Effect of diets low and high in refined sugars on gut transit, bile acid metabolism, and bacterial fermentation

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Abstract

Increasing consumption of refined sugar has been implicated in many gastrointestinal disorders on epidemiological grounds. Nine volunteers agreed to participate in a study comparing the effects of a diet containing 165 g refined sugar/day with a diet of only 60 g/day on gut transit, bile acid metabolism, and fermentative activity of the intestinal flora. The wet and dry weight, pH, and water content of the stools were similar on the two diets. On the high sugar diet mouth-to-anus transit time was significantly prolonged, despite a shortened mouth-to-caecum transit time. The faecal concentration of total bile acids and the faecal concentration of secondary bile acids increased significantly. Diet affected neither the serum bile acid pattern nor the concentration. Breath hydrogen tests showed significantly enhanced H_2 production on the high sugar diet. We conclude that the quantity of refined sugar in the diet can significantly influence gut function and the composition of bowel contents.

It has been suggested that changing eating habits in industrialised countries play a part in the pathophysiology of some common gastrointestinal diseases such as the formation of gall stones,[1] colonic cancer,[2] irritable bowel syndrome,[3] diverticulosis of the colon,[4] and inflammatory bowel disease.[5] One of the changes in our nutrition is the increasing refined sugar intake. In the 100 years from 1870 to 1970 sugar consumption doubled.[6] The average annual intake of sugar per capita is 35 kg in the Federal Republic of Germany, 50 kg in Britain and Australia, but only 20 kg world wide.[7]

A diet containing refined foods results in excess energy intake – that is, it is fattening.[8] Average calorie intake and obesity are related to the incidence of large bowel cancer[9,10] and gall stones.[11] In particular, high consumption of refined sugar is associated with the development of colorectal cancer,[12] the formation of gall stones,[13] and Crohn's disease.[14]

The ability, however, of epidemiological data to answer pathophysiological questions is limited. Experimental data on the effects of refined sugar on stool characteristics, gastrointestinal transit, bacterial fermentation, and bile acid metabolism are rare. We therefore designed a study to try to determine the influence of diets that are low and high in refined sugar on those functions of the gut.

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Methods

Nine healthy volunteers (five women, median age 24 years, range 23–26 years) gave informed consent and were included in the study, which was approved by the Ethics Committee of the Klinikum Grosshadern, University of Munich. All subjects were on a typical Western diet. Before entering the study they ingested 20 g Glaubers salt (sodium sulphate) to attain microbiological steady state conditions in the intestines as quickly as possible. Thereafter they took only 2000 ml/day of a commercially available formula diet (Fresubin plus, Fresenius AG, Oberursel, FRG, ingredients as listed in Table I), with the exception that additional water, tea, and black coffee were allowed ad libitum. At the end of the 14 day control period marker studies, breath hydrogen tests, 48 hour stool collections, and fasting venous blood sampling were performed. The volunteers began the 14 day study period by again taking 20 g Glaubers salt. The diet now consisted of the same formula diet as in the control period, but with the addition of 120 g of refined sugar/day, thus increasing the total refined sugar content from 60 g/day to 165 g/day. To keep the diets roughly isocaloric, the total amount of formula diet was restricted to 1500 ml/day during the study period. The mean (SEM) weight of the subjects was similar during the control period (64·1 (5·2) kg) and the study period (63·5 (5·1) kg). At the end of the study period marker studies, breath hydrogen tests, and stool and blood collections were again carried out.

STOOL COLLECTION, WEIGHT, WATER, AND PH

Stools were collected for 48 hours urine free in plastic containers. Immediately after defecation stool pH was determined by a pH electrode, the wet stools were weighed, and 100 ml of 70% isopropanol was added to inhibit bacterial degradation of bile acids. The stools were stored at −20°C.

MARKER STUDIES

Transit through the whole gut was assessed by means of the single stool technique according to Cummings and Wiggins.[15] The three different marker pellets (spheres, rings, and 'B'-formed markers) had a mean (SEM) weight of 30 (1) mg.

BREATH HYDROGEN TESTS

All subjects were studied in the morning at the

same time after a 12 hour overnight fast; 20 g lactulose in 300 ml water was given by mouth. End-expiratory hydrogen concentrations were measured using a H_2 sensitive electrode (GMI-monitor, Scotland). Breath samples were collected at 5 minute intervals by the single breath technique for at least 120 minutes. Mouth-to-caecum transit was defined by the first rise of breath hydrogen above two standard deviations of baseline fluctuations. Fasting and maximum H_2 production were recorded and H_2 production quantified by calculating the area under the curve over two hours.[16] For no apparent reason one subject did not respond to lactulose when she was on a high sugar diet.

DETERMINATION OF FAECAL BILE ACIDS

Stool samples were vacuum dried at 120°C and weighed (stool dry weight). The difference between wet and dry weight was defined as the stool water content. Further steps in stool preparation (50 mg) included heating with alkaline methanol (80°C), centrifugation, extraction of bile acids with Bond Elute C18 cartridges,[17] methylation, and silylation.[17] Analysis of the bile acid methylester trimethylsilane ether derivatives was performed by capillary gas chromatography on a 25 m×0·32 mm CP Sil 19 CB column (Chrompack, Middelburg, The Netherlands). Analysis conditions have been described previously.[17 18]

DETERMINATION OF SERUM BILE ACIDS

Individual conjugated and unconjugated fasting serum bile acids were analysed quantitatively using recently described methods applying group separation of conjugated and unconjugated bile acids on DEAP-LH-20 anion exchange columns and capillary gas chromatography.[18 19]

STATISTICS

Results are given as median (range). Differences were assessed by the Wilcoxon matched pairs signed ranks test. A p value of $<0·05$ was considered significant.

Results

GENERAL FAECAL CHARACTERISTICS

No significant differences were observed in faecal wet and dry weight, in faecal water content, and in faecal pH on the different diets. Faecal wet weight was 66 (29–93) g/24 h *v* 44 (22–161) g/24 h (low sugar diet *v* high sugar diet, faecal dry weight was 26 (12–37) g/24 h *v* 15 (9–65) g/24 h, faecal water content was 66 (42–72) % *v* 60 (42–73) %, and faecal pH was 6·90 (6·40–7·05) *v* 6·90 (6·25–7·55).

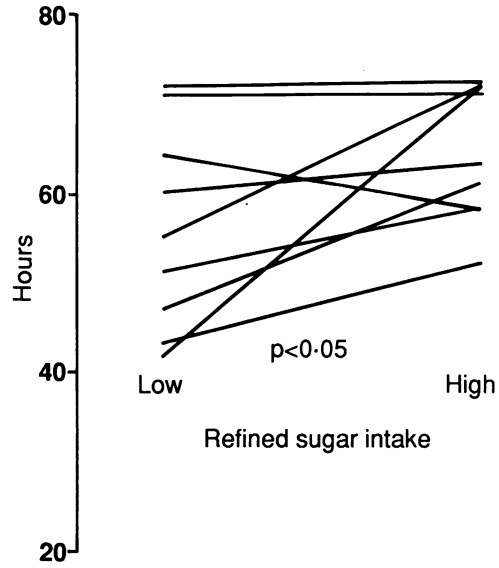

Figure 1: Transit time through the gut in nine healthy subjects taking diets low and high in refined sugar.

INTESTINAL TRANSIT AND HYDROGEN BREATH TESTS

The mean transit time increased significantly ($p<0·05$) from 55 (42–72) h in the low sugar period to 63 (53–72) h during the high sugar period (Fig 1). Transit time from mouth to caecum (Fig 2) as measured by the breath hydrogen test was 27 (15–70) min on the low sugar diet and 15 (3–65) min on the high sugar diet ($p<0·01$). Bacterial hydrogen production was increased ($p<0·05$) on the high sugar diet as assessed by the calculation of the area under the curve (Fig 3, Table II). Neither maximum end-expiratory hydrogen concentration nor fasting breath hydrogen concentration was influenced by the different diets (Table II).

FAECAL BILE ACIDS

Total bile acid excretion was simiar on the low (132 (99–279) mg/24 h) and high sugar diets (122 (60–470) mg/24 h). But faecal total bile acid concentration expressed as mg bile acids per g stool dry weight increased significantly ($p<0·05$)

TABLE I *Composition of the formula diet used in the study, which contained all necessary vitamins, minerals, and electrolytes*

Ingredient	*Amount per litre*
Protein	38 g
Total fat	34 g
Essential fatty acids	10 g
Total carbohydrates	138 g
Sucrose	30 g
Fibre (soluble/insoluble = 1/3; cellulose 45%, hemicellulose 35%, lignin 10%, pectin 10%)	20 g
Energy	0·418 MJ
Osmolality	35 mOsmol/l

TABLE II *Results of the breath hydrogen test in eight healthy subjects during low and high refined sugar diet (median and range)*

	Refined sugar intake		
	Low	*High*	
Orocaecal transit time (min)	27 (15–70)	p<0·01	15 (3–65)
Fasting H_2–concentration (ppm)	10 (4–30)		8 (4–20)
Maximum H_2–concentration (ppm)	156 (30–300)		150 (90–355)
Area under the H_2–exhalation curve (ppm×h)	114 (36–298)	p<0·05	123 (22–403)

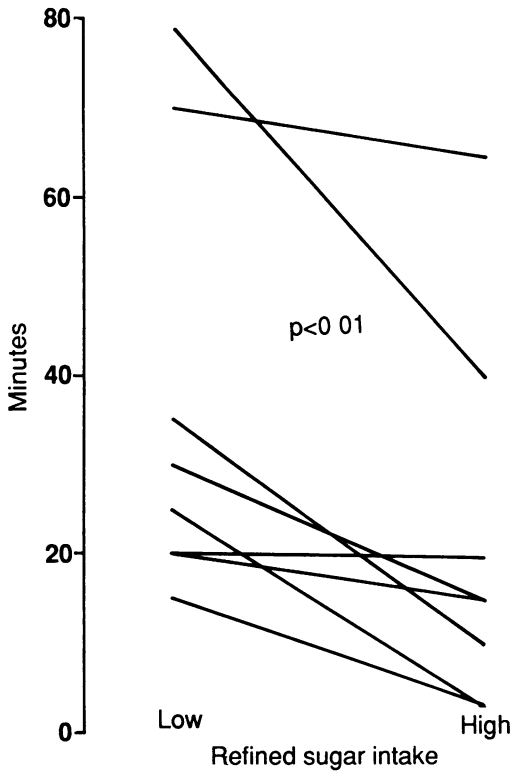

Figure 2: Mouth-to-caecum transit time in eight healthy subjects taking diets low and high in refined sugar.

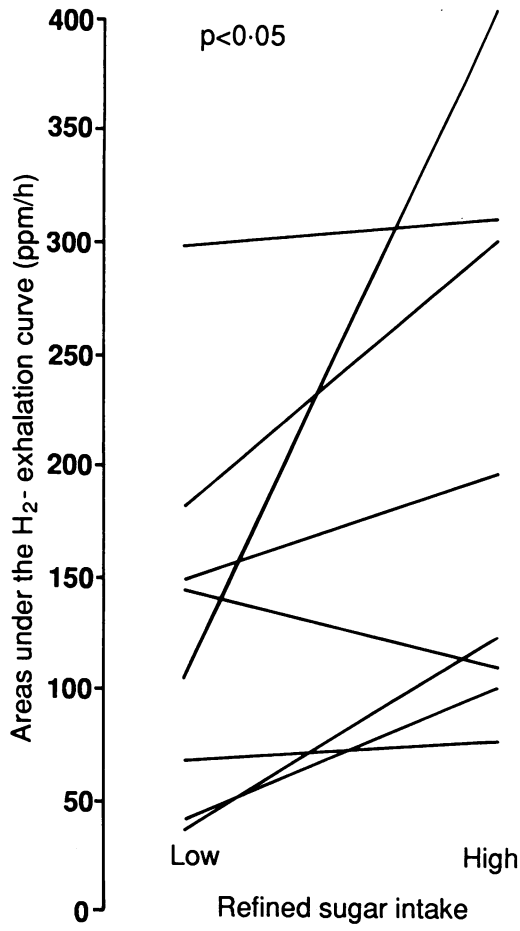

Figure 3: Area under the breath hydrogen curve after taking 20 g oral lactulose on diets low and high in refined sugar.

from 6·4 (4·0–10·1) mg/g on the low sugar diet to 7·2 (4·9–10·8) mg/g on the high sugar diet. This increase was due to a significant ($p<0·05$) change of faecal lithocholic acid from 2·6 (1·5–3·7) mg/g to 3·2 (1·2–6·3) mg/g. Faecal deoxycholic acid was 3·3 (2·0-5·4) mg/g *v* 3·6 (2·7–4·3) mg/g. As Figure 4 shows there was a significant ($p<0·05$) increase in the faecal content of secondary bile acids from 6·0 (3·5–8·9) mg/g to 6·6 (4·2–10·7) mg/g. The faecal content of primary bile acids did not change significantly: cholic acid 0·05 (0·05–0·44) mg/g *v* 0·05 (0·05–0·65) mg/g; chenodeoxycholic acid 0·47 (0·17–0·59) mg/g *v* 0·35 (0·05–0·91) mg/g. The faecal content of ursodeoxycholic acid (0·05 (0·05–0·4) mg/g *v* 0·05 (0·05–0·17) mg/g) remained unchanged.

SERUM BILE ACIDS

The serum concentration of conjugated cholic acid dropped significantly ($p<0·05$) during the high sugar period. Otherwise the serum concentrations of total, conjugated, and unconjugated bile acids were not influenced by the change in diet. The data are summarised in Table III.

TABLE III *Serum bile acid concentration in nine healthy subjects on diets low and high in refined sugar (median and range)*

	Refined sugar intake (μmol/l)	
	Low	*High*
Total bile acids:	2·11 (0·55–2·51)	1·40 (0·80–2·94)
Cholic acid	0·13 (0·04–0·45)	0·09 (0·04–0·22)
Chenodeoxycholic acid	0·44 (0·04–0·90)	0·45 (0·17–0·80)
Deoxycholic acid	0·49 (0·30–0·79)	0·37 (0·13–1·20)
Conjugated total bile acids:	0·96 (0·45–1·34)	0·75 (0·46–1·26)
Cholic acid	0·10 (0·02–0·19)	0·02 (0·02–0·08)*
Chenodeoxycholic acid	0·28 (0·02–0·50)	0·26 (0·15–0·45)
Deoxycholic acid	0·28 (0·18–0·42)	0·25 (0·11–0·48)
Unconjugated total bile acids:	0·91 (0·10–1·65)	0·60 (0·10–1·93)
Cholic acid	0·02 (0·02–0·28)	0·02 (0·02–0·20)
Chenodeoxycholic acid	0·16 (0·02–0·54)	0·17 (0·02–0·46)
Deoxycholic acid	0·18 (0·02–0·58)	0·23 (0·02–0·79)

*$p<0·05$.

Discussion

When studying the physiological effects of a single nutrient it is essential that the rest of the diet is kept constant and that experimental periods are long enough to allow for steady state conditions. It is also desirable that the composition of the diet resembles that of the subject's normal diet. We used a formula diet produced according to the suggestions of the DGE (German Association of Nutrition)[20] for the nutrition of adults. The fibre content of this formula diet is 20 g/2000 ml, reflecting the average daily fibre consumption in industrialised countries.[21] In our study the different diet periods lasted for 14 days, which has been found[22] to result in steady state conditions for faecal excretion of sterols and bile acids. Feeding chemically defined diets led to microbiological steady state conditions after 13 days,[23] which could be shortened to four days by giving laxatives between the different diet periods.[24] The control diet included 60 g/day of refined sugar, which is similar to the average sugar intake world wide,[7] while the high sugar diet comprised 165 g/day, as in some Western countries.[7]

Transit through the whole gut slowed down on the high sugar diet. This change may have been

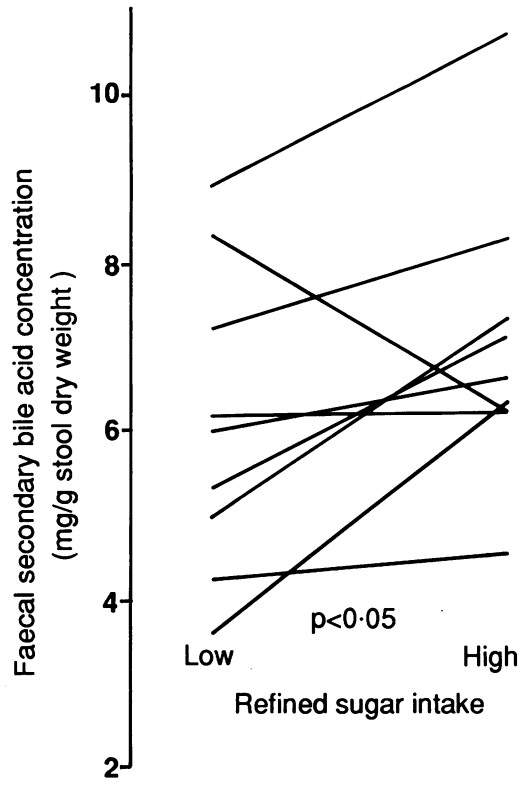

Figure 4: Faecal secondary bile acid concentration in nine healthy subjects taking diets low and high in refined sugar.

influenced by the fibre content of the diets. In fact there are conflicting results on the effect of fibre ingestion on gastrointestinal transit, either causing acceleration[25 26] or showing no effects.[27 28] In our study during the high sugar period fibre content was diminished by 5 g/day; this amount of fibre has not yet been proved to alter bowel transit. The weight and water content of the stools were not significantly different during the two diet periods. Thus the prolongation of whole gut transit time was probably caused by the high sugar intake, though the mechanism is obscure.

In contrast to transit through the whole gut, mouth-to-caecum transit, calculated from the increments in breath hydrogen, was accelerated. Increased hydrogen production is due to bacterial fermentation of lactulose in the caecum. In cases with bacterial overgrowth, however, it may also be caused by fermentation in the upper gut. An early peak of breath hydrogen followed by a prolonged increase in hydrogen corresponding to the passage of lactulose into the caecum is characteristic for bacterial growth of the proximal small bowel.[29] We did not observe this pattern of hydrogen exhalation and, in addition, fasting breath hydrogen concentration, another marker of bacterial growth of the intestine,[30] was not raised. Analysis of the area under the breath hydrogen curve showed an increase on the high sugar diet, indicating altered colonic bacterial fermentation.[16] Thus breath hydrogen testing in subjects on a high sugar diet showed faster small bowel transit and raised fermentative bacterial activity in the colon.

One of the many enzymatic actions of intestinal bacteria is 7-α-dehydroxylation of primary to secondary bile acids. Altered bowel flora may lead to an increase of faecal secondary bile acids,[31] as we observed on the high sugar diet. Bacterial conversion from primary to secondary bile acids depends on the transit time.[32] Thus the lengthened whole gut transit time on the high sugar diet may also favour an increase of secondary bile acids. The raised faecal secondary bile acid concentration was not accompanied by a decrease in primary bile acids, but it was associated with significantly higher faecal concentration of total bile acids on the high sugar diet. In previous studies[33 34] pool sizes tended to enlarge on a diet with refined sugar. Bile acids that are deconjugated in the upper small bowel by gut bacteria can easily pass into the portal blood and therefore be recognised in the systemic circulation.[18] Indeed, raised unconjugated serum bile acids have been described[18] in patients with intestinal bacterial overgrowth. We observed no changes in the serum concentration of unconjugated bile acids, which further supports the breath hydrogen findings of a bacterial growth of the small bowel not affected by the intake of refined sugar.

This study shows significant changes of gut transit, of fermentative colonic bacterial activity, and of intestinal bile acids in healthy subjects fed a diet high in refined sugar. Whether those alterations have a patho-physiological role is not yet clear but has been suggested in epidemiological studies.

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1 Heaton KW. Diet and gallstones. In: Fisher MM, Goresky CA, Schaffer EA, Strasberg SM, eds. *Gallstones*. New York: Plenum Press, 1979: 371–89.
2 Burkitt DP. Epidemiology of cancer of the colon and rectum. *Cancer* 1971; **28**: 3–13.
3 Thompson WG. The irritable bowel. *Gut* 1984; **25**: 305–20.
4 Painter NS, Burkitt DP. Diverticular disease of the colon: deficiency disease of Western civilisation. *BMJ* 1971; ii: 450–4.
5 Sachar DB, Auslander MO, Walfish JS. Aetiological theories of inflammatory bowel disease. *Clin Gastroenterol* 1980; **9**: 231–57.
6 Trowell HC. Refined carbohydrate foods and fibre. In: Burkitt DP, Trowell HC, eds. *Refined carbohydrates: food and disease*. Oxford: Academic Press, 1975: 23–41.
7 Food and Agricultural Organisation. *Food balance sheets 1975–1977 average*. Rome: FAO, 1980.
8 Heaton KW. Dietary factors. In: Truelove SC, Lee E, eds. *Cancer of the large bowel. Topics in gastroenterology*. Oxford: Blackwell, 1977; 5: 29–44.
9 Armstrong B, Doll R. Environmental factors and cancer incidence and mortality in different countries, with special reference to dietary practices. *Int J Cancer* 1975; **15**: 617–31.
10 Garfinkel L. Overweight and cancer. *Ann Intern Med* 1985; **103**: 1034–6.
11 Friedman GD, Kannel WB, Dawber TR. The epidemiology of gall bladder disease. Observations in the Framingham study. *J Chron Dis* 1966; **19**: 273–92.
12 Bristol JB, Emmett PM, Heaton KW, Williamson RCN. Sugar, fat, and the risk of colorectal cancer. *BMJ* 1985; **291**: 1467–70.
13 Scragg RKR, McMichael AJ, Baghurst PA. Diet, alcohol, and relative weight in gall stone disease: a case-control study. *BMJ* 1984; **288**: 1113–9.
14 Martini GA, Brandes JW. Increased consumption of refined carbohydrates in patients with Crohn's disease. *Klin Wochenschr* 1976; **54**: 367–71.
15 Cummings JH, Wiggins HS. Transit through the gut measured by analysis of a single stool. *Gut* 1976; **17**: 219–33.
16 Bond JH, Levitt MD. Use of pulmonary hydrogen (H_2) measurements to quantitate carbohydrate absorption. *J Clin Invest* 1972; **51**: 1219–25.
17 Stellaard F, Sackmann M, Sauerbruch T, Paumgartner G. Simultaneous determination of cholic acid and chenodeoxycholic acid pool sizes and fractional turnover rates in human serum using ^{13}C labelled bile acids. *J Lipid Res* 1984; **25**: 1313–9.
18 Stellaard F, Sauerbruch T, Luderschmidt Ch, Leisner B, Paumgartner G, Intestinal involvement in progressive systemic sclerosis detected by increased unconjugated serum bile acids. *Gut* 1987; **28**: 446–50.
19 Schindlbeck NE, Heinrich C, Stellaard F, Paumgartner G,

Müller-Lissner SA. Healthy controls have as much bile reflux as gastric ulcer patients. *Gut* 1987; **28:** 1577–83.
20 Deutsche Gesellschaft für Ernährung e.V. Ernährungsbericht. Frankfurt, 1984: 20.
21 Southgate DAT, Bingham S, Robertson J. Dietary fibre in the British diet. *Nature* 1978; **274:** 51–2.
22 Moore RB, Anderson JT, Taylor HL, Keys A, Frantz ID. Effect of dietary fat on the fecal excretion of cholesterol and its degradation products in man. *J Clin Invest* 1968; **47:** 1517–34.
23 Connor WE, Wittiak DT, Stone DB, Armstrong ML. Cholesterol balance and fecal neutral steroid and bile acid excretion in normal men fed dietary fats of different fatty acid composition. *J Clin Invest* 1969; **48:** 1363–75.
24 Winitz M, Adams RF, Seedman DA, Davis PN, Jayko LG, Hamilton JA. Studies in metabolic nutrition employing chemically defined diets. II. Effects on gut microflora populations. *Am J Clin Nutr* 1970; **23:** 546–59.
25 Cummings JH, Jenkins DJA, Wiggins HS. Measurement of the mean transit time of dietary residue through the human gut. *Gut* 1976; **17:** 210–8.
26 Baird JM, Walters RL, Davies PS, Hill MJ, Drasar BS, Southgate DAT. The effects of two dietary fiber supplements of gastrointestinal transit, stool weight and frequency, and bacterial flora, and fecal bile acids in normal subjects. *Metabolism* 1977; **26:** 117–28.
27 Eastwood MA, Kirkpatrick JR, Mitchell WD, Bone A, Hamilton T. Effects of dietary supplements of wheat bran and cellulose on faeces and bowel function. *BMJ* 1973; iv: 392–4.
28 Metcalf AM, Phillips SF, Zinsmeister AR, MacCarty RL, Beart RW, Wolff BG. Simplified assessment of segmental colonic transit. *Gastroenterology* 1987; **92:** 40–7.
29 Rhodes JM, Middelton P, Jewell DP. The lactulose hydrogen breath test as a diagnostic test for small bowel bacterial overgrowth. *Scand J Gastroenterol* 1979; **14:** 335–6.
30 Perman JA, Modler S, Barr RG, Rosenthal P. Fasting breath hydrogen concentration: normal values and clinical application. *Gastroenterology* 1984; **87:** 1358–63.
31 Carey MC. The enterohepatic circulation. In: Arias I, Popper H, Schachter D, Shafriz DA, eds. *The liver: biology and pathobiology*. New York: Raven Press, 1982: 429–65.
32 Garbutt JT, Wilkins RM, Lack L, *et al*. Bacterial modification of taurocholate during enterohepatic recirculation in normal man and patients with small intestinal disease. *Gastroenterology* 1970; **59:** 553–66.
33 Werner D, Emmett PM, Heaton KW. Effects of dietary sucrose on factors influencing cholesterol gall stone formation. *Gut* 1984; **25:** 269–74.
34 Hepner GW. Effect of decreased gallbladder stimulation on enterohepatic cycling and kinetics of bile acids. *Gastroenterology* 1975; **68:** 1574–81.