

Altered postprandial glucose, insulin, leptin, and ghrelin in liver cirrhosis: correlations with energy intake and resting energy expenditure¹⁻³

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ABSTRACT

Background: Liver cirrhosis is associated with reduced energy intake and increased resting energy expenditure.

Objective: We aimed to investigate the possible role of glucose, insulin, leptin, and ghrelin in the pathogenesis of these alterations.

Design: Nutritional status, energy intake, resting energy expenditure, and fasting glucose, insulin, and leptin were assessed in 31 patients with cirrhosis. Postprandial glucose, insulin, C-peptide, leptin, and ghrelin responses were studied in a subgroup of patients after a standard meal. Ten healthy subjects served as controls.

Results: Patients with cirrhosis had a lower energy intake ($P < 0.05$), higher resting energy expenditure ($P < 0.05$), higher fasting leptin ($P < 0.05$), and higher insulin resistance ($P < 0.001$) than did the healthy control subjects. In the patients with cirrhosis, fasting leptin was negatively correlated with resting energy expenditure ($r = -0.38$, $P < 0.05$) but not with energy intake. In control subjects, leptin was negatively correlated with energy intake ($r = -0.72$, $P < 0.05$) but not with resting energy expenditure. The patients with cirrhosis had higher postprandial glucose ($P < 0.001$) and lower ghrelin ($P < 0.05$) concentrations at 4 h postprandially than did the control subjects. The increase in ghrelin from its minimal postmeal value to 4 h postmeal was negatively correlated ($r = -0.66$, $P = 0.014$) with weight loss in the patients with cirrhosis. Energy intake was negatively correlated ($r = -0.42$, $P < 0.01$) with the postprandial increase in glucose.

Conclusions: In cirrhosis, altered postprandial glucose and ghrelin are associated with reduced energy intake and weight loss, respectively, and the effects of leptin on energy intake and expenditure seem to be altered. Insulin resistance might be involved in these altered postprandial responses. *Am J Clin Nutr* 2007;85:808–15.

KEY WORDS Glucose, insulin, leptin, ghrelin, insulin resistance, energy intake, resting energy expenditure, liver cirrhosis, malnutrition

INTRODUCTION

Malnutrition is common in patients with liver cirrhosis, with a reported prevalence as high as 80% depending on the severity of liver disease (1–3). The mechanisms of malnutrition in cirrhosis are not completely understood. Both poor dietary intake (3–5) and increased basal energy expenditure have been reported to contribute to a negative energy balance in patients with cirrhosis (1, 6–11).

Insulin resistance is common in patients with cirrhosis (3, 7, 12) and is possibly associated with impairment of nutritional status (12). An elevated postprandial insulin concentration has been proposed as a factor that induces satiety and a subsequent reduction in energy intake in liver cirrhosis (12). However, the relation of postprandial hyperglycemia to energy intake, which has been shown to occur in cirrhosis (12), is unexplored in this group of patients.

Leptin and ghrelin are known to influence energy expenditure and energy intake in humans (13). Leptin circulates in free and bound form, and it has been shown to suppress energy intake and stimulate energy expenditure, whereas ghrelin has been shown to rise before a meal thus enhancing appetite and food intake (13). The basal concentrations of leptin and ghrelin have been reported to be deranged in liver cirrhosis (5, 9, 14–19), but only few studies are available on the relations of leptin and ghrelin to energy intake and resting energy expenditure (REE) in these patients (5, 18, 19). In a previous report, no correlation was found between total leptin concentration and REE in patients with cirrhosis with adequate food intake (18). Also, bound (but not free) leptin was shown to be increased and positively correlated with REE in patients with postviral cirrhosis on a weight-maintaining diet (9). To our knowledge, the relation of leptin to spontaneous energy intake and REE in patients with cirrhosis of various etiologies has not been previously investigated. Also, data are lacking on postprandial changes in leptin and ghrelin in patients with cirrhosis.

Insulin has been reported to be essential for meal-induced ghrelin suppression (20–22) and to acutely increase leptin in healthy persons (23). An inverse relation between leptin and ghrelin has been observed, and it has been proposed that leptin could be of importance for suppression of basal ghrelin in normoinsulinemic subjects (24). Thus, to study the potential importance of these hormones for energy intake and REE, they need to

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be investigated together, a study not previously undertaken in liver cirrhosis.

The main aim of the current study was to investigate the relation of basal and postprandial concentrations of plasma glucose, insulin, leptin, and ghrelin to energy intake and REE. A secondary aim was to study the interrelations of postprandial plasma glucose, insulin, leptin, and ghrelin in patients with cirrhosis.

SUBJECTS AND METHODS

Thirty-one consecutive patients with liver cirrhosis attending the outpatient clinic of the Department of Internal Medicine at Sahlgrenska University Hospital, Gothenburg, Sweden, were enrolled in the study. The diagnosis of liver cirrhosis was established histologically; on the basis of its clinical, laboratory, endoscopic, or imaging features; or both. The severity of liver disease was assessed according to the Child-Pugh and the Model for End Stage Liver Disease scores (25). Patients with malignancy, infections, known gastrointestinal or renal disease, significant respiratory or cardiac dysfunction, insulin-dependent diabetes mellitus, hepatorenal syndrome, untreated thyroid dysfunction, and hepatic encephalopathy grade II–IV were excluded. Patients with alcoholic cirrhosis had been abstinent for ≥ 6 mo at inclusion. All had normal serum creatinine and had undergone gastroscopy in the previous 6 mo. Twenty-six of the 31 patients had endoscopic evidence of esophageal varices, and 20 of the 31 had evidence of portal hypertensive gastropathy. None of the patients had macroscopic evidence of gastric mucosal atrophy. Two patients were found to have diabetes mellitus on blood sampling for purposes of this study. Six patients had mild ascites detectable by ultrasonography at inclusion and were treated with spironolactone. None had peripheral edema. Ten age-, sex-, and body mass index (BMI)-matched healthy weight-stable volunteers, mainly health-care professionals, acted as controls. Most of them had participated in several studies as healthy volunteers before, none was taking any medications, none was obese, all denied alcohol overconsumption, and all had normal liver function tests. The study was approved by the ethics committee of the University of Gothenburg and informed consent was obtained from all subjects.

Assessment of nutritional status

Weight was measured without shoes and in light clothing. Of 6 patients with mild ascites, every effort was made to calculate dry weight, which is defined as body weight after taking into consideration water overload. The dry weight was considered equal to the current weight if no ascites was present. In patients with ascites, a review of the patient files was performed to find data on weight after last paracentesis or before recent ascites development. BMI was calculated and weight change that could not be explained by ascites or edema during the previous 6 mo was noted. Dry weight loss was expressed as a percentage of actual body weight. Skinfold thickness at the tricep, bicep, subscapular, and supriliac sites as well as midarm muscle circumference were measured 3 times by the same research dietitian, and the mean value was used. The sum of the tricep, bicep, subscapular, and supriliac skinfolds was used to assess percentage body fat according to previously published age- and sex-specific tables (26). This method has been shown to have comparable results with dual energy X-ray absorptiometry in patients with cirrhosis without overt fluid retention (27). Fat-free mass

(FFM) was calculated as body weight minus fat mass. Patients were considered malnourished when the triceps skinfold thickness, midarm muscle circumference, or both were below the 5th percentile, according to standard tables for the Swedish population based on age and sex (28), or if BMI (in kg/m^2) was < 18.5 .

Dietary intake

To assess the subjects' dietary intake, a 4-d food diary was used as previously described (29). Total daily energy intake is reported in absolute amounts, as a ratio of body weight in kg (energy intake:body weight), and as a ratio of REE (energy intake:REE).

Indirect calorimetry

REE was determined for all subjects in the morning after an overnight fast (10 h) by indirect calorimetry (Deltatrac; Datex, Helsinki, Finland) from 0730 to 0830. To compare REE between the different groups, REE was adjusted for FFM by the use of a linear regression model. Adjusted REE was calculated as the group median REE plus measured REE minus predicted REE, where group median REE is the median absolute REE, measured REE is the metabolic rate measured in each subject, and predicted REE is the calculated rate obtained by using the individual FFM in the linear regression equation generated from the cirrhotic or control group as appropriate (30). Hypermetabolism was defined as a ratio of measured REE to predicted REE > 1.1 (29).

Test meal

On another day, about one week apart from indirect calorimetry, from 0730 to 0800 after an overnight fast, a subgroup of 18 patients with cirrhosis (group A) and all healthy control subjects had a 480 kcal test meal of oatmeal porridge and one cheese sandwich with set amounts of macronutrients (55% of energy as carbohydrate, 31% of energy as fat, and 14% of energy as protein). The test meal is a common kind of breakfast in Scandinavia. The subjects were instructed to eat the meal within 10 min. Blood samples for serum insulin, plasma glucose, and serum C-peptide measurements were drawn from an indwelling cannula at baseline and at 30 min, 60 min, 90 min, 2 h, and 4 h after the meal. In a subgroup of group A—13 patients with cirrhosis (group B)—and all healthy control subjects blood samples were also drawn for plasma leptin and ghrelin analysis at the same intervals.

Blood sample analysis

Blood samples for glucose, insulin, and leptin were drawn after an overnight fast on the day of the test meal from subjects who participated in this part of the study and on the day of indirect calorimetry from all others. Insulin resistance was expressed as homeostasis model assessment index (HOMA-IR) (31). Plasma was immediately separated by centrifugation for 5 min at $1000 \times g$ (4°C) and then stored at -80°C until subsequent leptin, ghrelin, or C-peptide analysis. Plasma total ghrelin concentrations were measured by commercial RIA (Linco Research Inc, St Louis, MO) by using ^{125}I -labeled ghrelin as a tracer and ghrelin antiserum specific for total ghrelin. The detection limit for the assay was 93 pg/mL. Ghrelin was expressed in absolute values. Plasma leptin concentrations were measured by using a commercial enzyme-linked immunosorbent assay (Quantikine human leptin, R&D Systems, Oxford, United Kingdom). The



TABLE 1
Basic characteristics in all subjects¹

	Group A ³						Healthy control subjects (n = 10)
	All cirrhotic patients (n = 31)	<i>P</i> ²	All patients with cirrhosis in group A (n = 18)	<i>P</i> ²	Group B ⁴ (n = 13)	<i>P</i> ²	
Age (y)	57 (51–63) ⁵	0.41	57 (52–63)	0.62	56 (48–62)	0.93	54 (49–63)
Sex (M/F)	18/13	0.91	11/7	0.95	11/2	0.18	6/4
Weight	77 (70–88)	0.92	83 (70–89)	0.69	86 (76–91)	0.17	77 (72–84)
BMI	26.3 (24.3–29.3)	0.60	26.5 (24.7–29.3)	0.52	26.5 (24.4–29.7)	0.61	25.7 (24.1–27.2)
Fat ⁶							
(%)	36.2 (31.2–40.3)	0.11	38.2 (34.8–46.9)	0.04	38.0 (28.3–49.6)	0.11	31.1 (26.8–37.9)
(kg)	26.9 (23–35)	0.23	29.4 (23.5–41.2)	0.10	33.8 (23.5–43.1)	0.07	25.2 (20.2–29.1)
MAMC (cm)	24.7 (21.6–27.7)	0.10	23.6 (21.3–26.3)	0.04	24.0 (21.2–27.6)	0.10	27.1 (24.0–28.5)
Diabetes (n)	2	0.41	1	0.45	1	0.37	0
Weight loss (%) ⁷	0 (0–1.3)		0 (–5 to 0)		0 (–5.8 to 0)		
Malnutrition (n) ⁸	5		2		2		
Etiology (n)							
Alcoholic	13		6		5		
Viral	5		2		1		
PBC	4		4		1		
Cryptogenic	6		4		4		
Other ⁹	3		2		2		
Ascites (n)	6		3		3		
MELD score	11 (9–14)		9.5 (9–14)		10.5 (9–14)		
Child-Pugh score	8 (6–9)		7 (6–9)		8 (6–10)		
Child-Pugh							
A	11		8		5		
B	15		7		5		
C	5		3		3		
Encephalopathy grade I (n)	3		0		0		

¹ MAMC, midarm muscle circumference; PBC, primary biliary cirrhosis; MELD, Model for End-Stage Liver Disease.

² Compared to healthy control subjects. The Mann-Whitney *U* or the chi-square test was used as appropriate for comparisons between groups.

³ Group A was the subgroup of all cirrhotic patients in which postprandial glucose and insulin were measured.

⁴ Group B was the subgroup of group A in which postprandial leptin and ghrelin were measured.

⁵ Median; interquartile range in parentheses (all such values).

⁶ Fat (expressed as a percentage of body weight or in kg) was calculated from anthropometric data.

⁷ Dry weight loss expressed as a percentage of actual body weight during the previous 6 mo (negative values represent weight gain).

⁸ Number of malnourished patients (based on a triceps skinfold thickness, midarm muscle circumference 5th percentile, or BMI < 18.5 kg/m²).

⁹ Of all patients with cirrhosis, 1 had autoimmune hepatitis, 1 had autoimmune hepatitis and primary sclerosing cholangitis, and 1 had nonalcoholic steatohepatitis (NASH); in group A and B, 1 patient had autoimmune hepatitis and 1 had NASH.

detection limit for the assay was 15.6 pg/mL. Leptin was expressed in absolute values and as a ratio of leptin to weight (leptin:body weight), of leptin to BMI (leptin:BMI), and of leptin to fat in kg (leptin:fat). Patients in subgroup B underwent serological testing for the detection of *Helicobacter pylori* performed according to standard in-house methods.

Statistics

Data are expressed as medians and interquartile ranges (IQRs). The Mann-Whitney *U* test was performed for calculations of differences between groups. For correlation analysis, the Spearman coefficient was calculated. Partial correlation analysis was performed to control for covariates. The chi-square test was used for comparisons between qualitative variables (sex, presence of diabetes, or hypermetabolism). To evaluate plasma glucose, insulin, leptin, and ghrelin changes postprandially, the Friedman's test was used. When the *P* value was < 0.05, a post hoc analysis with the Wilcoxon's signed rank test was performed. Multivariate repeated-measures analysis of variance was used to test the interaction between time and group. When

the *P* value was < 0.05, the Mann-Whitney *U* test was used to compare the 2 groups at each time point. Stepwise linear regression analysis was used to determine the correlation of independent variables with the energy intake:body weight or the area under the glucose curve (dependent variables), which were transformed into a normal score by using the Blom's method. All tests were two-tailed and conducted at a 5% significance level. Statistical analysis was done by using SPSS version 11.0.2 (SPSS Inc, Chicago, IL).

RESULTS

The basic characteristics of the patients and healthy control subjects are shown in **Table 1**. The patients with cirrhosis had higher insulin resistance, leptin, and REE (adjusted for FFM) as well as lower energy intake than did the healthy control subjects (**Table 2**). No significant differences in any of the variables in **Table 2** were observed between the patients with alcoholic and those with nonalcoholic cirrhosis, the patients with Child-Pugh class A and those with Child-Pugh class B or C, the patients with

TABLE 2

Metabolic and dietary data in patients with cirrhosis and healthy control subjects¹

	Patients with cirrhosis	Healthy controls subjects	P
Fasting plasma glucose (mmol/L)	5.4 (4.8–6.6) ²	4.7 (4.4–5)	0.005
Fasting serum insulin (mU/L)	19 (11–30)	7.3 (5.3–8.9)	<0.001
Fasting C-peptide (nmol/L)	0.55 (0.50–0.67)	1.10 (0.89–1.30)	0.001
Fasting leptin (pg/mL) ³	25 500 (15 950–34 525)	9995 (6528–28 525)	0.039
Fasting leptin:fat mass (pg · mL ⁻¹ · kg ⁻¹)	1030 (604–1546)	536 (228–769)	0.021
HOMA-IR	6 (2–7)	1 (1–1)	<0.001
REE (kcal/24 h)	1500 (1400–1790)	1430 (1320–1477.5)	0.112
REE adjusted for FFM (kcal · 24 h ⁻¹ · kg ⁻¹)	1509 (1412–1689)	1353 (1318–1477)	0.031
Energy intake (kcal/24 h)	1798 (1537.3–1985.8)	2271 (1768.8–2932.3)	0.011
Energy intake:body weight (kcal · 24 h ⁻¹ · kg ⁻¹)	22.1 (17.5–27.8)	26.7 (24.4–37.7)	0.028
Energy intake:REE	1.17 (0.96–1.4)	1.6 (1.27–2.04)	0.001
Hypermetabolism (n) ⁴	7	0	0.068

¹ Fasting leptin:fat mass, ratio of fasting leptin to fat mass (in kg); energy intake:body weight, ratio of energy intake to body weight (in kg); HOMA-IR, insulin resistance expressed as homeostasis model assessment index; REE, resting energy expenditure; FFM, fat-free mass; energy intake:REE, ratio of daily energy intake to REE. The Mann-Whitney *U* test was used for comparisons between groups.

² Median; interquartile range in parentheses (all such values).

³ Similar results were obtained when leptin:BMI or leptin:body weight were used (data not shown).

⁴ Hypermetabolism was defined as measured REE:predicted REE > 1.1.

malnutrition and those without malnutrition, and the patients with hepatic encephalopathy and those without hepatic encephalopathy (data not shown).

Fasting leptin was positively correlated with BMI in patients with cirrhosis ($r = 0.48$, $P = 0.007$). Also, leptin was positively correlated with body fat (in kg) in the healthy control subjects ($r = 0.78$, $P = 0.008$) but not in patients with cirrhosis ($r = 0.18$, $P = 0.4$). After control for BMI (partial correlation analysis), fasting leptin was positively correlated with HOMA-IR ($r = 0.4$, $P = 0.034$), negatively correlated with REE ($r = -0.38$, $P = 0.042$), and not significantly correlated with energy intake ($r = -0.04$, $P = 0.8$) in patients with cirrhosis. After control for BMI (partial correlation analysis), fasting leptin was negatively correlated with energy intake ($r = -0.72$, $P = 0.029$) but not to HOMA-IR ($r = -0.48$, $P = 0.2$) or REE ($r = -0.49$, $P = 0.2$) in control subjects.

Postprandial glucose

At 30 min postprandially, plasma glucose had risen in both the cirrhosis and the control groups but subsequently remained elevated only in the former (Figure 1). The interaction between time and group for glucose was found to be significant ($P = 0.037$). The area under the glucose curve (AUC) and the increase of glucose from baseline to 60 min postprandially were higher in the patients with cirrhosis than in the control subjects [respective median (IQR) AUCs: 13.7 mmol · l⁻¹ · h⁻¹ (11.9–15) compared with 10.9 mmol · l⁻¹ · h⁻¹ (8.8–11.2); $P < 0.001$; and respective median (IQR) increases: 54.8% (22.1–79.6%) compared with 20% (-21.3% to 31.9%); $P = 0.002$, respectively]. The increase of glucose from baseline to 60 min postprandially was negatively correlated with the ratio of energy intake to body weight in the patients with liver cirrhosis ($r = -0.53$, $P = 0.023$) but not in the healthy control subjects ($r = 0.37$, $P = 0.3$). HOMA-IR was positively correlated with the AUC of glucose in the patients with cirrhosis ($r = 0.75$, $P < 0.001$) but not in the control subjects ($r = 0.16$, $P = 0.7$).

Postprandial insulin

At 30 min, serum insulin had risen in both the patients with cirrhosis and the control subjects and remained elevated until 2 h postmeal in both groups (Figure 1). The interaction between time and group for insulin was not significant.

Postprandial C-peptide and serum insulin-to-C-peptide molar ratio

The interaction between time and group for C-peptide was significant ($P = 0.035$). The postprandial C-peptide response was higher in the patients with liver cirrhosis than in the healthy control subjects (Figure 1; AUC of C-peptide: 4.9 nmol · l⁻¹ · h⁻¹ (IQR: 4.2–6.7 nmol · l⁻¹ · h⁻¹) compared with 2.6 nmol · l⁻¹ · h⁻¹ (2.4–3.5 nmol · l⁻¹ · h⁻¹; $P < 0.001$). The postprandial insulin-to-C-peptide molar ratio response, a measure of portosystemic shunting, in patients with cirrhosis and healthy control subjects is shown in Figure 1. The interaction between time and group for the insulin-to-C-peptide molar ratio was not significant.

Postprandial leptin

The interaction between time and group for leptin was not significant (Figure 2). Similar results were obtained when leptin: BMI, leptin:body weight, or leptin:fat in kg were used instead of uncorrected leptin values.

Postprandial ghrelin

Postprandial ghrelin changed significantly compared with baseline only in the healthy control subjects (Figure 2). The interaction between time and group for ghrelin was significant ($P = 0.015$). At 4 h, ghrelin was higher in the healthy control subjects than in the patients with liver cirrhosis [1176 pg/mL (IQR: 679.3–1692 pg/mL) compared with 519 pg/mL (379.5–607 pg/mL); $P = 0.021$]. The increase of ghrelin from its minimal postmeal value to 4 h postmeal was higher in the healthy control subjects than in the patients with cirrhosis [39% (33.1–48.2%) compared with 14.2% (12.8–33.4%); $P = 0.005$], and it was

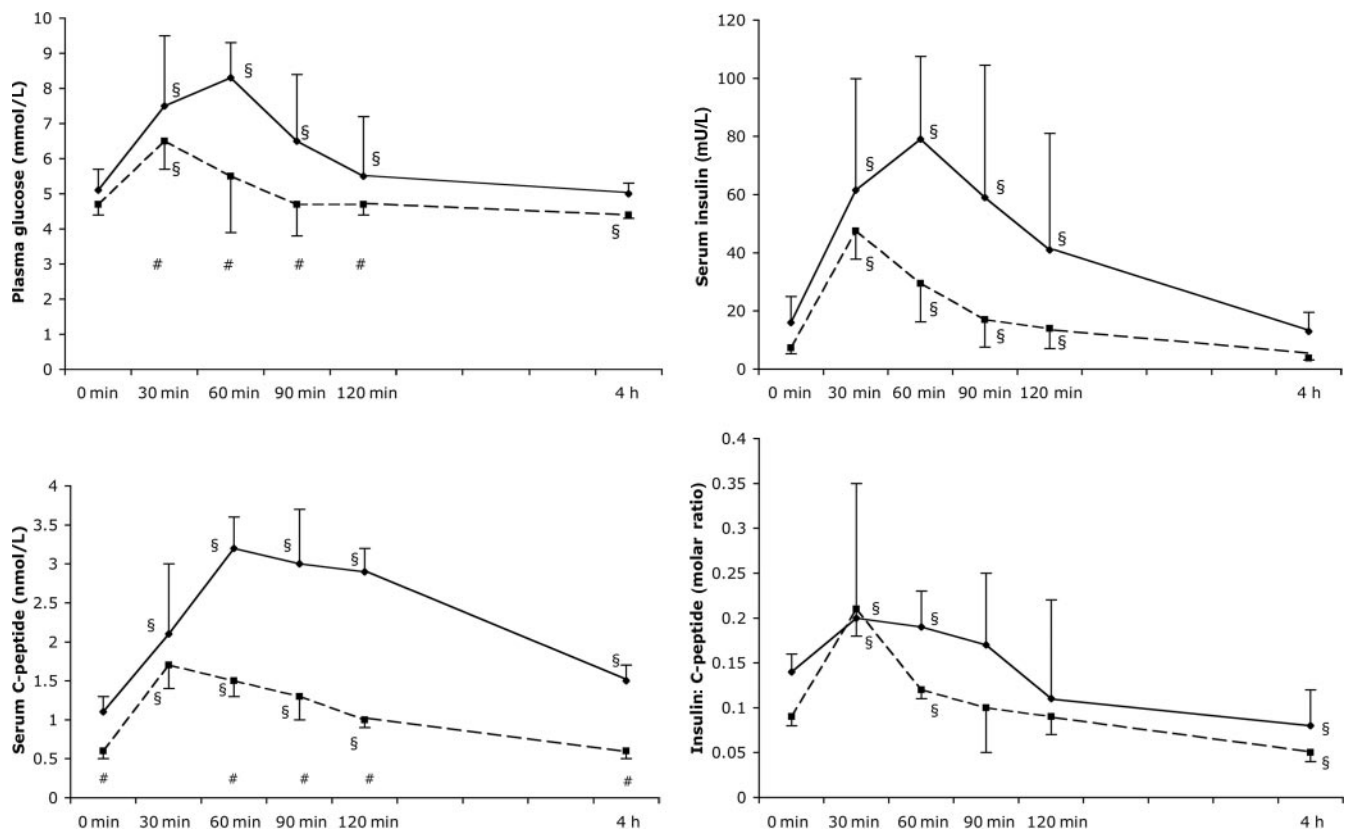


FIGURE 1. Median (half interquartile range) postprandial plasma glucose, serum insulin, and serum C-peptide concentrations and molar ratio of serum insulin to C-peptide in patients with cirrhosis (solid line; $n = 18$) and in healthy control subjects (dashed line; $n = 10$). Glucose changed significantly in patients with liver cirrhosis (Friedman's test, $P = 0.001$) and in healthy control subjects (Friedman's test, $P = 0.001$). The interaction between time and group was significant ($P = 0.037$). Insulin changed significantly in both the patients with liver cirrhosis and the healthy control subjects (Friedman's test, $P = 0.001$ for both). The interaction between time and group was not significant. C-peptide changed significantly in both the patients with liver cirrhosis and the healthy control subjects (Friedman's test, $P = 0.001$ for both). The interaction between time and group was significant ($P = 0.036$). The molar ratio of insulin to C-peptide changed significantly in both patients with liver cirrhosis and in healthy control subjects (Friedman's test, $P = 0.001$ for both). The interaction between time and group was not significant. §Significantly different from baseline values, $P = 0.05$ (Wilcoxon's signed rank test). #Significantly different from the healthy control subjects, # $P = 0.05$ (Mann-Whitney U test).

negatively correlated with weight loss in the previous 6 mo in the patients with cirrhosis ($r = -0.66$, $P = 0.014$). The AUC of ghrelin did not differ significantly between the patients with cirrhosis and the healthy control subjects (data not shown). Postprandial ghrelin concentrations were negatively correlated with glucose and insulin in both the patients with liver cirrhosis and the healthy control subjects (Table 3). The postprandial ghrelin decrease was positively correlated with leptin decrease in the healthy control subjects and negatively in the patients with liver cirrhosis (Table 3). Ghrelin concentrations were not significantly different at any time point between the patients with and those without portal hypertensive gastropathy and between the patients with ($n = 3$) and those without ($n = 10$) serological positivity for *Helicobacter pylori* (data not shown).

Regression analysis

Stepwise linear regression analysis was performed for the cirrhosis group with the ratio of energy intake to body weight as the dependent variable. Child-Pugh score, REE, the increase in glucose 60 min postprandially, and the increase in ghrelin from its minimal postmeal value to 4 h postmeal were used as independent variables. Only the increase in glucose 60 min postprandially was found to be independently correlated with energy intake ($\beta = -0.42$, $P = 0.019$).

In an attempt to identify factors involved in the increased postprandial glucose response, stepwise regression analysis was also performed for the cirrhosis group with AUC of glucose as the dependent variable. Percentage fat mass, HOMA-IR, baseline glucose concentrations, the Child-Pugh score, and the fasting serum insulin-to-C-peptide molar ratio (as a measure of hepatic shunt volume) were used as independent variables. Only insulin resistance expressed as HOMA-IR was found to be independently correlated with the postprandial glucose response ($\beta = 0.82$, $P = 0.001$) in the patients with cirrhosis.

DISCUSSION

In the current study, we observed altered postprandial responses of glucose and ghrelin associated with reduced energy intake and weight loss in patients with liver cirrhosis. The patients with cirrhosis exhibited insulin resistance with higher baseline and postprandial glucose concentrations compared with the healthy control subjects, which agrees with the results of previous studies (3, 7, 12). Although the patients with cirrhosis exhibited both higher fasting insulin and C-peptide concentrations than did the control subjects, indicating increased insulin production in the cirrhotic subjects, the postprandial glucose response was found to be independently related only to insulin

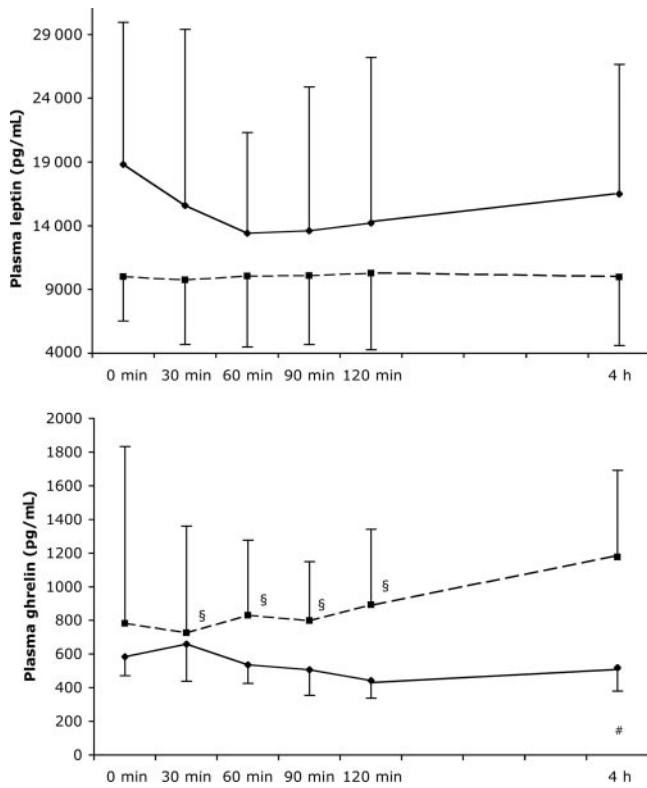


FIGURE 2. Median (half interquartile range) postprandial plasma leptin and ghrelin concentrations in patients with cirrhosis (solid line; $n = 13$) and in healthy control subjects (dashed line; $n = 10$). Leptin changed significantly in patients with liver cirrhosis (Friedman's test, $P = 0.001$) but not in healthy control subjects (Friedman's test, $P = 0.5$). The interaction between time and group was not significant. Ghrelin changed significantly in the healthy control subjects (Friedman's test, $P = 0.001$) but not in the patients with liver cirrhosis (Friedman's test, $P = 0.13$). The interaction between time and group was significant ($P = 0.015$). [§]Significantly different from concentrations at 4 h, $P = 0.01$ (Wilcoxon's signed-rank test). [#]Significantly different from healthy control subjects, $P = 0.021$ (Mann-Whitney U test).

resistance. Furthermore, the postprandial increase in glucose was found to contribute independently to the reduced energy intake in the patients with cirrhosis. Decreased hunger and slower gastric emptying were observed in healthy volunteers during induced hyperglycemia (32). Postprandial hyperglycemia has been reported to be associated with increased postprandial upper gastrointestinal symptoms (33, 34) compared with euglycemia in

healthy volunteers. We recently reported an increased prevalence of gastrointestinal symptoms (including early satiety) in patients with cirrhosis (35, 36). It is therefore possible that postprandial hyperglycemia results in reduced energy intake by contributing to early satiety and other gastrointestinal symptoms in patients with cirrhosis.

Baseline leptin in patients with cirrhosis was found to be elevated, as previously reported (14–18), and leptin effects on energy intake and REE were disturbed in these patients. Leptin has been shown to increase REE (13), but in a recent study performed in non-cirrhotic individuals, total and free leptin were reported to be negatively and bound leptin positively associated with REE (37). We observed a negative association between total leptin and REE in patients with cirrhosis. It might therefore be hypothesized that the resistance to the effects of leptin in cirrhotics observed in the current study is mediated by a proportional increase in free leptin. However, we did not measure free and bound leptin fractions in our series, which is mandatory to show this. Alternatively, the disturbed associations of leptin with energy intake and REE in cirrhosis might simply indicate disturbed metabolic regulation in these patients, documenting the central role of liver metabolism in whole-body fuel homeostasis. The results of the current study, however, do not support a role of postprandial leptin concentrations in the low energy intake seen in patients with cirrhosis.

Ghrelin concentrations after a meal have not been investigated previously in patients with liver cirrhosis. The patients with cirrhosis had a clearly altered postprandial pattern of ghrelin compared with the control subjects, with an attenuated ghrelin increase at 4 h postmeal. Ghrelin enhances appetite and food intake, and its concentration rises preprandially, thus playing a role in meal initiation (13). Therefore, the low ghrelin observed in the patients with cirrhosis at 4 h postmeal (ie, before expected lunch in our experiment setting) could be involved in the reduced energy intake in these patients. In a recent study, fasting ghrelin was found to be elevated in patients with liver disease compared with healthy control subjects (19). Marchesini et al (5) reported that fasting ghrelin was comparable in patients with cirrhosis and control subjects but increased concentrations were identified in a group of patients with low energy intake and malnutrition. In our study, we were also unable to confirm generally increased fasting ghrelin in patients with cirrhosis. These discrepancies could, at least in part, be explained by different patient selection, control subject selection, or both. Patients in the former study (19) were

TABLE 3

Spearman correlations of postprandial ghrelin with postprandial glucose, insulin, and leptin variables in patients with liver cirrhosis and healthy control subjects¹


	Ghrelin at 30 min postmeal		Ghrelin decrease at 30 min postmeal				Ghrelin at 90 min postmeal				Ghrelin decrease at 90 min postmeal					
	Patients with cirrhosis		Control subjects		Patients with cirrhosis		Control subjects		Patients with cirrhosis		Control subjects		Patients with cirrhosis		Control subjects	
	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>	<i>r</i>	<i>P</i>
Glucose increase at 90 min postmeal	-0.63	0.022	-0.64	0.048	0.10	0.748	0.04	0.907	-0.66	0.014	-0.65	0.043	-0.21	0.482	0.07	0.855
Insulin increase																
2 h postmeal	-0.48	0.112	-0.7	0.036	0.43	0.167	0.67	0.050	-0.74	0.006	-0.47	0.205	-0.13	0.681	0.50	0.138
4 h postmeal	0.25	0.443	-0.71	0.019	0.00	1.000	0.56	0.090	0.16	0.625	-0.61	0.060	-0.26	0.417	0.63	0.067
Leptin decrease																
30 min postmeal	0.14	0.665	-0.06	0.881	-0.54	0.058	0.04	0.910	-0.20	0.511	0.12	0.751	-0.12	0.707	0.29	0.430
90 min postmeal	0.37	0.209	-0.18	0.627	-0.59	0.035	0.75	0.013	-0.10	0.768	-0.06	0.881	-0.04	0.901	0.75	0.013

¹ $n = 13$ patients with cirrhosis and 10 control subjects.

transplantation candidates, some had malignancies and were not BMI-matched with control subjects, whereas in the current study, no patients with malignancies were included and BMI-matched control subjects were chosen.

The mechanisms of altered postprandial ghrelin response might involve glucose, insulin, leptin, or all three. Postprandial ghrelin was negatively related to glucose and insulin in both healthy control subjects and patients with cirrhosis, as previously reported (20–22). According to these studies, insulinemia is essential for postprandial ghrelin suppression with glucose having an additional effect (20–22). In our series, the postprandial ghrelin decrease was negatively related to leptin reduction in the patients with cirrhosis. This agrees with earlier data suggesting an inverse relation between leptin and ghrelin and that leptin could be important for suppression of ghrelin (24). Therefore, insulin resistance resulting in high postprandial glucose and insulin might be involved in the low ghrelin observed 4 h postmeal. Thus, it is conceivable that treatment of insulin resistance might reduce the hypoghrelinemia before a meal in patients with cirrhosis, possibly stimulating appetite. Although this is probably not the single most important reason for reduced energy intake in liver cirrhosis, it certainly warrants further studies.

Certain methodologic aspects should be taken into consideration when interpreting the results of the current study. Food intake was assessed by means of food diaries. This is an established method of food intake assessment (29, 37–39), which has been previously utilized in patients with liver cirrhosis (4, 5, 12). However, it is known that both normal-weight and obese subjects may underestimate their dietary intake (39), and it is conceivable that patients with hepatic encephalopathy might also be prone to underreporting when filling in detailed food diaries. In the current study, no patients with encephalopathy grade II or higher were included and food intake was not statistically different between the patients with and those without hepatic encephalopathy grade I. Furthermore, our findings confirm previous studies showing reduced energy intake in patients with cirrhosis (3–5) and reports of a negative correlation between leptin and food intake in healthy subjects (39). Second, in the current study, fasting data were obtained from all subjects but postprandial data were obtained from a smaller subgroup of the main patient population. Although the patients with cirrhosis were carefully matched with the group of healthy control subjects, a type 2 error in the assessment of the postprandial responses cannot be ruled out. Lastly, the current study was a cross-sectional one. Thus, statistical correlations between hormonal disturbances and energy intake or REE in cirrhosis do not necessarily implicate a cause-effect relation.

In conclusion, altered postprandial glucose and ghrelin concentrations correlated with reduced energy intake and weight loss in liver cirrhosis. The effects of leptin on energy expenditure and energy intake seem to be altered in patients with cirrhosis. Insulin resistance might be involved in the altered postprandial glucose and ghrelin responses. 

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writing of the manuscript. None of the authors have a personal or financial conflict of interest.

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