Disposal of Intravenously Administered Fat in Subjects with Atherosclerosis and in Normal Controls


Following a fatty meal, the plasma becomes turbid because the concentration of chylomica in it rises. Several studies have shown that the intensity and sometimes the duration of this alimentary lipemia is greater on the average in subjects who have had cardiac infarction than in those who have not.1-4

Some of these studies have been open to criticism because the possible effects of a recent infarction or treatment given for it have not been considered or because the controls were not well matched. However, the study of Bronte-Stewart and Blackburn3 is not open to these criticisms and accords well with the earlier work. A difference in alimentary lipemia between patients with cardiac infarction and other subjects has also been revealed by tracer studies in which triolein labeled with 131 has been given orally.5-7

It is difficult to interpret differences in alimentary lipemia revealed by studies of this sort for the extent of the lipemia depends on contemporary processes of absorption and clearing of fat. The variable effect of absorption on lipemia could be excluded if fat could be given intravenously. Ideally such fat should be in the form of a standard chyle, but its preparation for study in man presents difficulties. Meanwhile, since a stable and safe emulsion of cottonseed oil (Lipomul I.V.)* for intravenous use has become available, we have studied the lipemia produced by it in healthy subjects and in patients who have suffered cardiac infarction. In addition, a group of patients with peripheral vascular disease was studied.

Methods

Five groups of patients were studied. They were all ambulant at the time of the test, and the age structure of the groups is shown in table 1.

Group I: The control group consisted of 12 subjects who were either healthy or who were recovering from illnesses unrelated to the cardiovascular system. They were free of clinical evidence of coronary or other arterial disease.

Group II: The untreated infarct group consisted of 10 patients who had suffered a myocardial infarction 5 to 6 months before the test was performed and who were receiving no treatment for their condition.

Group III: The heparin-treated infarct group consisted of 11 patients who had suffered a myocardial infarction 5 to 12 months before the test and who were receiving 250 mg. of aqueous heparin subcutaneously twice a week for at least 3 months before the test. At the time of the test 3 to 4 days had elapsed since the last injection.

Group IV: The phenprocoumon (phenprocoumon)-treated infarct group consisted of 11 patients who had suffered a myocardial infarction 3 to 9 months before the test and who had been receiving anticoagulant therapy continuously since their illness in dosage sufficient to maintain a prothrombin index of 20 to 30 per cent. The drug used was phenprocoumon except in 1 patient who was receiving phenylindanedione.

Group V: The peripheral vascular disease group consisted of 8 patients with intermittent claudication which was associated with angina pectoris in 4 patients and a history of myocardial infarction in 2 patients. They were receiving no treatment at the time of the test.

The patients in group II comprised the first patients of suitable age who could be collected from among the past or present patients in one ward at the Royal Melbourne Hospital. The patients in groups III and IV had been selected at random for each treatment for the purposes of a clinical trial. Subjects with diabetes mellitus were excluded from all groups.

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*We are grateful to the Upjohn Co., Kalamazoo, Mich., for generous supply of Lipomul I.V. The constituents of Lipomul I.V. are cottonseed oil 15 per cent, soybean phosphatide, Pluronic F68 and dextrose 4 per cent in aqueous vehicle.
Each subject fasted overnight and in the morning was given an intravenous infusion of Lipomul I.V. into the left arm. This was administered over a 30-minute period, the total dose being approximately 80 ml. During the first 5 minutes the rate of infusion was 10 drops/min. to avoid the so-called “colloid reaction,” but for the remaining 25 minutes the rate was maintained at 60 drops/min.

Before the infusion was begun, an indwelling needle was placed in a vein of the right arm. Blood samples (5 to 10 ml.) were withdrawn through it into oxalate solution before the infusion started, at 10-minute intervals after the start of the infusion for 1 hour, and at 20-minute intervals during a second hour. The plasma from each sample was spun off at 500 gravity units for 20 minutes and its optical density (O.D.) read in a Unicam spectrophotometer at 650 m\(\mu\) and 1 cm. light path.

### Results

Obvious lipemia developed in all subjects so that curves relating optical density to time could be constructed and compared.

There was great variation in the shapes of the curves. In most cases there was a linear rise in O.D. to a peak which coincided with the end of the infusion at 30 minutes followed by an exponential fall. However, sometimes the peak O.D. occurred before or after the infusion was stopped at 30 minutes, or the 30- and 40-minute readings were similar. Table 2 shows the frequency of the different times at which peak O.D.'s were reached in the 5 groups of subjects. It shows that delayed or sustained peaks occurred in all but the control group and that among groups II to V increase in O.D. after the end of the infusion was most frequent in the phenprocoumon-treated group (group IV).

Figure 1 shows the mean curve for each group. Measurements of various components of the curves are shown in table 3 and compared in table 4.

The rate of rise of O.D. was calculated as peak O.D. minus initial O.D./time. As an index of fall in O.D., the time taken for the O.D. to reach 50 per cent of the peak level (T\(\frac{1}{2}\)) was used.

Figure 1 shows that the mean curves for the control and untreated infarct groups resemble each other closely. Table 3 shows that the variation between the curves within the control group was greater than within the untreated infarct group. The mean curve of the peripheral vascular disease group was very similar to those of the control and untreated infarct group, but the mean fall in O.D. was slower. This was due largely to one very slow fall and was not statistically significant.

The mean curves for the heparin and phenprocoumon-treated groups differ strikingly from those of the untreated infarcts.

While the mean initial O.D.'s do not differ significantly, the mean peak O.D. of the heparin-treated infarcts is lower and that of the phenprocoumon-treated infarcts higher.

These differences are reflected in the slower rise in O.D. in the heparin-treated and the faster rise in the phenprocoumon-treated groups. However, the difference in rate of rise between the phenprocoumon-treated and untreated infarct group just fails to reach significance at the 5 per cent level. The differences between the control and the two treated infarct groups are not very significant at the 5 per cent level, due to the large variance in the former.

Table 3 shows that the time for O.D. to fall to 50 per cent of peak level was longer in the untreated infarcts than in the heparin-treated group and longer still in the phenprocoumon-treated group. However, these differences were not statistically significant.

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**Table 1**

<table>
<thead>
<tr>
<th>Age Frequency Distribution by Decades for Groups Studied</th>
<th>31-40</th>
<th>41-50</th>
<th>51-60</th>
<th>61-70</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group I: Controls</td>
<td>2</td>
<td>6</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>Group II: Untreated infarcts</td>
<td>0</td>
<td>3</td>
<td>4</td>
<td>3</td>
</tr>
<tr>
<td>Group III: Heparin-treated infarcts</td>
<td>1</td>
<td>4</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>Group IV: Phenprocoumon-treated infarcts</td>
<td>1</td>
<td>6</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>Group V: Peripheral vascular disease</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>3</td>
</tr>
</tbody>
</table>
INTRAVENOUSLY ADMINISTERED FAT

Table 2
Frequency of Occurrence of Peak Lipemia at Different Times After Commencement of Infusion

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Untreated Infarct</th>
<th>Heparin Infarct</th>
<th>Phenprocoumon Infarct</th>
<th>Peripheral vascular disease</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number</td>
<td>12</td>
<td>10</td>
<td>11</td>
<td>11</td>
<td>8</td>
</tr>
<tr>
<td>Peak at 20 min.</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Peak at 30 min.</td>
<td>11</td>
<td>10*</td>
<td>10*</td>
<td>4</td>
<td>7*</td>
</tr>
<tr>
<td>Peak at 40 min.</td>
<td>0</td>
<td>2*</td>
<td>2*</td>
<td>5*</td>
<td>2*</td>
</tr>
<tr>
<td>Peak at 50 min.</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>1*</td>
</tr>
</tbody>
</table>

*Peak lipemia persisted for more than 1 reading.

Discussion

Under the conditions of this experiment both the untreated myocardial infarct group and the peripheral vascular disease group had normal ability to clear their bloodstream of the particles or the artificial emulsion Lipomul I.V. or at least of that moiety capable of scattering light. The abnormal lipemia produced when fat was given orally to patients who had suffered a myocardial infarction contrasts with the results described above. Two explanations offer themselves. First, the infarct patients may absorb fat abnormally. In all of the studies where fat was given orally, the area under the abnormal curve is greater than that under the normal one. To explain this as an increased flux of fat through the blood due to increased total absorption would imply that the abnormal group absorbs more fat than do normal subjects. However, the normal absorption of dietary fat is at least 95 per cent complete; increasing absorption even to 100 per cent would not explain the observed differences. It is possible, however, that the absorptive process may result in emulsions of fat having different physical properties in the 2 groups. The same concentration of fat in 2 subjects could give rise to widely different optical densities if a greater proportion of the particles were large enough to scatter light in one than in the other. Thus, the greater turbidity produced in the myocardial infarct group could reflect an abnormal state of emulsification of the fat rather than abnormal concentration of triglyceride.

A second explanation of the disparity between the oral and intravenous findings is that the clearing of the artificial emulsion particles may proceed in the same way in both groups despite a relative inability of the infarct patients to remove from their bloodstream chylomicra derived from a fatty meal. There is evidence that fat given intravenously as an artificial emulsion rapidly enters the same metabolic path as ingested fat, although the mechanisms of removal from the bloodstream may differ. Thus, it is probable that the defect causing the development of abnormal lipemia is present at or before the stage of removal of fat particles from the blood. Davies has produced evidence that the chylomicra of patients who had suffered a myocardial infarction are indeed abnormal. Using the technic of free electrophoresis, he has shown decreased mobility of the chylomicra of patients who had suffered a myocardial infarction and he has demonstrated that the factor responsible resides in the plasma. He interprets this as indicating a difference in the surface activity of the plasma which leads to decreased colloidal stability.
Table 3

Mean Values with Standard Errors of Components of the Lipemia Curves in Each Group

<table>
<thead>
<tr>
<th>Group</th>
<th>Number</th>
<th>Mean initial O.D. ± S.E.</th>
<th>Mean rate of increase/ min. in O.D. ± S.E.</th>
<th>Mean peak O.D. ± S.E.</th>
<th>Mean time from peak O.D. to 50% of peak O.D. (T½) ± S.E. (min.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I. Control</td>
<td>12</td>
<td>22.2 ± 4.99</td>
<td>4.6 ± 0.62</td>
<td>158.3 ± 14.93</td>
<td>29.5 ± 4.03</td>
</tr>
<tr>
<td>II. Untreated infarct</td>
<td>10</td>
<td>23.1 ± 3.63</td>
<td>4.3 ± 0.36</td>
<td>159.0 ± 9.49</td>
<td>31.9 ± 2.78</td>
</tr>
<tr>
<td>III. Heparin-treated infarct</td>
<td>11</td>
<td>29.0 ± 3.76</td>
<td>3.1 ± 0.45</td>
<td>124.9 ± 12.69</td>
<td>24.8 ± 2.58</td>
</tr>
<tr>
<td>IV. Phenoprocoumon-treated infarct</td>
<td>11</td>
<td>21.9 ± 3.33</td>
<td>5.4 ± 0.41</td>
<td>200.3 ± 10.34</td>
<td>41.6 ± 5.29</td>
</tr>
<tr>
<td>V. Peripheral vascular disease</td>
<td>8</td>
<td>19.4 ± 3.52</td>
<td>4.7 ± 0.58</td>
<td>166.0 ± 17.72</td>
<td>35.6 ± 6.07</td>
</tr>
</tbody>
</table>

Table 4

Comparison Between Groups of Different Components of Lipemia Curves and Levels of Significance of Differences

<table>
<thead>
<tr>
<th></th>
<th>Control vs. untreated infarct</th>
<th>Control vs. heparin-treated infarct</th>
<th>Control vs. phenoprocoumon-treated infarct</th>
<th>Control vs. peripheral vascular disease</th>
<th>Control vs. untreated infarct and peripheral vascular disease</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial O.D.</td>
<td>Not significant</td>
<td>Not significant</td>
<td>Not significant</td>
<td>Not significant</td>
<td>Not significant</td>
</tr>
<tr>
<td>Rate of increase in O.D.</td>
<td>Not significant</td>
<td>Not significant</td>
<td>Not significant</td>
<td>p &lt; 0.05</td>
<td>Not significant</td>
</tr>
<tr>
<td>Peak O.D.</td>
<td>p &lt; 0.05</td>
<td>p &lt; 0.01</td>
<td>p &lt; 0.01</td>
<td>Not significant</td>
<td>Not significant</td>
</tr>
<tr>
<td>Fall (T½)</td>
<td>Not significant</td>
<td>Not significant</td>
<td>Not significant</td>
<td>Not significant</td>
<td>Not significant</td>
</tr>
</tbody>
</table>

thetic emulsifiers which stabilize the Lipomul particles may adequately replace diminished plasma surface activity and thus disguise a deficiency responsible for impaired clearing of an alimentary lipemia in the infarct group.

Thus, the demonstrated ability of the untreated infarct group to clear the Lipomul normally does not necessarily conflict with the earlier findings relating to alimentary lipemia, but it does serve to focus attention on the chylomicra themselves as a likely site for these differences.

In view of the great similarity in the behavior of the untreated infarct, peripheral vascular disease and control groups, the differences seen in the 2 treated groups are particularly noteworthy. The major part of the chylomicra cleared from the blood are hydrolyzed soon afterwards, probably by lipoprotein lipase in the tissues. It has been shown that intravenous injection of Lipomul I.V. causes the appearance of lipoprotein lipase in the blood, and so it seems likely that the enzyme plays a part in the utilization of the artificial emulsion similar to its role in the physiologic situation after a fatty meal. We have observed that long-term heparin therapy was more closely associated with increased in vitro clearing activity after a stimulating injection of heparin in some of the patients studied here than was found in the untreated infarct group. It is possible that this "priming" of clearing factor production may explain the enhanced ability of the heparin-treated group to remove Lipomul from the blood.

The phenoprocoumon-treated group reached a significantly higher peak level than the untreated group, and more of the individual subjects exhibited a rise in O.D. after the end
of the infusion than the subjects in any other group. This rise may be related to some changes occurring in the physical state of the particles, but the explanation is at present obscure.

Although many of the patients suffered from angina of effort and levels of lipemia comparable to those seen after a fatty meal occurred frequently in these experiments, none of them suffered any chest pain during the experiment, and there was not deterioration of effort tolerance afterwards. This contrasts with the findings of Kuo and Joyner who reported a high incidence of angina in such patients at the height of an alimentary lipemia.

Summary
A series of experiments has been performed in which a standard dose of an artificial oil emulsion (Lipomul I.V.) was given to 5 groups of subjects and the disappearance of fat from their blood was observed by plasma optical density measurements. The groups were age-matched and consisted of a control group; 3 groups who had suffered a myocardial infarction and were respectively untreated, receiving heparin and receiving phenprocoumon after recovery from the acute stage of the illness; and the last group which consisted of patients who suffered from intermittent claudication. The control, untreated infarct and peripheral vascular disease groups exhibited similar curves of optical density on time. The mean peak optical density was significantly lower in the heparin-treated than in the untreated group. Conversely, the mean peak optical density of the phenprocoumon-treated group was significantly higher than that of the untreated or control groups.

Acknowledgment
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References
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