Intestinal Absorption of Fatty Acids, and Blood Lipid Composition in Sheep

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ABSTRACT

Interrelationships between intestinal uptake of fatty acids and their concentrations in lipids of blood plasma of sheep were assessed by quantities of individual fatty acids that flowed through and were absorbed from the intestinal tract under different dietary conditions.

Major long-chain fatty acids were approximately 90% digested, thus demonstrating that dietary fatty acids of high melting points can be absorbed efficiently by ruminants provided they are well dispersed.

Relationships were linear between uptakes of 16:0, 18:0, 18:1, and 18:2 fatty acids from the gut and their concentrations in both triglycerides and triglyceride-free plasma lipids. The proportion of each transferred to triglyceride-free plasma lipids was in order 18:2 > 18:1 > 16:0 > 18:0, whereas in plasma triglycerides the order was 16:0 = 18:0 = 18:2 < 18:1. Interconversion of 18:0 to 18:1 by intestinal mucosa may explain the anomalous behavior of 18:1 triglycerides.

These results are consistent with the hypothesis that the intrinsic nature of the fatty acid primarily determines the composition of triglyceride-free plasma lipids whereas the relative amount of each acid absorbed by the intestine determines that of plasma triglycerides and, hence, of milk and depot fats of ruminants.

INTRODUCTION

In ruminants there are differences in partitioning of the four major dietary fatty acids (16:0, 18:0, 18:1, and 18:2) among the various lipid fractions of blood plasma. For example, by far the greatest amount of total fatty acids in blood is 18:2; yet only a small quantity of it is in the triglycerides (TG), thus contrasting with the distribution of 18:0 within the other plasma lipid classes (17, 18). Because plasma TG are the major source of long-chain fatty acids for milk fat synthesis (1), it follows naturally that the amount of 18:2 in ruminant milk fat is also low.

Bickerstaffe et al. (3) used labeled C18 fatty acids to examine the transfer of these acids in high-oil diets from the intestine to lymph and milk at one fixed intake in goats. They concluded that at that particular intake the rate of transfer of each acid from one site to another was approximately the same. However, in another experiment with sheep (16) the uptake of 16:0 from the gut was much greater than that for 18:0. Christie (6) suggested that the concentration of 18:2 in plasma TG may not respond in a linear manner to its uptake from the gut of ruminants. Therefore, to increase markedly 18:2 in plasma TG of animals receiving low-fat diets, it may be necessary to have a disproportionately larger increase in the amount of this acid reaching the small intestine.

In view of current interest (13) in feeding high-fat rations to ruminants, it is important to establish how concentrations of major long-chain fatty acids in blood change in relation to the amount of each that is absorbed from the gut over the range of intakes of current feeding practice.

It is particularly important to find out if the relationship differs significantly at low and high intakes of dietary lipid.

Palmquist and Mattos (11) showed that the rate of transfer of 18:2 from blood TG to milk fat can be affected by the metabolic state of the animal. Therefore, if the same conditions apply to the rate of transfer from gut to blood, then it is essential that these rates be measured simultaneously for all four major dietary fatty acids in the same animal for the comparisons to be valid. Results of such an investigation with sheep are reported here.
MATERIALS AND METHODS

Animals and Diets

Four sheep with average weight 70 kg were fitted with permanent rumen and duodenal fistulas and kept in metabolism cages. The duodenal cannula was placed as close as possible to the pylorus without interfering with its function and well away from the point of entry of the common bile duct.

Each animal was given, in turn, one of four diets that consisted of 500 g DM (dry matter) per day of either ryegrass hay or ryegrass silage as the forage and either 570 g DM per day of a low- or 515 g DM per day of a high-oil concentrate mixture. The concentrate was based on ground barley, extracted soybean oil meal, starch, molasses, and minerals. Soybean oil made up 8% of the weight of the concentrate in the high-oil treatment and replaced starch on an isoenergetic basis. Chromic oxide (Cr₂O₃) was incorporated into the concentrate portion of the ration to measure the rate of passage of certain components through the alimentary tract.

Animals received their daily ration in two equal portions at 0600 and 1800 h, and water was available ad libitum. Each sheep was assigned randomly to one treatment sequence of a 4 × 4 Latin square balanced for carryover effects. Each period lasted 28 days, and changeovers between treatments were abrupt. All results were analyzed statistically by methods outlined by Snedecor and Cochran (15).

At various times during the last 2 days of each experimental period samples of blood were taken from the jugular vein whereas samples of liquor were taken from the rumen by a suction probe (9 mm, i.d.) through the fistula and of duodenal contents by gravity through the cannula. On days 19 to 26 inclusive of each period, all the feces were collected from the sheep; a representative sample of each day's collection was stored at -20°C and bulked at the end of each period for chemical analyses.

Chemical Analyses

Amounts of Cr₂O₃ in the samples of food, duodenal contents, and feces were measured by the method of Stevenson and De Langen (19), and quantities of Ca and Mg in food and feces were estimated by the method of Bligh and Dyer (4) and quantified by that of Christie et al. (5). The TG were separated from other lipid classes by thin-layer chromatography (8).

RESULTS AND DISCUSSION

Nonlipid Components

The digestion coefficient of Ca was approximately 25% of that for Mg on all four diets. There was a tendency (P>.05) for both minerals to be absorbed more efficiently on silage compared to hay diets. Although significantly less Ca (P<.05) and Mg (P<.01) were taken up from the gut on high- compared to low-oil diets (Table 1), this may not be reflected necessarily by concentrations in blood (18). Reductions in uptake of Ca agreed with results of other researchers for both sheep (21) and cows (12). However, Palmquist and Conrad (12) also found that if absorption of Ca was low (i.e., about 4%), addition of oil to the diet also could increase uptake. Although they found Mg absorption was variable, in general it was less on high-fat diets. If Ca and Mg form indigestible soaps with fatty acids in the intestine, then in my experiment the extra fatty acids excreted in the feces on high-oil diets

<table>
<thead>
<tr>
<th>Mineral</th>
<th>Forage concentrate</th>
<th>Hay</th>
<th>Silage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Low-oil</td>
<td>High-oil</td>
</tr>
<tr>
<td>Ca</td>
<td></td>
<td>11.9</td>
<td>7.3</td>
</tr>
<tr>
<td>Mg</td>
<td></td>
<td>44.0</td>
<td>35.1</td>
</tr>
</tbody>
</table>

\textsuperscript{a}Standard error of difference between treatment means.
would be sufficient to account for the excess loss of these two minerals (Table 1).

Addition of oil to the diet reduced the amount of fat-free organic matter (OM) taken up from the rumen-reticulum of the sheep on both the hay- \( (P<.05) \) and silage-based \( (P<.01) \) diets (Table 2). Although the amount digested postduodenally increased \( (P>.05) \) on high-oil diets, it was insufficient to prevent reduction of the overall digestibility coefficient of the OM. The fat-free OM of the silage was absorbed more completely than that of the hay \( (P<.001) \). The reduction in absorption of the OM associated with increased oil intakes confirms \( (10, 16) \) and is probably the result of reduction in digestion of the fiber fraction \( (16) \) in the preduodenal part of the intestine. The partial compensatory increase in uptake in the postduodenal part of the alimentary tract is more difficult to explain. One possible reason, however, may be that when the fat concentration of the diet is increased, there is an enhanced flow of highly digestible microbial matter to the duodenum \( (7) \).

**Alimentary Lipids**

Variation in concentration of each of the major fatty acids in rumen liquor throughout the day depended on the nature of the fatty acid as well as on composition of the diet (Figure 1). The inverse relationship between amounts of unsaturated C\(_{18}\) fatty acids and those of 18:0 in the rumen results from microbial hydrogenation \( (14) \).

The rumen exerted a moderating influence on variations of concentration of lipid that appeared in duodenal fluid (Figure 2); not only was the amplitude of peaks and troughs of lipid concentrations less in the duodenal fluid, but the rates at which these maxima and minima were reached also were much slower as compared to those in the rumen.

Most of the unsaturated fatty acids were hydrogenated before they reached the duodenum with a larger percentage of those in the food saturated on the hay- compared to the silage-based diets \( (P<.01) \). However, the amount of lipid in the silage portion of the diet was sufficiently greater than that in the hay to ensure that the total quantity of stearic acid produced on silage diets exceeded \( (P<.001) \) those based on hay (Table 3).

The quantity of fatty acids in the diet was the same as that passing to the duodenum. This finding contrasts with results of Sutton et al. \( (20) \), who reported an increase in the amount of fatty acids reaching the duodenum over that in the food. However, Bickerstaffe et al. \( (3) \) found that in the goat the apparent amount of synthesis of fatty acids in the rumen ranged between 0 and 20 g per day. The major difference between the diets in my investigation and those used by Sutton et al. \( (20) \) was that the hay which the latter used contained only about 20\% C\(_{18}\) fatty acids compared to over 60\% in my trial.

In my experiment, recovery of fatty acids from food, especially from forage, was much greater when the material was incubated first with 5 M HCl acid, whereas fatty acid contents of duodenal fluid were the same whether the material was subjected first to treatment with strong acid.

Individual and total fatty acids in the low-oil

**TABLE 2. Percentage of dietary fat-free organic matter disappearing from different sites of the alimentary tract of sheep given diets containing hay or silage with either low- or high-oil concentrates.**

<table>
<thead>
<tr>
<th>Site</th>
<th>Forage concentrate</th>
<th>Hay Low-oil</th>
<th>Hay High-oil</th>
<th>Silage Low-oil</th>
<th>Silage High-oil</th>
<th>SE(^a)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Preduodenum</td>
<td></td>
<td>47.4</td>
<td>42.4</td>
<td>52.1</td>
<td>43.4</td>
<td>1.74</td>
</tr>
<tr>
<td>Postduodenum</td>
<td></td>
<td>24.5</td>
<td>26.0</td>
<td>28.0</td>
<td>31.9</td>
<td>1.53</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>71.9</td>
<td>68.4</td>
<td>80.1</td>
<td>75.3</td>
<td>.92</td>
</tr>
<tr>
<td>Preduodenum as percent of total</td>
<td>65.9</td>
<td>62.0</td>
<td>64.7</td>
<td>57.6</td>
<td>1.81</td>
<td></td>
</tr>
</tbody>
</table>

\(^a\) Standard error of difference between treatment means.

with silage diet were absorbed more completely in the duodenum than those in the low-oil hay-based diet \((P<.01)\). Inclusion of soybean oil in the diet increased the proportion of both total \((P<.01)\) and most of the individual fatty acids absorbed from the lower gut (Table 4) with this increase being entirely due to added fatty acids. With the exception of 18:2, the constituent fatty acids of the soybean oil tended to be absorbed more from the hay than from the silage-based diets \((P>.05)\).

Digestibility coefficients for fatty acids in Table 4 are much greater than those in (16), especially those for stearic acid. This disparity may be related to the difference in the origin of 18:0 in both experiments. In (16), direct addition of large paricles of stearic acid would hinder formation of small micelles because of the high melting point (70°C) of this acid. In comparison, it is reasonable that monomeric dispersion of \(C_{18}\) unsaturated fatty acids is an obligatory prerequisite for conversion to stearic acid by rumen microorganisms. This in turn would lead to the saturated product.
being dispersed monomerically and, therefore, would increase the probability of formation of small micelles, which enhances efficiency of absorption (9).

The experiment also demonstrates that efficiency of lipid uptake from intestine may be facilitated by the moderating influence of the rumen (Figure 1) in releasing fatty acids in such
a way that the duodenum receives a more even flow throughout the day, thereby reducing the likelihood of excessive concentration building up in the fluid and interfering with intestinal uptake (Figure 2).

Blood Lipids

Relationships between amounts of each of the three acids — 16:0, 18:0, and 18:2 — that were absorbed from the intestine and their concentration in the blood TG was linear and significant \((P<.01, .001, .01, \text{respectively})\), and slopes of lines describing these relationships were approximately the same (Figure 3). Although this relationship for the 18:1 acid also was linear \((P<.001)\), the slope of the line describing it was much greater than that for the other three acids. The most probable reason for this marked increase is that about 7 to 9% of 18:0 is desaturated to 18:1 in the intestinal wall of ruminants (3). Because the amount of 18:0 absorbed per day was about seven times that of 18:1, this interconversion would be insufficient to cause any major change in the slope of the line for stearic acid but would cause a substantial change in that for 18:1.

Relationships between intestinal uptake of fatty acids and their concentration in the TG-free plasma lipid was linear (Figure 4) and significant \((P<.01 (16:0), <.001 (18:0), <.01 (18:1), <.05 (18:2))\) but differed markedly from those in plasma TG (Figure 3). The rate of increase in TG-free lipid of plasma differed also between acids and was in order 18:2 > 18:1 > 16:0 > 18:0 for each incremental increase in intestinal uptake.

Intercepts on Figures 3 and 4 show that the concentration of each of the major fatty acids in plasma TG has a much greater dependence

### Table 3

<table>
<thead>
<tr>
<th>Dietary unsaturated fatty acids</th>
<th>Forage concentrate</th>
<th>Hay Low-oil</th>
<th>Hay High-oil</th>
<th>Silage Low-oil</th>
<th>Silage High-oil</th>
<th>SE^a</th>
</tr>
</thead>
<tbody>
<tr>
<td>Percentage of total</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weight (g/day)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

^aStandard error of difference between two treatments.

### Table 4

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>Hay Low-oil</th>
<th>Hay High-oil</th>
<th>Silage Low-oil</th>
<th>Silage High-oil</th>
<th>Soybean oil when added to diets of</th>
</tr>
</thead>
<tbody>
<tr>
<td>16:0</td>
<td>67.3</td>
<td>85.6</td>
<td>78.9</td>
<td>87.6</td>
<td>97.3</td>
</tr>
<tr>
<td>18:0</td>
<td>91.9</td>
<td>90.5</td>
<td>95.5</td>
<td>92.1</td>
<td>90.0</td>
</tr>
<tr>
<td>18:1</td>
<td>73.0</td>
<td>90.9</td>
<td>88.6</td>
<td>92.1</td>
<td>93.7</td>
</tr>
<tr>
<td>18:2</td>
<td>69.1</td>
<td>81.9</td>
<td>70.1</td>
<td>87.8</td>
<td>88.4</td>
</tr>
<tr>
<td>Total</td>
<td>80.6</td>
<td>88.7</td>
<td>87.2</td>
<td>90.1</td>
<td>92.9</td>
</tr>
</tbody>
</table>

^aStandard error of difference between two treatments.
Figure 3. Regression of fatty acid concentration of TG in plasma on fatty acid uptake from intestine. (Standard deviation of regression coefficient.)

Figure 4. Regression of fatty acid concentration of TG-free lipid in plasma on fatty acid uptake from intestine. (Standard deviation of regression coefficient.)
on intestinal uptake than that exhibited by the other plasma lipids.

These results are consistent with the hypothesis that it is primarily the intrinsic nature of the fatty acid that determines the fatty acid composition of the TG-free lipid of the plasma, whereas it is the relative amount of each long-chain fatty acid that is absorbed from the gut that is the main determinant of plasma TG composition and, hence, of milk and depot fats of ruminants.

REFERENCES


