SHORT COMMUNICATION

Cholesterol and bile acid metabolism in obesity

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Summary
1. The present study was undertaken to determine the influence of obesity on bile acid kinetics and cholesterol balance in man.
2. Fourteen obese and normolipidaemic patients (160 ± 6% of ideal body weight, mean ± SEM) were studied under standardized dietary conditions. Bile acid kinetics were determined with the aid of 14C-labelled cholic acid and chenodeoxycholic acid. Cholesterol balance was calculated as the sum of bile acid synthesis plus daily faecal excretion of neutral C27 steroids minus dietary intake of cholesterol. The results obtained were compared with previously published data on control subjects (n = 13).
3. The cholesterol balance was higher in the obese patients (2.61 ± 0.27 mmol/day) than in the control subjects (1.78 ± 0.22 mmol/day), owing to a higher excretion of neutral steroids. When expressed per kg of body weight the cholesterol balance was quite normal in the obese patients.

Key words: bile acid, chenodeoxycholic acid, cholic acid, neutral faecal steroids, steroid balance.

Introduction
Body weight has been suggested as an important factor in the determination of total body cholesterol synthesis in man (Nestel, Whyte & Goodman, 1969; Miettinen, 1971; Nestel, Schreibman & Ahrens, 1973). Previous studies from our laboratory on normolipidaemic, normal-weight subjects were unable to demonstrate any relationship between body weight and bile acid formation or net cholesterol balance (Einarsson, Hellström & Kallner, 1974a; Angelin, Einarsson, Hellström & Kallner, 1976; Hellström & Einarsson, 1977).

The present study was undertaken to evaluate the influence of being overweight on cholesterol and bile acid turnover in man. Bile acid kinetics and cholesterol balance were studied in 14 obese normolipidaemic patients. The results obtained were compared with previously published data from normolipidaemic control subjects (Einarsson et al., 1974a; Angelin et al., 1976).

Materials and methods

Patients
The study comprised 14 obese (more than 125% of ideal weight) and normolipidaemic subjects (age, 54 ± 2 years; relative body weight, 160 ± 6%). The normal upper limits for serum cholesterol and serum triglyceride concentrations were 7.4 and 2.2 mmol/day respectively. Nine subjects were female and five male. Five of the subjects had cholelithiasis or had undergone cholecystectomy. Controls were 13 non-obese and normolipidaemic subjects, six female and seven male (age, 54 ± 3 years; relative body weight, 101 ± 3%), who had been presented previously (Einarsson et al., 1974a, Angelin et al., 1976).

Experimental procedure
The patients were hospitalized during the study. All subjects were interviewed about their dietary habits by a dietitian in order to estimate their caloric requirements. For 4–7 days before and during the experimental period they were maintained on a standardized diet of solid food with the energy intake adjusted to keep the body
weight constant. The control subjects were on the same diet (Einarsson et al., 1974a; Angelin et al., 1976). About 40% of the calories were supplied as fat, most of which contained saturated fatty acids. The major part of the carbohydrates were given as starch. The intake of cholesterol was comparatively low (on average 0.26 mmol/day in each subject). Faeces were collected for 7–10 days and stored at −20°C until analysed.

An informed consent was obtained from all subjects and research was carried out according to the Declaration of Helsinki. The ethical aspects of the study were approved by the Committee on Ethics of the Karolinska Institutet, Stockholm.

**Methods**

The pool size and turnover of cholic acid and chenodeoxycholic acid were determined by the isotope dilution technique after [24-14C]cholic acid (4 μCi) and [24-14C]chenodeoxycholic acid (4 μCi) were given perorally as described previously (Angelin, Einarsson, Hellström & Leijd, 1978). The control group received [3H]chenodeoxycholic acid instead of [14C]chenodeoxycholic acid and on account of non-specific losses of tritium, the pool size and turnover rate of chenodeoxycholic acid were overestimated by 14 ± 2 (SEM) and 17 ± 2% respectively (Einarsson, Hellström & Kallner, 1974b). In the present study, these results have been corrected to achieve values comparable with those obtained with [14C]chenodeoxycholic acid.

Neutral faecal steroids were analysed as described previously (Einarsson, Hellström & Kallner, 1974c).

Results are presented as means ± SEM and the significance of differences between mean values has been determined with Student's t-test. Correlations were tested by estimating the correlation coefficient, r (Snedecor & Cochran, 1974).

**Results**

In obese patients the pool sizes of cholic acid and chenodeoxycholic acid, as well as the combined pool size of the two bile acids, were not significantly larger than in the control subjects (Table 1 summarizes the results). This was true also after patients with gallbladder disease (gallstones or cholecystectomy) had been excluded. Neither was the synthesis of cholic acid (0.73 ± 0.13 mmol/day), chenodeoxycholic acid (0.38 ± 0.04 mmol/day) nor the total formation of bile acids significantly increased in the obese patients.

The combined faecal elimination of cholesterol and its neutral metabolites was about 60% higher in the obese patients than in the control subjects. Coprostanol was the major compound, accounting for about 85% of the neutral steroids excreted.

Neglecting the steroids excreted through the skin and in the urine, net steroid balance was calculated as bile acid synthesis plus faecal excretion of total C27 steroids minus dietary cholesterol. Results are means ± SEM; significantly different from controls, *P < 0.05 and **P < 0.01. The control data have been published previously (Einarsson et al., 1974a; Angelin et al., 1976).

**Table 1. Bile acid kinetics and cholesterol balance in control and obese subjects**

<table>
<thead>
<tr>
<th></th>
<th>Control subjects (n = 13)</th>
<th>Obese patients (n = 14)</th>
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<tbody>
<tr>
<td>Bile acid pool size (mmol)</td>
<td></td>
<td></td>
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<tr>
<td>Cholic acid</td>
<td>1.62 ± 0.22</td>
<td>2.43 ± 0.64</td>
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<tr>
<td>Cholesterol</td>
<td>1.09 ± 0.14</td>
<td>1.88 ± 0.48</td>
</tr>
<tr>
<td>Cholic acid and chenodeoxycholic acid</td>
<td>2.71 ± 0.32</td>
<td>4.31 ± 1.09</td>
</tr>
<tr>
<td>Bile acid synthesis (mmol/day)</td>
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<td></td>
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<tr>
<td>Cholic acid</td>
<td>0.67 ± 0.09</td>
<td>0.73 ± 0.13</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>0.29 ± 0.04</td>
<td>0.38 ± 0.04</td>
</tr>
<tr>
<td>Total bile acid synthesis</td>
<td>1.05 ± 0.12</td>
<td>1.11 ± 0.16</td>
</tr>
<tr>
<td>mmol/day</td>
<td>13.43 ± 1.58</td>
<td>10.89 ± 1.51</td>
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<tr>
<td>Total C27 steroids in faeces (mmol/day)</td>
<td>1.09 ± 0.14</td>
<td>1.76 ± 0.18**</td>
</tr>
<tr>
<td>μmol day⁻¹ kg⁻¹ body wt.</td>
<td>15.12 ± 1.48</td>
<td>16.64 ± 1.56</td>
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<td>Net steroid balance (mmol/day)</td>
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<tr>
<td>mmol/day</td>
<td>1.78 ± 0.22</td>
<td>2.61 ± 0.27*</td>
</tr>
<tr>
<td>μmol day⁻¹ kg⁻¹ body wt.</td>
<td>25.07 ± 2.60</td>
<td>25.02 ± 1.35</td>
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</table>
calculated as the sum of the bile acid synthesis and faecal excretion of neutral steroids (C_{27} steroids) minus cholesterol intake. Obese patients displayed significantly higher values (2.61 ± 0.27 mmol/day) than the control subjects (1.78 ± 0.22 mmol/day).

The formation of bile acids, the faecal excretion of neutral steroids and the net steroid balance were plotted against body weight. No correlations were obtained between bile acid formation and body weight. The faecal excretion of neutral steroids correlated significantly with body weight in the controls (r = 0.576, P < 0.05) and in the group comprising both controls and obese subjects (r = 0.654, P < 0.001). The net steroid balance also correlated with body weight when calculated for control subjects combined with obese patients (r = 0.530, P < 0.01). Bile acid synthesis, faecal excretion of C_{27} steroids and net steroid balance were also calculated as μmol day^{-1} kg^{-1} body wt.; values of bile acid synthesis and net steroid balance were now normal in the obese patients.

Discussion

Bile acid kinetics in the obese patients revealed values within normal limits for the synthesis of cholic acid and chenodeoxycholic acid as well as for the total formation of bile acids. The ratio between the synthesis of the two bile acids averaged 1.9, which is quite normal. Several of the obese patients displayed enlarged bile acid pool sizes, a finding also reported by other investigators (Bennion & Grundy, 1975; Mok, von Bergmann & Grundy, 1977; Shaffer & Small, 1977), but the mean values of the pool sizes of cholic acid and chenodeoxycholic acid were not significantly different from those of the control subjects. The net steroid balance increased on average by 54% in the obese patients, but when calculated as μmol day^{-1} kg^{-1} body weight it was normal. This suggests that obese subjects represent an extension of the normal weight population with respect to cholesterol synthesis. The increased cholesterol turnover in obesity was due to an enhanced faecal excretion of neutral steroids.

The present results confirm previous findings of an increased production of cholesterol in obesity (Miettinen, 1971; Nestel et al., 1973). Miettinen (1971) performed sterol balance studies in obese normolipidaemic patients with an average body weight of 109 kg and found a mean cholesterol synthesis double that of lean subjects (mean weight 61 kg). Sterol balance, when expressed per kg of body weight, was no longer higher in the obese patients than in the control subjects. Similarly Nestel et al. (1973) demonstrated with both the sterol balance technique and compartmental analysis that grossly obese but normolipidaemic patients (mean weight 141 kg) had cholesterol turnover values of more than twice those found in non-obese subjects. These two groups found that the main increment in faecal steroid excretion in the obese patients appeared in the neutral steroid fraction but they also noted an increase in bile acid excretion. This latter finding is in disagreement with our results but may be explained by differences in dietary management.

The mechanisms responsible for the increased synthesis of cholesterol in obesity are not known. The possibility that cholesterol synthesis in adipose tissue may contribute a substantial portion of the total body cholesterol turnover has gained some support from data in vitro (Kekki, Miettinen & Wahlström, 1977). However, studies in vitro have suggested that cholesterol synthesis in adipose tissue is negligible and that the excess formation of cholesterol in obesity occurs in the liver (Schreibman & Dell, 1975).

Hyperlipoproteinaemia type IV is also often associated with an enhanced formation of bile acids and an increased net cholesterol balance (Einarssson et al., 1974a; Angelin et al., 1976; Hellström & Einarssson, 1977). Although these patients are often slightly overweight, the cholesterol balance remains elevated even after correction for body weight (Angelin et al., 1976), which is obviously in contrast to the findings in obesity.

It is apparent from the present study that cholesterol production in obesity is increased and that cholesterol is excreted from the body preferentially as neutral steroids. Accordingly, obese patients have an elevated secretion of biliary cholesterol, resulting in a bile supersaturated with cholesterol (Bennion & Grundy, 1975; Shaffer & Small, 1977). In hyperlipoproteinaemia type IV, on the other hand, the excess of cholesterol is preferentially converted into bile acids in the liver. Nevertheless, most patients with type IV hyperlipoproteinaemia also have supersaturated bile (Ahlberg, Angelin, Einarssson, Hellström & Leijd, 1980). Previous investigations have given evidence for a certain compartmentalization of liver cholesterol in man (Schwartz, Berman, Vlahcevic, Halloran, Gregory & Swell, 1978; Einarssson, Ahlberg, Angelin & Holmström, 1979). Thus bile acids are to a larger extent than biliary cholesterol derived from newly synthesized cholesterol. Such a compartmentalization may be one possible expla-
nation for the different metabolism of excess cholesterol in obesity and type IV hyperlipoproteinaemia. Another possibility is that further metabolism of excess cholesterol is determined by the ratio between the rate-determining enzymic step of bile acid biosynthesis (7α-hydroxylase) and that of cholesterol synthesis (3-hydroxy-3-methylglutaryl-CoA reductase). It may be speculated that in patients with type IV hyperlipoproteinaemia this ratio is increased compared with that in obese subjects.

Acknowledgments
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References