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INVERSE RELATIONSHIP OF CIRCULATING GHRELIN AND LEPTIN PEPTIDE HORMONES IN OBESE AND NON-OBESE SUBJECTS

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ABSTRACT

OBJECTIVE: *Ghrelin is an appetite hormone activates food intake and participates in the regulation of energy homeostasis. Ghrelin and leptin levels are influenced by body fat. The aim of the study was to compare fasting ghrelin and leptin levels between obese and non-obese subjects and to investigate possible correlations of these peptide hormones with body mass index.*

STUDY DESIGN: *This was a prospective comparative study, conducted in the Department of Physiology, Basic Medical Sciences Institute, Jinnah Postgraduate Medical Centre, Karachi*

SUBJECTS AND METHODS: *Sixty subjects age range 22 to 52 years were included in this study. These subjects were divided in two groups according to body mass index (BMI). The 30 subjects age (36.50 ± 1.76) with BMI >25, (32.88 ± 0.83 kg/m^2) were considered obese (16 women and 14 men) and 30 subjects age (37.07 ± 1.81) with BMI<25 (21.08 ± 0.42 kg/m^2) were considered non-obese (21 women and 9 men). In all participants, height, weight and body mass index (BMI) were measured. Fasting blood sugar, ghrelin and leptin levels using kit were determined.*

RESULT: *In obese, BMI (32.88 ± 0.83, P<0.01) was significantly higher as compared to non-obese controls (21.08 ± 0.42). Comparing obese to non-obese subjects ghrelin level was significantly decreased (172.09 ± 3.37 vs. 288.98 ± 4.59; p<0.01) and leptin levels was significantly increased (56.28 ± 4.15 vs. 22.30 ± 3.02; p<0.01). Triacylglycerol showed significant difference (p<0.01) while cholesterol was non significant. BMI was positively correlated with leptin and negatively with ghrelin levels. A negative correlation between circulating leptin and ghrelin levels was found.*

CONCLUSION: *We observed significant inverse relationship between ghrelin and leptin serum levels in obese subjects. Ghrelin might be able to increase the appetite as leptin fails to suppress food intake in obese subjects because of the resistance of hypothalamic centers to leptin*

KEY WORDS: Obesity, Body mass index, Ghrelin, Leptin

INTRODUCTION

Obesity is an excessive accumulation of energy in the form of body fat which impairs health. Obesity is so strongly associated with medical comorbidities and mortality that it has begun to overtake infectious diseases as the most significant contributor to ill health worldwide.[1] The etiology of obesity is very complex and is based upon the interplay of genetic and environmental factors. Delgado-Aros et al., (2004)[2] suggested that decreased satiety perception represents an important risk factor for the development of obesity. Obese subjects have delayed onset of satiety after meal consumption which might be due to alterations in hormonal responses to food intake.[3] Modulation of satiety perception through the changes of gastrointestinal hormone ghrelin secretion represents a promising approach to prevent obesity complications.[4]

Gastrointestinal hormones play an important role in the neuroendocrine regulation of food intake and postprandial satiety. Ghrelin is a 28-amino acid orexigenic peptide hormone produced mainly by the stomach that is involved in both the long-term regulation of body weight and the short-term regulation of postprandial satiety. Impairments in ghrelin secretion may in concert with other factors play an important role in the development of both obesity and anorexia nervosa.[5] Ghrelin identified as the endogenous ligand for growth hormone secretagogue receptor (GHS-R)[6], stimulates growth hormone (GH) secretion

from the pituitary and biological actions including increases food intake and inducting adiposity. Nass et al., (2010)[7] suggests that wide tissue distribution of ghrelin may have other functions including increased appetite, altered gastrointestinal motility and regulated lipid and glucose metabolism.

Leptin is a 167 amino-residue peptide hormone encoded by the obesity gene.[8] It is secreted by fat tissue and suppresses food intake, while increasing energy expenditure. Leptin levels are influenced by the amount of body fat, as they are found high in obese and low in lean individuals.[9]

Kapica et al., (2008)[10] states that leptin is also considered as a gastrointestinal hormone, since leptin and leptin receptors are found in gastrointestinal system. In humans, plasma leptin level peaks 1-2 hr after food intake and significant reduction below the control level is observed after 24 hr-fasting, thus circulating leptin after meal inhibit food intake. Thus leptin act as an appetite suppressing factor that transmits satiety signals to the brain. Hunger signals from peripheral tissues, however, had remained unidentified until the discovery of ghrelin.[11]

Leptin acts at the central nervous system (CNS) in suppressing food intake and stimulating energy expenditure.[12] Yildiz et al., (2004)[13] studies pointed to adipose tissue as an active endocrine organ that secretes a range of hormones such as leptin and adiponectin. Leptin and ghrelin are part of the regulation of energy metabolism. The neurons of the lateral hypothalamus are involved in the release of neuropeptide Y (NPY) and agouti releasing peptide hormone (AgRP) that then activate the paraventricular nucleus (PVN) or the "feeding center".[12] Inhibition of the PVN induces satiety. Fasting stimulates the release of ghrelin from the oxyntic mucosa and releases growth hormone from hypothalamus promotes the release of NPY and AgRP to stimulate the PVN and increase feeding behavior. Disruption in the balance between the anorexigenic and orexigenic factors can lead to disorders such as obesity or cachexia (weight loss).[14]

Studies report that as weight loss occurs, ghrelin levels rise and leptin levels decrease.[12] Obesity is seen as a chronic adaptation to the hormonal changes that occur due to the increase in adipose tissue. With weight gain, there are increased leptin and decreased ghrelin levels but there may also be an up or down regulation of receptors in the hypothalamus, which in turn will cause sensitization or desensitization to the effects of the hormones.[11]

Overall, gastrointestinal (GI) hormones constitute a significant part of the complex neuroendocrine regulation of energy balance. Most of GI tract hormones increases satiety and decrease food intake. The only known exception is a hormone ghrelin that has the opposite effects.[15] Therefore study is planned 1) to compare the status of ghrelin and leptin in obese compared to non-obese subjects and 2) to investigate any correlation between these two hormones and body mass index.

TABLE # 1

Comparison of biophysical parameters of non-obese and obese subjects
The values are expressed as mean ± s.e.m.
The numbers of observations are shown in parenthesis.

Parameters	Non-obese (n = 30)	Obese (n = 30)
Age (Years)	37.07 ± 1.81	36.50 ± 1.76
Sex Male	21 (70.0%)	14 (46.7%)
Female	09 (30.0%)	16 (53.3%)
Height (cm)	167.30 ± 1.54	163.30 ± 1.73
Weight (kg)	59.43 ± 1.49	87.75 ± 2.51*
BMI (kg/m^2)	21.08 ± 0.42	32.88 ± 0.83*

*P<0.01 significant difference when compared obese vs non-obese

TABLE # 2

Comparison of biochemical parameters of non-obese and obese subjects
The values are expressed as mean ± s.e.m.
The numbers of observations are shown in parenthesis.

Parameters	Non-obese (n = 30)	Obese (n = 30)
Ghrelin (pg/ml)	288.98 ± 4.59	172.09 ± 3.37*
Leptin (ng/ml)	22.30 ± 3.02	56.28 ± 4.15*
FBS (mg/dl)	87.36 ± 1.44	93.13 ± 1.51*
Cholesterol (mg/dl)	177.26 ± 6.60	193.80 ± 7.15
TAG (mg/dl)	135.53 ± 8.77	164.63 ± 10.56*

*P<0.01 significant difference when compared obese vs non-obese

TABLE # 3

Correlation co-efficient of ghrelin and leptin vs weight and BMI in obese subjects

S. No.	Parameter	Ghrelin (r-value)	Leptin (r-value)
1.	Weight (kg)	-0.77**	0.16
2.	BMI (kg/m^2)	-0.83**	0.40*
5.	Leptin (ng/ml)	-0.22	-

* Correlation is significant at the p < 0.05 level
**Correlation is significant at the p < 0.01 level

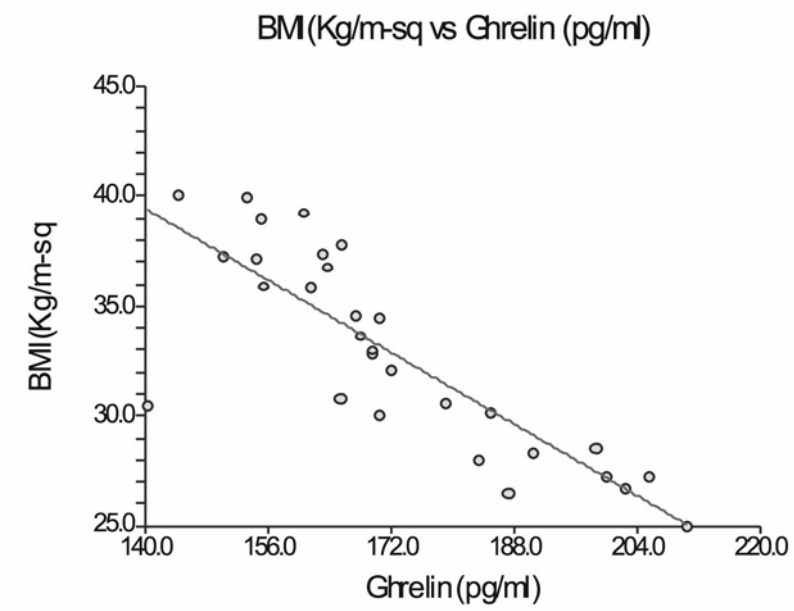
MATERIALS AND METHODS

This proposed study was carried out in the Department of Physiology, Basic Medical Sciences Institute (BMSI), Jinnah Postgraduate Medical Centre (JPMC) Karachi. This is a prospective comparative study performed on a total of 60 individuals age range 22 to 52 years of both sexes, divided by using body mass index (BMI) as 30 obese patients mean age 36.50 ± 1.76 and 30 of mean age 37.07 ± 1.81 matched non-obese normal healthy control subjects were selected from general population for comparison. The subjects were interviewed in detail regarding the name, age, address, occupation, medical and surgical history, diabetes and any other associated chronic illness on the questionnaire. All the participants were screened by medical history and physical examination. None of them had any chronic disease or was undergoing any pharmacological treatment. Body weight (kilograms) and height (meters) were measured and body mass index (BMI) was calculated (weight in kilograms divided by the square of height in meters).

After an overnight fast, morning blood samples were drawn from all subjects included in the study. Blood samples were centrifuged (10 min at 1500g) and the aliquots were stored at –20°C until assayed according to the instructions published by Groeschl et al.,[16] regarding ghrelin. In groups, fasting glucose, ghrelin, leptin, cholesterol and triglyceride levels were measured. Serum glucose was determined with the enzymatic method (Randox), ghrelin and leptin were determined by enzyme-linked immunosorbent assay (ELISA) based on a double-antibody sandwich technique (diagnostic system laboratories, U.S.A.).The cholesterol and triglyceride were determined by kit method (Randox). A written consent of the individuals was taken after explaining the procedure that underwent clinical and biochemical investigation.

FIGURE # 1

Correlation co-efficient of ghrelin vs BMI in obese subjects



RESULT

The anthropometric features of obese and non-obese are presented in Table 1. In all participants, height, weight and body mass index (BMI) were measured, statistically no significant difference in age or gender distribution was observed among both groups. In obese, BMI (32.88 ± 0.83, P<0.01) was significantly higher as compared to non-obese controls (21.08 ± 0.42).

Comparing the ghrelin and leptin of obese showed significantly inverse relationship with non obese subject group (Table 2). In this table the ghrelin level was significantly decreased (172.09 ± 3.37 vs. 288.98 ± 4.59; p<0.01) and the leptin levels was significantly increased (56.28 ± 4.15 vs. 22.30 ± 3.02; p<0.01) of obese compared to the non-obese subjects. Regarding lipids; triacylglycerol showed significant difference (p<0.01) and cholesterol showed slightly increased but non significant levels in obese as compared to non-obese. Whereas fasting blood sugar was under the normal limits in both groups but was significantly high in obese group (Table 2).

BMI was positively correlated with leptin and negatively with ghrelin levels. A negative correlation between circulating leptin and ghrelin levels was found. A suggestive negative correlation between ghrelin levels and weight, independent of BMI, was also observed (r= -0.77, P=0.01) (Table 3). Figure 1 shows negative correlation of BMI with ghrelin.

DISCUSSION

Ghrelin is at present the only known Gastrointestinal (GI) hormone that increases food intake. Plasma ghrelin levels are inversely correlated with body weight and rise following weight loss in humans.[17] Ghrelin levels are remarkably decreased in states of obesity, a disorder prone to develop hyperinsulinemia and insulin resistance. An opposite relationship also exists between serum leptin and plasma ghrelin levels[13] and rodent studies showed that these hormones exert counteracting effects on neuropeptide Y-positive hypothalamic neurons controlling appetite. Our study confirms that ghrelin secretion is less oscillatory and there is an overall decrease in obese compared with non-obese (p<0.01). Leptin, a valuable parameter of obesity was observed significantly higher in obese when compared to non-obese.

We also found that fasting plasma ghrelin concentration in patients with obesity correlated negatively with BMI. Marzullo et al., (2006)[18] states that fasting ghrelin levels are decreased in human obesity, which might represent a compensatory response to a sustained positive energy balance. Obese subjects are not resistant to the effects of ghrelin administration and ghrelin could play an important role in the endocrine abnormalities commonly present in obesity.[19] Obese people appear to be more sensitive to the appetite stimulating effects of exogenous ghrelin than normal-weight subjects and inhibition of circulating ghrelin could be a useful therapeutic approach in the treatment of obesity.[20] Moreover, Nass et al., (2010)[7] states that the current epidemic of obesity in the face of a surfeit is based on caloric restriction. Our observations confirm previous data showing that ghrelin secretion was decreased in obesity (p<0.01) as we obtained the significant negative correlation of BMI with ghrelin (r= -0.83, p<0.01) in obese.

Buyukbese et al., (2004)[21] proposed that one potential factor involved in the control of ghrelin is leptin, an adipocytokine that signals the amount of energy accumulation

to the hypothalamus and is implicated in long-term control of energy balance. All experimental conditions of our study showed higher leptin levels in obese than non-obese normal subjects. Moreover, we think that obesity may play an important role in the release of ghrelin as we observed negative correlation between ghrelin and leptin.

In our study the mean value of triacylglycerol (TAG) was highly significant ($p<0.01$) in obese when compared to non-obese subjects, it may be due to mobilization of more free fatty acids in obese which can be explained by reduced action of insulin at the tissue level . In obese, level of cholesterol was greater but non significant as compared with non-obese subjects. Our results are similar to Bener et al., (2007).[22]

When the mean heights of obese was compared with non-obese control group we observed no difference, it clearly indicated that height had no effect with obesity, when the mean weight and BMI of both groups were compared to the non-obese control group significant difference was observed. Our results related to BMI are in accordance to Sarah et al., (2008).[23]

Finally our study indicates as a whole the relationship of ghrelin with BMI and leptin proved ghrelin as an appetite hormone which by increasing the food intake becomes the probable the reason of obesity in obese. BMI showed inverse correlation with ghrelin and observed decrease in ghrelin concentration in obese. Further it was found that there was some what relation of lipid particularly hypertriglyceridemia in obese. Conclusion: we observed significant inverse relationship between ghrelin and leptin serum levels in obese subjects. Ghrelin might be able to increase the appetite as leptin fails to suppress food intake in obese subjects because of the resistance of hypothalamic centers to leptin.

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