2024, 21(11): 3438-3445
https://doi.org/10.21123/bsj.2024.8990
P-ISSN: 2078-8665 - E-ISSN: 2411-7986
Baghdad Science Journal

The role of gut hormonal aspect in Iraqi patients subjected to sleeve Gastrectomy

Rafida M. Al-Amiri*[1], Hanaa S. Kadhum[2], Falih M. Ali[3]

[1] Department of Basic Sciences, College of Dentistry, University of Basrah, Basrah, Iraq.
[2] Department of Pathological Analysis, College of Science, University of Basrah, Basrah, Iraq.
[3] Al-Basra metabolic and Bariatric Surgery Center/Al-Sadder Teaching Hospital, Basrah, Iraq.
*Corresponding Author.

Received 24/04/2023, Revised 26/09/2023, Accepted 28/09/2023, Published Online First 20/04/2024, Published 01/11/2024

Abstract

There are a variety of obesity surgeries and procedures in which obese individuals undergo. Primarily, they alter satiety and hunger hormone levels in the gut. Moreover, standard method of such procedures can result in long-term weight loss. Ghrelin (Ghr) is a gut hormone that regulates food intake. Laparoscopic Sleeve gastrectomy (LSG) is one method for treating morbid obesity (MO), which could modulate its secretion. The aim of the present study was to demonstrate the effect of sleeve gastrectomy on gut hormones. This study was conducted at Al-Basrah metabolic and Bariatric Surgery Center, Iraq, from December 2021 to March 2022, on 24 patients with morbid obesity treated by sleeve gastrectomy. The included patients were categorized into two main groups according to the periods of their operation. Glucagon –like peptide-1 GLP-1, Ghrelin, Leptin hormones and body mass index (BMI) were significantly decreased after surgery. There was a statistically significant correlation, found between patient's BMI and Leptin level after three months of the surgery, while a statistically significant correlation was found between patient's BMI and GLP-1 level before the surgery when BMI increase by one unit (Kg/m^2), the GLP-1 would be increased by half unit 0.45 (50%). The present study has reported that the three hormones GLP-1, Ghrelin and Leptin were decreased after three months of the surgery.

Keywords: Ghrelin, GLP-1, Leptin, Obesity, Sleeve Gastrectomy.

Introduction

Obesity is a complex morbid condition that is resulted from an imbalance between energy intake and energy consume [1]. It is considered a significant risk factor for several chronic and major health conditions, including cardiovascular disease, diabetes mellitus, musculoskeletal disorders, and cancer [2] .Bariatric surgery remains the most effective treatment for obesity and its complications [3]. Among the most common, are Roux en-Y bypass, sleeve gastrectomy (SG) and other types of procedures that aim to promote sustained weight loss like laparoscopic adjustable gastric banding, that result in a new hormonal weight set point can be achieved by primarily modifying the levels of gut hormones that are accountable for hunger and satiety [4]. However, the metabolic and physiological alteration after SG is not fully understood [5]. Recently, the scope of addressing obesity and its related medical issues has broadened to include bariatric endoscopy, serving as a link between guidance on diet and lifestyle modifications, medical therapies along with the proven efficacy of bariatric surgery [6]. The (SG) has gained popularity as both a

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https://doi.org/10.21123/bsj.2024.8990
P-ISSN: 2078-8665 - E-ISSN: 2411-7986
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first-stage procedure for high-risk patients and a standalone option to address morbid obesity. During the procedure, the majority of the stomach volume is vertically resected, removing the antrum, most of the body, and the entire fundus [7]. Using this procedure ensures a combination between volume restriction, a high-pressure system, and favourable hormonal [8]. Endocrine changes occur after operation, gut and metabolic hormones such as ghrelin and GLP-1. Ghrelin is an orexigenic hormone primarily secreted by the ga stric fundus, recently, it has been proposed that ghrelin, may also have effects on glucose homeostasis in addition to i ts effects on caloric intake[9]. So, ghrelin, as appetite-stimulating hormone, was initially discovered in the stomachs of both rats and humans[10]. Ghrelin is the only powerful orexigenic hormone that is released during fasting from the stomach, initiates feeding, and controls meal frequency. It is a pleiotropic and widely distributed hormone that affects many metabolic processes, including appetite regulation, body weight, glucose metabolism, heart pressure, adiposity, fertility, memory, learning, and reward processing [11,12]. GLP-1, a hormone discharged by enteroendocrine L-cells within the intestine in response to undigested food, promotes the secretion of insulin, suppresses the secretion of glucagon, and induces a feeling of fullness. After a (SG), both the postprandial hyperglucagonemia and rapid gastric emptying resulted in increased GLP-1 levels.[8] GLP-1 has various metabolic effects, including insulin secretion stimulation that depends on glucose levels, reduced gastric emptying, inhibited food intake, as well as increased natriuresis and diuresis[13].However, after SG, there are several factors that put patients at risk for developing nutritional deficiencies, including reduced dietary intake, decreased hydrochloric acid and intrinsic factor secretion, poor food choices and food intolerance[14]. Adipose tissue and small intestinal cells release the hormone leptin to control energy balance in relation to triglycerides via specific brain pathways, primarily the hypothalamus, to reduce appetite, which in turn reduces fat storage in adipocytes [15]. Leptin is an adipocytokine derived from the obesity gene (ob gene), which may play a role in long-term energy balance[16]. This hormone is mostly produced by adipocytes, and its major purpose is to control caloric intake. It primarily affects the hypothalamus in order to reduce food intake and increase energy expenditure. Adipose tissue secretes leptin, which can pass through the blood-brain barrier and enter the arcuate nucleus of the hypothalamus to regulate appetite[17,18]. Instead of macronutrients, adipose tissue mass controls plasma leptin concentrations[19].

Materials and Methods

Patients' selection and study protocol

This is a prospective study was conducted on 24 patients with morbid obesity by SG, with age ranged between 18-59 years for about 3-months duration at Al-Basrah metabolic and Bariatric Surgery Center, Iraq. All patients were subjected to questioner including: age, sex, marital state BMI, chronic disease and drugs history. Blood samples were collected from patients at preoperative time and three months. Hormonal assays including Ghrelin, GLP-1 and Leptin was performed before and three months of the surgery. According to the periods of their operation, the patients were classified into two groups. The first group was collected before the operation and the second group was collected after three months of the operation.

Blood sampling

Venous blood samples (2 ml) were collected from 24 SG patients preoperational and post operation of the same patients. Gel tubes (2ml) were used and then left for short time for blood clotting and then serum samples were obtained by centrifugation at room temperature at 3500 rpm for 10 minutes, 1.5 ml was used for hormonal determination.

Eppendorf safe-lock tubes (0.5ml) were used for dividing the serum before storage in deep freezing at -20°C.

Biochemical parameters measurement

The BMI was calculated as:

Body Mass Index=Weight (kg) / height (m^2).

The measured biochemical parameters are hormones including: Ghrelin, GLP-1and Leptin by ELISA (Elisa-kit. Biotin, China).

Statistical analysis

The Shapiro-Wilk test and Kolmogorov – Smirnov was performed for the numerical data whether the normal or non-normal distribution. The paired T-test analysis was used for parametric data measurement, while Wilcoxon Signed Ranks Test was performed for non- parametric data. The data were expressed as mean± standard deviation (SD). For the purpose of

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https://doi.org/10.21123/bsj.2024.8990
P-ISSN: 2078-8665 - E-ISSN: 2411-7986
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investigating correlations between numeric variables, Spearman Correlation Test was used. A value of $p \leq 0.05$ was considered as statistically significant.

Results and Discussion

As shown in Table 1, 14 males comprise 58.3%, while 10 females comprise 41.7%, minimum age is 18 years, maximum 59 years, mean of male age is 34.07 and mean of female age is (35.60). So, mean of male and female height is 174.43, 160.10 respectively, means of male and female weight are 135.79, 107.00 respectively.

Table 1. Baseline characteristics of participants enrolled in the present study.

Variable	Sex	Number	Mean	SD
Age	Male	14	34.07	9.72
	Female	10	35.60	14.30
Height	Male	14	174.43	10.15
	Female	10	160.10	5.09
Weight	Male	14	135.79	27.66
	Female	10	107.00	13.75

Table 2. The BMI, GLP-1, Ghrelin, and Leptin hormones levels in all studied groups.

Variable	Group I Before surgery N=24		Group II After surgery N=24		P. Value
	Mean ±SD	Median (Min-Max)	Mean ±SD	Median (Min-Max)	
GLP -1 hormone (ng/ml)	10.71±9.42	7.69 (0.81-30.50)	3.72±2.33	3.78 (0.38-10.50)	0.0001*
Ghrelin hormone (ng/ml)	1.92 ±1.65	1.54 (0.78-9.35)	1.28±0.65	1.19 (0.62-4.01)	0.0001*
Leptin hormone (ng/ml)	39.02±18.07	--------	25.67±7.89	--------	0.001**
BMI	44.84±8.83	43.38 (31.48-69.20)	36.89±7.85	35.58 (24.07-58.48)	0.0001*

* Wilcoxon Signed Ranks Test
** Paired t-test

As shown in Table .2 total GLP -1serum levels Significantly (p=0.0001) decreased from 7.69 (0.81-30.50) ng/mL before LSG to 3.78 (0.38-10.50) ng/mL after three months of LSG.

Table 2, also showed significantly (p=0.0001) decreased in the Ghrelin level before LSG was 1.54(0.78-9.35) ng/mL while three months after LSG was 1.19 (0.62-4.01) ng/ml. Moreover, Leptin before LSG was decreased significantly from 39.02±18.07 while after LSG was 25.67±7.89 and BMI before LSG was 43.38 (31.48-69.20) but after LSG was 35.58 (24.07-58.48) (p=0.001 and 0.0001) for leptin and BMI, respectively.

Table 3. Spearman's non-Parametric Correlations before surgery

		BMI 1	Ghrelin 1	GLP 1	Leptin 1
Age	R	0.019	0.091	-0.271-	-0.226-
	P-value	0.931	0.672	0.200	0.300
	N	24	24	24	23
BMI 1	R		0.038	0.453*	0.292
	P-value		0.861	0.026	0.176
	N		24	24	23
Ghrelin 1	R			-0.314-	0.090
	P-value			0.134	0.681
	N			24	23
GLP 1	R				0.329
	P-value				0.126
	N				23

Table 3 has reported that the only statistically significant correlation was found between patient's BMI and GLP-1 level before the surgery. As shown, the correlation was directly proportional with correlation coefficient 45.3%. That means when the BMI has increased by one unit (Kg/m^2), the GLP-1 would be increased by half unit 0.45 (50%).

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https://doi.org/10.21123/bsj.2024.8990
P-ISSN: 2078-8665 - E-ISSN: 2411-7986
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Table 4. Correlations between the hormonal aspect after surgery.

Variables	GLP 2 N=24		Ghrelin 2 N=24		BMI 2 N=24	
	R	P. Value	R	P. Value	R	P. Value
Ghrelin 2 Hormone (ng/ml)	-----	-------	-------	-------	0.058	0.788
GLP2 hormone (ng/ml)	------	-------	0.308-	0.143	0.095	0.658
Leptin2 hormone (ng/ml)	0.112	0.603	0.036-	0.869	0.641**	0.001

There was only one statistically significant correlation, found between patient's BMI and Leptin level after three months of the surgery, as presented in Table 4. It was direct proportional correlation with a coefficient of 64.1%.

The present study has suggested a significant correlation between GLP -1 hormone and BMI before surgery and between Leptin and BMI after surgery.

The current study focused on the interactions between gut peptides. The results of the present study have shown that the patient's group SG before operation had significantly higher GLP-1 level than patients' group after three months of operation. It has been suggested clarified the role of this hormone in several processes controlling body homeostasis, including glucose homeostasis and food intake regulation. A recent study concerning GLP-1in bariatric surgery have been published [20]. The current result was in agreement with previous finding [21] which have revealed that plasma GLP-1 level was higher after LSG than its level before surgery.

Fasting levels of GLP-1 were increase after bypass surgery as well as after diet-induced weight loss, and stay high after even one year of weight maintenance[22]. In any case, the mechanism underlying postprandial (GLP-1) changes after metabolic surgery remains partially understood [23].

Furthermore, the mechanisms involved a range of from electrogenic transporters, ion channel modulation and nutrient-activated G-protein coupled receptors that converge on the release machinery controlling hormone secretion. Elucidation of these mechanisms will provide much needed insight into postprandial physiology and identify tractable dietary approaches to potentially manage nutrition and satiety by altering the secreted gut hormone profile [24]. The provided information hinted at the possibility of intestinal physiology adjusting to fast nutrient intake via elevating the count of enteroendocrine cells or enhancing the nutrient sensitivity of the current enteroendocrine cell population. There is a study that has reported an increase in L-cell count by SG in rats, A number of studies demonstrated an increase in the number of GLP-1–secreting enteroendocrine cells (EECs) after SG without any gross morphological changes in the intestinal epithelium. The (EECs), along with numerous other intestinal cell types (goblet cells, enterocytes, Paneth cells), are dispersed separately throughout the intestinal epithelium and release gut peptides [25,26], but another did not, Clinical studies have demonstrated that both Roux-en-Y gastric bypass (RYGB) and vertical sleeve gastrectomy (VSG) have resulted in comparable speedy enhancements in body mass and glycemic control. The available data points towards the fact that the advantageous impact on glucose homeostasis after RYGB may be attributed to the adaptive tissue growth and restructuring of intestinal glucose processing, however, it is uncertain if these adaptive changes also take place after sleeve gastrectomy[27]. Anyway, the reasons for these discrepant findings after VSG are unclear; they have no agreement with the present study.

Of the many available options, (RYGB) and (VSG) are currently the most widely used procedures. RYGB and VSG have very different anatomical restructuring but both surgeries are effective, to varying degrees, at inducing weight loss. However, Bariatric-metabolic surgery has multi-faceted effects that cause alterations in various gut hormones, including ghrelin. To devise a customized strategy for patients and identify potential adverse factors that can result in bariatric surgery failure, a comprehension of the biological modifications and

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P-ISSN: 2078-8665 - E-ISSN: 2411-7986
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behaviors that occur post the surgical procedure is highly essential [28, 29]. Additionally, removal of the stomach fundus decreases the number of cells producing ghrelin – the "hunger hormone". The reduction in plasma ghrelin level promotes the feeling of satiety and restrains food intake [30]. This is supported by the present study which reported postoperative decreases in serum ghrelin of the follow-up after 3 months.

Even though SG is commonly perceived as a surgery that limits, it could trigger metabolic changes that are believed to be linked to the reduction of the secretion of the appetite-stimulating hormone ghrelin. This is why the decrease of ghrelin level that was observed in our study.

The reduction of ghrelin-producing cells, i.e., the removal of oxyntic glands (located in the gastric fundus), may simply account for the observed difference resulting from the sleeve procedure [31].

Though, a comparison of Leptin level before and after three months of surgery have been shown a significant decreasing level.

Salman *et al*. [32] a study has discovered that there was a significant reduction in serum leptin levels during a 1-year follow-up of patients who underwent bariatric surgery, in agreement with our findings. Similarly, other study has reported a significant decline in serum leptin levels one year after RYGB and LSG surgeries, with no notable distinction between the two procedures[33]. Additional report exhibited the reduction of Leptin has been reported in all the bariatric procedures (RYGB, LSG, LAGB), and it has been linked directly with weight loss[34]. Postoperative improvements in endogenous Leptin-resistance status can be attributed to the positive effect of RYGB and LSG on serum leptin levels, which is commonly associated with obesity.

In respect of BMI measurement, the obese patients in the current study have shown the significantly lower level after surgery in comparison with the same patients before three months. Our data was in line with a recent study by Salminen *et al*. (2018) [35]. The observed difference resulting from the sleeve procedure may simply be due to the reduction of ghrelin-producing cells, specifically the removal of oxyntic glands. There is a significant correlation between GLP-1 and BMI were noted in patients before surgery. A significant correlation between leptin and BMI were noted in SG group at 3 months after surgery (r=0.641) [33]. In fact, "Leptin resistance" is a feature of extreme obesity, and bariatric surgery may boost Leptin sensitivity, which may aid in weight loss[36]. Obese individuals who exhibit resistance to the satiety-inducing impacts of Leptin may have already experienced this in the cases of operated patients who slowly regain weight [37].

Conclusion

The present study has reported SG is an effective treatment for morbid obesity because it has a favorable risk-benefit ratio. Firstly, as obesity considered a significant risk factor for several chronic and major health illness. So, reduction in BMI can decrease these risks. Secondly, decrease in Ghrelin and Leptin hormones lead to reduction in the satiety.

Acknowledgment

We thank the research assistants at Al-Basrah metabolic and Bariatric Surgery Center in Iraq for their assistance in samples collection and peptide hormone analysis in the laboratory for Biology Department / College of science /Basrah University for their collaboration.

Authors' Declaration

- Conflicts of Interest: None.
- We hereby confirm that all the Figures and Tables in the manuscript are ours. Furthermore, any Figures and images, that are not ours, have been included with the necessary permission for re-publication, which is attached to the manuscript.
- Authors sign on ethical consideration's approval.
- Ethical Clearance: The project was approved by the local ethical committee at University of Basrah.

Authors' Contribution Statement

This work was carried out in collaboration between all authors. F. M. A. diagnosis the cases then collected the samples and R. M. A. doing the tests and wrote and edited the manuscript with revisions idea, H. S. K., analysis the data with revisions idea. All authors read and approved the final manuscript.

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2024, 21(11): 3438-3445
https://doi.org/10.21123/bsj.2024.8990
P-ISSN: 2078-8665 - E-ISSN: 2411-7986
Baghdad Science Journal

دور الجانب الهرموني للقناة الهضمية لمرضى عراقيين خاضعين لعملية قص المعدة

رافدة مجيد العامري[1]، هناء سلمان كاظم[2]، فالح محسن علي [3]

[1] قسم العلوم الاساسية، كلية طب الاسنان ، جامعة البصرة، البصرة، العراق.
[2] قسم التحليلات المرضية، كلية العلوم ، جامعة البصرة، البصرة، العراق.
[3] مركز جراحة السمنة ، مستشفى الصدر التعليمي، دائرة صحة البصرة ،البصرة ،العراق.

الخلاصة

ان هناك عدة انواع من العمليات الجراحية التي تجرى للاشخاص ذوي الاوزان العالية لعلاج السمنة المفرطة عندهم . وذلك عن طريق التغيير الحاصل في مستوى الهرمونات المعوية المسؤلة عن الجوع والشبع . ونتيجة لهذه العمليات سيحصل فقدان كبير بالوزن بعد اجراء الجراحة الناظورية وهو من خيارات علاج السمنة المرضية ونتيجة لذلك فأن افراز هرمون الكريلين ممكن ان يتعدل او يتغير. الهدف من الدراسة هو توضيح او اكتشاف اي تأثير لعمليات تكميم المعدة على بعض الهرمونات المعوية. لقد قمنا بأجراء دراسة مستقبلية في مركز البصرة لجراحة الأيض والسمنة ، في جنوب العراق من كانون الأول (ديسمبر) 2021 إلى آذار (مارس) 2022 ، على 24 مريضاً كانو يعانون من السمنة المرضية واجريت لهم عمليات تكميم المعدة. تم تصنيف المرضى المشمولين إلى مجموعتين رئيسيتين حسب فترات العملية. وقد عولجت النتائج باستخدام الإصدار 25 من برنامج SPSS (شركة SPSS) للحسابات الإحصائية. و قمنا بحساب قيمة P. النتائج: انخفض معدل GLP -1 و Ghrelin و Leptin و BMI بشكل ملحوظ بعد الجراحة. لم نلاحظ اي تأثير معنوي سوى علاقة ذات دلالة إحصائية واحدة بين مؤشر كتلة الجسم ومستوى اللبتين للمريض بعد ثلاثة أشهر من الجراحة ، بينما كان هناك ارتباط معنوي بين مؤشر كتلة الجسم ومستوى GLP-1 قبل الجراحة. الاستنتاج نستنتج أن الهرمونات الثلاثة GLP-1 و Ghrelin و Leptin تنخفض بعد ثلاثة أشهر من الجراحة ويزداد مؤشر كتلة الجسم بمقدار وحدة واحدة (Kg / m2) ، وسوف يزيد GLP-1 بمقدار نصف وحدة 0.45 (50٪).

الكلمات المفتاحية: الكريلين ،اللبتين، كلوكاكون لايك ببتايد ، السمنة ، تكميم المعدة.