Antiatherogenic Effect of Vegetable Oil Supplemented with ω-3 Polyunsaturated Fatty Acids

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Considerable interest has recently been expressed in the antiatherogenic effect of ω-3 polyunsaturated fatty acids and the development of new drugs on this basis [6-8]. However, these substances are not readily available, since they are present mainly in fish oil (16-19% of total fatty acids) [6], whereas in more abundant sources, for example of vegetable or animal origin, ω-3 fatty acids occur in trace amounts. Therefore, despite numerous data on their positive antiatherogenic effect, their therapeutic application is still seriously limited. There are some data on Maxepa, a preparation with maximal content of eicosapentaenoic (ω-3) acid [5]; however, it is extremely expensive. At the same time, due to the high rate of atherosclerosis morbidity, the development of new accessible drugs remains an urgent problem. Of all the variously acting compounds, those based on polyunsaturated fatty acids merit special attention, since they possess a well-known antiatherogenic effect and are thus recommended by dietitians for the prevention of atherosclerosis [3,12,13]. As a rule, these are the abundant ω-6 fatty acids from vegetable oils [1,10,11]. However, most studies report not a therapeutic but, rather, a preventive antiatherogenic effect. The hypocholesterolemic effect of vegetable oil has mainly been studied on cholesterol-fed experimental animals [11,13].

In our previous investigation we showed that in rabbits with experimental atherosclerosis after cessation of a cholesterol-enriched diet, during spontaneous regression, corn oil (with 70% linolenic acid [1]) promotes cholesterol clearance, which manifests itself in a more rapid decline of the blood cholesterol concentration, as well as in a substantially reduced percentage of aortic atherosclerotic lesions [4].

In the present study the therapeutic effect of this oil supplemented with linolenic acid was studied in rabbits with alimentary atherosclerosis. In parallel (and for comparison) one group of animals received the same doses of fish oil as a source of ω-3 fatty acids.

MATERIALS AND METHODS

The experiments were carried out on 39 male rabbits with an average weight of 3 kg. The rabbits received cholesterol (3 g per day) during 3 weeks and were then switched to a standard diet. Before treatment the rabbits were divided into 3 blood cholesterol level-matched groups. Rabbits of group I received 2 ml linolenic acid-supplemented (<5%) vegetable oil per animal. Preliminary experiments carried out for determining the optimal antiatherogenic dose of vegetable oil (2, 4, and 6 ml) re-

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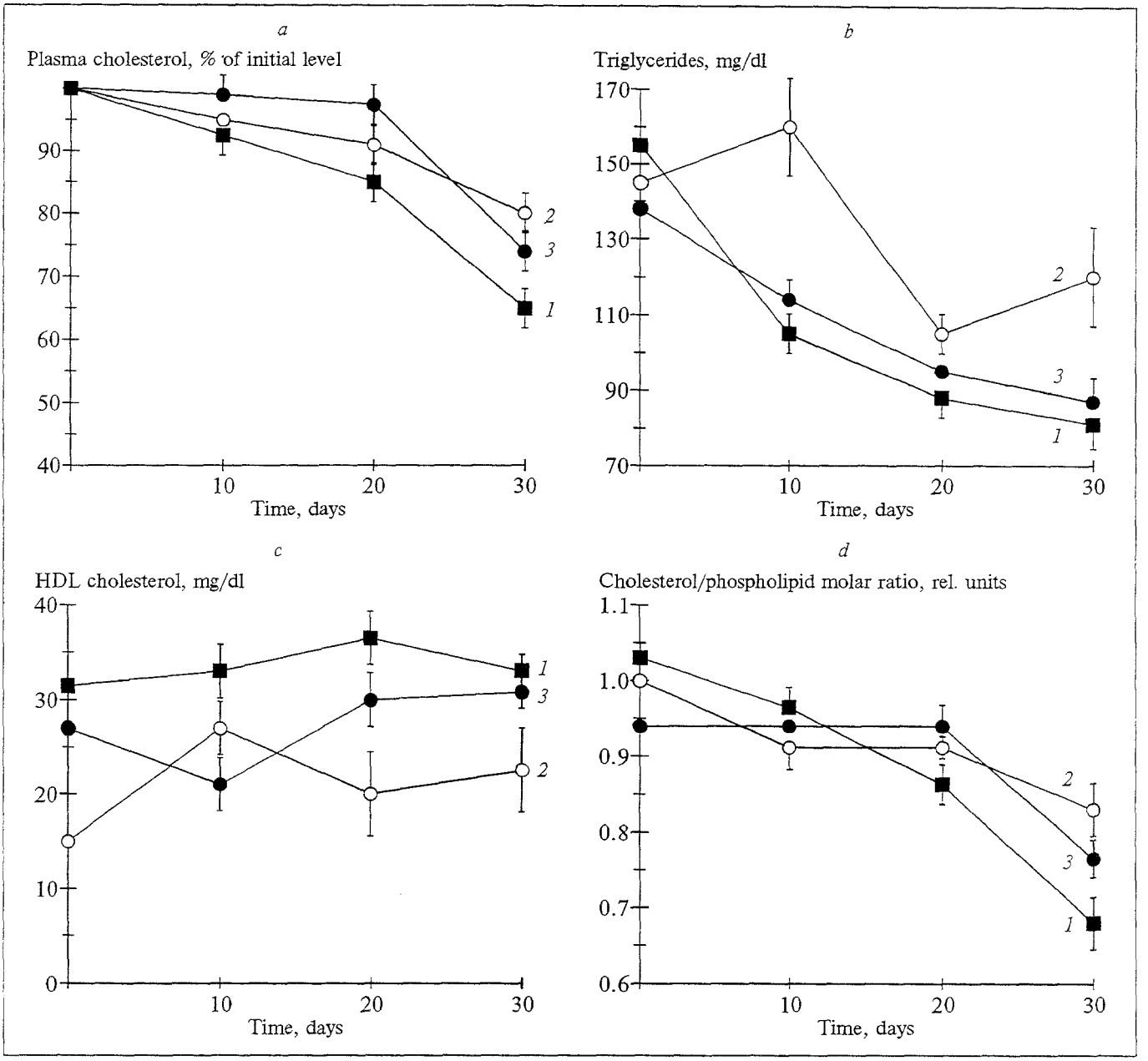

Fig. 1. Parameters of rabbit blood for treatment with linolenic acid (*1*), and fish oil (*2*) and during spontaneous regression (*3*).

vealed no reliable differences between the doses used, and therefore we used the minimal dose (2 ml) in further experiments. Group II received fish oil (2 ml per animal), with a high ω-3 content [8]. Group III comprised controls, i.e., spontaneously recovering rabbits.

The treatment lasted 30 days. Blood for the study was sampled every 10 days. The blood concentrations of cholesterol, triglycerides, and cholesterol of high density lipoproteins (HDL) were determined with a Centrifichem-400 autoanalyzer; the cholesterol to phospholipids molar ratio in erythrocyte membranes was routinely determined after Abel' and Vas'kovskii, as described elsewhere [4]. After the protocol was completed, the animals were killed and the percentage of atherosclerotic damage of the aorta was assessed using an IBAS scanner [2]. The results were processed statistically using the Student *t* test.

RESULTS

Figure 1 shows the dynamics of blood parameters in rabbits with alimentary atherosclerosis treated with linolenic acid-supplemented vegetable oil (*1*), and fish oil (*2*) and during spontaneous regression (*3*).

As is seen from Fig. 1, *a*, differences between groups in the blood cholesterol content first ap-

TABLE 1. Percentage of Aorta Lesions in Rabbits

Group	Lesions, %
1. Rabbits fed linolenic acid–supplemented oil	10.8±1.5
2. Rabbits fed fish oil	39.3±8.2
3. Spontaneous regression (control)	36.2±4.3

peared on day 20 of treatment, the minimal cholesterol concentration being observed in the group receiving linolenic acid-supplemented vegetable oil. At the end of the experiment these differences had increased and the blood cholesterol level in the oil-treated group was reliably lower than that in the group with spontaneous regression. Fish oil did not affect the blood cholesterol concentration.

The level of triglycerides (Fig. 1, *b*) dropped gradually in both the oil-treated and spontaneously recovering groups. No differences between these groups were found. In the fish oil-treated group, the triglyceride level did not drop until the 20th day of the experiment and then showed a reverse tendency to rise.

The concentration of HDL cholesterol remained unchanged over the entire experimental period in all experimental groups (Fig. 1, *c*).

However, the cholesterol to phospholipids molar ratio, which reflects the state of biomembranes, decreased more intensively in the linolenic acid+oil-treated group in comparison to the other two groups (Fig. 1, *d*). After 20 days this parameter dropped from 1.05 to 0.85 vs. 0.95 in the control. These differences were even more pronounced at the end of the experiment. Fish oil did not affect this parameter.

When determining the area of aorta atherosclerotic lesions after completion of the protocol (Table 1), we found the minimal area affected by atherosclerosis in rabbits treated with linolenic acid-supplemented vegetable oil (10.8±1.5% vs. 36.2±4.3 in the group with spontaneous regression)

A relatively high percentage of aorta involvement was unexpectedly found in the fish oil-treated group (39.3±8.2%). This result, however, may be explained in the light of recent data from Japanese investigators, who showed that in rabbits (possibly, due to some species-specific features), unlike other animals, fish oil possesses a hepatotoxic effect and probably causes secondary dyslipidemia [9].

Thus, we demonstrated an antiatherogenic effect of vegetable oil supplemented with linolenic acid. In the present experiment on rabbits with alimentary atherosclerosis it manifested itself in accelerated clearance of cholesterol from the plasma, cell membranes (erythrocytes), and ultimately, from the aorta. Although we investigated just the overall effect of this oil without studying separately the effect of linolenic acid, it may be assumed from published data [6-8] that when ω-3 fatty acid is added to vegetable oil, its antiatherogenic effect is enchanced by affecting the coagulation processes and properties of HDL. The data indicate a possible beneficial use of various combinations of vegetable oils, in particular, those supplemented with ω-3 fatty acids, as readily available antiatherogenic preparations.

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