Adipokine Levels in the Course of Mild Biliary Pancreatitis

Selim Demirci^a Erdem Akbal^b Erdem Koçak^b Adnan Taş^b Seyfettin Köklü^b

Departments of ^aInternal Medicine and ^bGastroenterology, Ankara Education and Research Hospital, Ministry of Health, Ankara, Turkey

Key Words

Acute pancreatitis • Adiponectin • Insulin • Leptin

Abstract

Objectives: Obesity markedly increases the risk of severe acute pancreatitis (AP). Several adipokines have been ascribed a role as a predictor of clinical severity in AP. Therefore, the aim of this study was to investigate a possible relationship between leptin and adiponectin and mild biliary AP.

Methods: We included 24 consecutive patients with mild biliary AP and 24 consecutive healthy age- and sex-matched controls. Clinical severity was classified by the Ranson score. ELISA was used to assess leptin and adiponectin levels on admission and in remission. Complete blood cell counts and other laboratory tests were also performed at baseline and in remission. **Results:** Leptin, adiponectin, insulin and HOMA-IR measurements showed no difference in pancreatitis patients both on admission and in remission compared to the control group. No difference was found in leptin, insulin or HOMA-IR levels in the course of pancreatitis. However, adiponectin levels were higher in remission compared to admission. **Conclusions:** Increased adiponectin levels in remission may be an indication of improvement in this condition. Further studies are needed to determine whether adiponectin provides protection from AP.

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Introduction

Acute pancreatitis (AP) is an inflammatory disease associated with autodigestion of the pancreas due to intra-pancreatic activation and release of digestive enzymes. Obesity and lipid metabolism are associated with the severity of AP [1]. Adipocyte hyperplasia is a trigger that initiates the inflammatory state, which results in the release of adipocytokines, such as TNF α , leptin, adiponectin, resistin, ghrelin and visfatin. Obesity markedly increases the risk of severe AP, possibly through the action of adipokines [2–4]. Several investigations also show an increased mortality and morbidity in obese AP patients, and the implications of the adipokines leptin and adiponectin [2–6]. However, the role of leptin and adiponectin in AP is unclear.

Leptin is one of the most important cytokines secreted by adipocytes, and it plays vital roles in controlling food intake and body energy balance [7]. Experimental studies on AP in rats have shown that leptin levels increase after cerulein-induced pancreatitis, presumably from pancreatic origin, and that exogenous administration is associated with diminished histological manifestations of pancreatitis and reduced plasma TNF α [3, 5, 8]. Similarly, leptin administration reduced the severity of ischemia/reperfusion-induced pancreatitis by diminishing morphological features of pancreatic damage and serum IL-1 β levels, while accelerating pancreatic repair by increas-

ing pancreatic blood flow and DNA synthesis [8]. These results are contrary to what would be expected according to other experimental studies evaluating the effect of leptin in non-pancreatic tissue, and point to a possible tissue-specific anti-inflammatory role in AP.

Adiponectin is recognized as an anti-inflammatory and antifibrotic protein derived from adipocytes. It has anti-inflammatory properties in addition to various biological functions, and low serum adiponectin levels are present in obesity [9]. Adiponectin plays a protective role in cerulein-induced AP [6, 10, 11]. Hypoadiponectinemia could enhance the severity of pancreatitis [12]. In AP patients, serum adiponectin levels are negatively correlated with organ dysfunction [2]. However, the role of adiponectin has not been carefully investigated in the course of AP. Recent studies have highlighted the relationship between obesity and pancreatic disease. However, the role of adipokines, including leptin and adiponectin, in AP remains uncertain. The aim of this study was to determine adipokine levels in the course of AP.

Patients and Methods

A total of 24 patients with mild biliary pancreatitis and 24 controls were included in the study. The control group consisted of sex- and age-matched healthy subjects. We excluded causes of AP other than gallstones, such as alcoholism, abdominal surgery, medication (estrogens, corticosteroids, thiazide diuretics and azathioprine), hereditary pancreatitis, hypercalcemia, hypertriglyceridemia, infection, injury to the abdomen, pancreatic cancer and the association with endoscopic retrograde cholangiopancreatography. Diagnosis of AP was based on typical clinical manifestations with at least a 3-fold increase in serum amylase and/or lipase. Whenever uncertainty about the diagnosis existed, a CT scan was performed to confirm/rule out AP. All patients were hospitalized and given conservative management. Prognostic severity determined by modified Ranson's criteria was recorded for each patient [13]. A score ≤ 3 on admission was defined as mild pancreatitis according to Ranson's criteria. Weight and height were measured in all patients during the first 48 h following admission for body mass index (BMI) calculation. Oral feeding was initiated after clinical and laboratory findings had improved. Remission was defined as the disappearance of clinical symptoms and resolution of the pancreatic abnormalities on imaging studies. Patients in remission were started on oral feeding.

Blood samples were obtained on admission and in AP remission. During the first 24 h following admission and on the 1st day of remission (initiation of oral feeding), aliquots of a serum sample obtained from all patients were stored at -40°C until analysis. Insulin resistance was calculated by the HOMA-IR formula (fasting insulin value $\times$ fasting blood glucose/22.5) [14]. Assessment of adiponectin and leptin levels is based on the principle of a solid-phase ELISA. The study protocol was approved by the local ethics committees.

Data analysis was carried out using SPSS 15.0 software. Continuous variables are shown as means $\pm$ SD or medians (ranges), while categorical variables are expressed in percent. Student's *t* test or the Mann-Whitney test was employed to assess significant differences between groups in terms of characteristics and parameters. Linear correlations between continuous variables were assessed using Spearman's correlation test. The χ^2 test was used for categorical variables. Power and sample size calculations were performed according to Schesselman [15]. The power of the study is 80%. A value of $p < 0.05$ is considered to be statistically significant.

Results

Demographic and clinical characteristics of the patient and control groups are summarized in table 1. No differences were observed for age, sex, hypertension or smoking status. Leptin, adiponectin, insulin and HOMA-IR levels were not different for pancreatitis patients both on admission and in remission compared to the control group. No difference was found in leptin, insulin or HOMA-IR levels in the course of pancreatitis. However, adiponectin levels were higher in remission than on admission. Median leptin concentrations on admission were 6 ng/ml (range 1.54–85.0) and 6.9 ng/ml (range 1.54–47.0) in remission ($p = 0.094$). Adiponectin concentrations on admission were 12,360 ng/ml (range 210–39,900) and 19,395 ng/ml (range 4,290–56,190) in remission ($p = 0.015$).

We investigated with correlation analysis whether there was a relationship between leptin and adiponectin and lipase, amylase, liver tests, sedimentation, CRP, insulin or BMI. Leptin correlated significantly with BMI ($r = 0.555$, $p < 0.001$) and insulin ($r = 0.301$, $p = 0.037$). However, there was no significant correlation between leptin and lipase ($p = 0.434$); amylase ($p = 0.368$); AST ($p = 0.688$); ALT ($p = 0.466$); ALP ($p = 0.364$); GGT ($p = 0.920$) and CRP ($p = 0.267$). There was also no correlation between adiponectin and lipase ($p = 0.404$); amylase ($p = 0.998$); AST ($p = 0.221$); ALT ($p = 0.557$); ALP ($p = 0.295$); GGT ($p = 0.903$); CRP ($p = 0.395$); insulin ($p = 0.366$) and BMI ($p = 0.490$).

The patients were classified into two groups according to their BMI; 13 patients had a normal BMI (<27) and 11 patients were obese (BMI >27). In the normal BMI group, insulin levels were $5.4 \mu\text{U/ml}$ (range 1.6–17.1), HOMA-IR values were 1.49 ± 0.97 and leptin levels were 4.7 ng/ml (range 1.54–17). In the obese group, insulin levels were $15.8 \mu\text{U/ml}$ (range 2.2–31), HOMA-IR values were 5.45 ± 2.57 and leptin levels were 31 ng/ml (range 3.24–85). Leptin, insulin and HOMA-IR were higher in obese patients than in patients with normal BMI ($p < 0.008$, $p <$

Table 1. Demographic and clinical characteristics of the patient and control groups

	Control group (n = 24)	AP (n = 24)		p value	
		on admission	in remission	control vs. admission ^a	admission vs. remission ^b
Age, years	46.6 ± 16.6	53.9 ± 19.4		0.13	
Sex					
Male, %	37.5 (n = 9)	33.3 (n = 8)		0.763	
Female, %	62.5 (n = 15)	66.7 (n = 16)			
BMI	27.3 ± 4.1	27.2 ± 4.1		0.942	
Smokers, %	4.2 (n = 1)	20.8 (n = 5)		0.07	
Hypertension, %	8.3 (n = 2)	16.7 (n = 4)		0.379	
Hyperlipidemia, %	4.1 (n = 1)	16.7 (n = 4)		0.015	
Ranson's score		2 (1–3)			
AST, U/l	20.5 (14–45)	91.0 (18–321)	41 (15–88)	<0.001	<0.001
ALT, U/l	17.5 (8–38)	96.5 (13–600)	40 (12–112)	0.003	0.003
ALP, U/l	71 ± 20.8	175.7 ± 115.6	143.5 ± 79.1	<0.001	0.121
GGT, U/l	19.5 (12–55)	167.0 (9–1,011)	137.5 (9–788)	<0.001	0.039
Bilirubin, mg/dl					
Total	0.6 (0.3–1.3)	0.9 (0.3–7.7)	0.7 (0.3–3.1)	0.004	0.01
Direct	0.1 (0.06–0.1)	0.2 (0.07–5.0)	0.19 (0.08–1.68)	0.005	0.017
LDH, U/l	194.3 ± 41.7	327.2 ± 97.2	284.7 ± 112.6	<0.001	0.126
Amylase, U/l	69.2 ± 22.1	852 ± 491	94.6 ± 29.7	<0.001	<0.001
Lipase, U/l	14.5 (3–57)	249.5 (9–2,276)	69.5 (9–639)	<0.001	<0.001
Glucose, mg/dl	91.7 ± 8.4	103.7 ± 29.7	109 ± 46.2	0.062	0.470
BUN, mg/dl	30.6 ± 15.4	26.7 ± 20.8	20.5 ± 15.6	0.463	0.003
Creatinine, mg/dl	1 ± 0.2	0.97 ± 0.39	0.9 ± 0.3	0.552	0.220
Albumin, g/l	4.3 ± 0.27	3.5 ± 0.4	3.4 ± 0.4	<0.001	0.891
Calcium, mg/dl	9.8 ± 0.5	8.5 ± 0.6	8.8 ± 0.6	<0.001	0.002
CRP, mg/l	0.45 ± 0.37	8.6 ± 7.42	3.3 ± 2.8	<0.001	0.088
Hemoglobin, g/dl	13.8 ± 1.6	12.3 ± 1.8	12.7 ± 1.5	0.005	0.599
Leukocytes, n/mm ³	6,750 ± 1,604	11,758 ± 5,113	8,433 ± 4,291	<0.001	<0.001
Insulin, µU/ml	10.7 (0.25–20.8)	8.1 (1.65–31)	7.11 (1.7–32)	0.557	0.995
Leptin, ng/ml	3.94 (1.58–50.9)	6 (1.54–85)	6.96 (1.54–47)	0.339	0.094
Adiponectin, ng/ml	17,280 (210–37,020)	12,360 (210–39,900)	19,395 (4,290–56,190)	0.604	0.015
HOMA-IR	2.6 ± 1.0	3.3 ± 2.7	3.3 ± 3.2	0.296	0.995

^a χ^2 test (gender difference, smoking, hypertension and hyperlipidemia) and Student's t test (other measurements).

^b Wilcoxon's rank test.

0.001 and $p < 0.001$, respectively). Adiponectin levels were 4,830 ng/ml (range 210–28,290) in the normal BMI group and 16,230 ng/ml (range 960–39,900) in the obesity group. Adipokine levels showed no difference between the two groups ($p = 0.369$).

Discussion

In this study, we assessed the serum levels of leptin, adiponectin and insulin. The main outcome of our study was that adiponectin levels were significantly higher in

remission than on admission in patients with mild AP. In contrast, no differences were observed regarding serum leptin and insulin levels. Furthermore, there was a correlation between leptin levels and BMI and insulin.

The role of leptin in AP remains puzzling. The role of leptin as an anti-inflammatory and protective signal that was demonstrated in some experimental rat models should have led to significantly increased or decreased levels in severe cases [5, 8, 16–20]. Several experimental studies in animal models have suggested a protective role for leptin in AP via suppression of the pro-inflammatory response, reduction in leukocyte infiltration in pancre-

atic tissue and increased pancreatic repair [8, 17, 18, 20]. However, though the serum concentrations of leptin increase in AP, there was no relationship between AP severity and circulating leptin concentrations [21]. In humans, leptin levels do not correlate with disease severity, suggesting that adipokines do not affect the course of AP [3, 19]. One study demonstrated that leptin was significantly elevated in patients with severe pancreatitis and was correlated with a radiological scoring system for extra-pancreatic necrosis [3]. Our study showed no differences in leptin levels from admission to remission in mild AP patients. However, there was a positive correlation between leptin and BMI and insulin. Increased leptin levels in AP would result in even further increased levels if superimposed on a chronic inflammatory condition. Differences in leptin levels were entirely explained by variations in BMI rather than disease activity. Our findings do not support an association between leptin and inflammatory response. Similar to leptin, the role of adiponectin in

AP is controversial. Adiponectin, an anti-inflammatory adipokine, has been investigated in experimental pancreatitis. It plays a protective role in the cerulein model of AP. Adiponectin levels were inversely correlated with BMI and were significantly lower for patients with severe than mild AP. Furthermore, in AP patients, serum adiponectin levels were inversely correlated with organ dysfunction. However, other studies detected no significant differences in serum adiponectin concentrations [2, 3, 10–12, 21].

In conclusion, leptin levels do not change in the course of mild biliary AP. The leptin level is correlated with BMI. According to our findings, increased adiponectin levels in remission may be an indication of improvement.

Disclosure Statement

The authors declare that they have no competing interest.

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