevention trials, the subjects of the study are free of detectable CHD at the outset, whereas in the secondary prevention trials the subjects are selected among survivors of a prior episode of myocardial infarction.

Primary Prevention Trials

In 1957 Dr N. Jolliffe and his associates started the Anticoronary Club in New York City. The subjects were middle-aged men considered to be at high risk of developing CHD because of high levels of serum cholesterol or obesity. Men of the same age attending the New York Cancer Detection Center, served as controls. The active members of the Club were instructed to modify their diets by reducing the total intake of fat, calories, and cholesterol and by increasing the intake of polyunsaturated fats. No dietary advice was given to the control group.[12] By the end of 1967, 17 confirmed new events had occurred during 3 954 man-years among the active members of the Club, as compared with 32 such events occurring during 3 122 man-years in the control group. The incidence rate of coronary events was therefore 2,4 times higher in the control group than in the active group, and the difference in incidence is statistically significant.[48]

The main criticism against this trial has to do with the selection of the subjects. It should be noted, however, that the control group in this study had actually a low incidence of CHD as compared with men of the same age group as those in the Framingham study. Moreover, the active group included high risk individuals who were expected to show high incidence of CHD during the period of observation.

Dayton *et al.*[34] have published the results of a study on 846 middle-aged and elderly men (mean age 66 years) living in the domiciliary unit of the Los Angeles Veterans Administration Center. The men were randomly allocated to an experimental (424 men) and a control group (422 men). The diet of the experimental group contained vegetable polyunsaturated oils incorporated in filled milk, imitation ice cream, unsaturated margarine, filled cheese, and special sausage. Vegetable oils were used in cooking and baking. The total fat content of the diet corresponded to 39% of the total calorie intake, and linoleic acid accounted for 38% of the total fatty acids in the diet. The cholesterol content of the diet was 365 mg/day. The control group continued eating the regular institutional diet with a total fat content equivalent to 40% of the total calorie intake, linoleic acid content corresponding to 10% of the total fatty acids, and cholesterol content of 653 mg/day.

The serum cholesterol level of the experimental group decreased by 12,7% from the average value at the outset, and persisted practically unchanged at this level throughout the trial. This change is in reasonable agreement with our prediction equation. The number of men sustaining definite overt myocardial infarction was 40 in the control and 27 in the experimental group. Corresponding figures for sudden death due to CHD were 27 and 18; and for definite cerebral infarction 22 and 13. The differences in sudden death or myocardial infarction were not statistically significant. When these data were pooled with those for cerebral infarction and other secondary end points the totals were 96 for the control group and 66 for the experimental ($P = 0,01$).

Miettinen *et al.*[44] and Turpeinen *et al.*[57] have reported a controlled trial started in 1959 in two mental hospitals in

Helsinki, Finland. The diet in one of the hospitals (Hospital N) was modified, replacing practically all the milk fat originally present in the diet by vegetable oil, mainly soya bean oil. Vegetable oil was used for cooking. The patients in the other hospital (Hospital K) continued eating the customary Finnish diet. After 6 years the diets were exchanged for another 6 years. Serum cholesterol decreased with the modified diet. The complete data recently published, show a marked and significant decrease in CHD for males. Mortality during the modified diet periods was about half that during the periods on the usual diet. There was also a decrease in coronary mortality among the females, but the difference for the pooled data was not significant. The authors concluded that in men a cholesterol-lowering diet considerably reduces the mortality from CHD.

Secondary Prevention Trials

There are a number of secondary prevention trials in the literature, but many of them have been criticised and should not be considered here.

The study conducted by Leren in Oslo, Norway, included 412 men below 68 years of age who had survived a myocardial infarction for an average of 20 months before being admitted to the study. The men were assigned at random to the control and treatment groups. The experimental diet was modified in order to reduce the intake of saturated fat and cholesterol and to increase that of polyunsaturated fats. The participants included in the experimental group were orally instructed to eliminate completely whole milk, cream, butter, and fat cheeses, to eat only one whole egg per week, and to reduce meat consumption, trimming all the visible fat. Use of low-fat cheeses, egg white, whale meat and poultry, was encouraged, as well as the use of fish and shellfish, and all kinds of vegetable products except coconut. Lard margarine and olive oil were restricted and the subjects were instructed to consume 0,5 litres of soya bean oil per week, mixed with dry skim milk as a substitute for butter, in baking and cooking, in salad dressings, and in daily amounts of 15 - 30 g as 'medicine'. The results after 11 years of observation[39] show significant reduction of coronary mortality for the diet group.

Although none of the studies considered provides incontrovertible evidence of the value of the diet in preventing CHD, they clearly support the view that dietary manipulation designed to decrease the serum cholesterol level is useful in reducing the risk of CHD. It is true that these studies show no differences in coronary mortality as great as those observed between populations in different countries. But it should be noted that these experiments were of limited duration and involved men who would be expected to have a sizeable degree of atherosclerosis, even if they had no previous history of myocardial infarction. Consequently, it seems reasonable to conclude that more prolonged trials begun earlier in life would have produced more striking differences. It is of interest in this connexion that in Leren's study the younger men appeared to benefit more than the older men. Bierenbaum *et al.*[10] have recently reported their 10-year experience with modified diets in younger men with coronary heart disease. They conclude: 'It seems that dietary management, if it is to have the most beneficial effect, should be started as early in life as possible in those susceptible to, or already affected by, coronary heart disease'.

The new Multiple Risk Factor Intervention Trial, about to begin in USA, is expected to give more information about the role of the diet in the prevention of CHD.

Possible Harmful Effects of the Diets Rich in Polyunsaturated Fatty Acids

In discussing the role of the diet in the prevention of CHD it is imperative to consider the possibility that diets with high content of polyunsaturated fatty acid glycerides, such as those being currently recommended, may have some undesirable effects.

It is impossible to evaluate the long-term effect of these diets because there is practically no example of a human population subsisting on a diet rich in polyunsaturated fatty acids. The Eskimos, who some years ago were cited as evidence that high fat intake has nothing to do with atherosclerosis, are perhaps the only population group subsisting on a diet which is believed to be rich in polyunsaturated fat. But the information about the Eskimos is very limited, with the exception of the recent work by Bang *et al.*[8] The data of these authors are of interest in showing that Eskimos in Greenland have low cholesterol, triglycerides and total lipids, as compared with Danes of the same age and sex, and with Eskimos living in Denmark. According to the data given by Bang *et al.*[8] CHD is rare among Eskimos and diabetes is practically unknown, but no data about other diseases are available.

Reservation about the use of diets rich in polyunsaturated fatty acids has been based mainly on two reasons: the possibility that these diets increase the requirements of vitamin E, and the possibility that high intake of polyunsaturated fatty acids may lead to an excessive production of free radicals.

Since most of the vegetable oils high in linoleic acid are believed to contain adequate amounts of alpha-tocopherol the diets used in the preventive trials can hardly be considered deficient in vitamin E. The data reported by Dayton *et al.*[13,14] indicate that the subjects eating the cholesterol-lowering diet were more abundantly supplied with vitamin E than the controls. Indeed, the experimental subjects had serum levels of alpha-tocopherol twice as high as the controls.

Polyunsaturated fatty acids are potential sources of free radicals in the organism,[46] and it has been postulated that free radicals arising from enzymatic and non-enzymatic reactions both in the cells and in the interstitial fluids, have deleterious effects. According to the free radical theory these effects may be responsible for the changes associated with ageing and with the development of certain degenerative diseases including atherosclerosis. Thus, Harman has suggested that the long-term ingestion of increased amounts of polyunsaturated fatty acids, by increasing the unsaturation of the plasma and tissue lipids

might actually enhance atherosclerosis, in spite of its cholesterol-lowering effect.[21]

These ideas obviously deserve the most careful consideration, but so far there is little evidence of untoward effects of the cholesterol-lowering diets in the clinical reports. The following statement by Malmros[42] seems to reflect the general opinion among clinicians: 'During the 14 years I have prescribed this diet for my patients I have never seen any side-effects.' The recent report describing a high frequency of cholelithiasis found in the autopsy of subjects eating highly polyunsaturated diets is an interesting and intriguing exception.[51] There is little evidence that polyunsaturated diets decrease the mortality due to CHD while increasing other causes of mortality. The analysis of the data of various preventive trials published by Ederer *et al.*,[16] concludes 'that potential health benefits of these diets in men of these ages are not likely to be outweighed by any as yet unidentified hazards.'

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