Original Article

Effect of Bariatric Surgery on Serum Glucagon like Peptide-1 Concentration and Metabolic Parameters in Obese Type 2 Diabetics.

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Abstract:

Background: Bariatric surgery has proven an effective anti-diabetic treatment modality. It results in significant weight loss and improvement in health and quality of life. Roux-en-Y gastric bypass (RYGB) surgery has been shown to be associated with lower overall mortality for subjects with T2DM. **Objective:** Assess the effect of RYGB surgery on serum glucagon like peptide-1 (GLP-1) concentration, metabolic and biochemical parameters pre and postoperatively in type 2 diabetics. **Methods:** This prospective study comprised 50 subjects divided into 3 groups. **Group I:** 20 obese diabetics (BMI > 35) that underwent Roux-en-y gastric bypass surgery (RYGB); **group II:** 20 lean subjects with T2DM and **group III:** 10 lean subjects as healthy controls. BMI, fasting blood glucose and 2hPP glucose, HbA1c, lipid profile, fasting serum insulin, fasting and 2hP plasma GLP-1were estimated. In group 1, tests were done pre and three months postoperatively. Insulin resistance was quantified using HOMA-IR. **Results:** Fasting and 2hPP GLP-1 levels were lower in group 1 compared to the other 2 groups respectively (FGLP-1: 4.33±1.10vs. 5.21±0.89, 6.14±0.42 ng/ml, p<0.001), 2hPP GLP-1 (4.94 ± 1.17 vs. 6.04 ± 0.99, and 9.32±0.97ng/ml, p<0.001); also the difference between groups 2 and 3 was statistically significant. Fasting and 2hPP GLP-1 levels showed significant negative correlation with all metabolic parameters except HDL-C, which showed a positive correlation (p<0.001). Regarding the postoperative results, we found an increase in the fasting GLP-1 levels (4.53 ±1.02vs 4.3±1.1 ng/ml, *p*=0.09), 2hPP GLP-1 levels (9.79±1.99 vs 4.9 ± 1.2 ng/ml, p<0.001) and HDL-C. Also, there was a statistically significant decrease in all metabolic parameters. Sixty percent of the patients stopped their anti-diabetic medications postoperatively. **Conclusion:** RYGB surgery resulted in a statistically significant reduction in BMI and all metabolic parameters. Fasting & 2hPP GLP-1 levels were low in diabetics. Postoperative 2hPP GLP-1 levels increased, possibly responsible for the metabolic benefits.

Keywords: Obesity, Type 2 Diabetes Mellitus (T2DM), glucagon like peptide-1 (GLP-1), HOMA-IR, Roux-en-y gastric bypass surgery (RYGB).

Introduction:

More people are diagnosed with type 2 diabetes mellitus due to increase in prevalence of obesity across continents. Conventional therapies seem to be unable to stop the progression of T2DM and certainly do not cure the disease[(1)]. In contrast, bariatric surgery has proven an effective anti-diabetic treatment modality. Additionally, it results in significant weight loss and improvement in health and quality of life[(2)]. Also, RYGB has been shown to be associated with lower overall mortality for subjects with T2DM. Currently the most commonly used form of bariatric surgery is RYGB[(3)].

Remission of T2DM and marked reduction of insulin resistance has been shown to occur a few days following RYGB operation, before any significant weight loss has taken place, indicating that the operation itself brings about acute endocrine changes improving glucose homeostasis[(4)].

Although the precise mechanisms through which type 2 diabetes remission occurs following metabolic surgery remain to be fully elucidated, it is clear that endocrine changes due to rearrangement of the gut

anatomy play an important role. Of the potential hormones involved in the endocrine changes are the in cretin hormones. Several studies have examined changes in levels of incretin hormones following bariatric surgery[5]. Stimulated GIP levels after a test meal were reported by Laferrère and colleagues to be increased 1 month after gastric bypass[6]. Several groups have reported decreased ghrelin levels after RYGB, which may partly account for the improved glycemia[7]. Also elevated PYY levels after gastric bypass have been reported by Valderas et al[8]. Early studies reported an increased fasting and postprandial enteroglucagon (previously used as a marker for GLP-1) after both gastric bypass and jejunoileal bypass[7]. Subsequent changes in both fasting and postprandial GLP-1 levels have been reported in several studies[5]. GLP-1 is known to stimulate insulin secretion from pancreatic *β* cells; inhibit glucagon secretion from pancreatic *α* cells and decrease gastrointestinal motility, appetite, and food intake and body weight[3].

Our aim was to assess the effect of Roux-en-Y gastric bypass surgery (RYGB) on serum glucagon like peptide-1(GLP-1) concentration and changes in metabolic and biochemical parameters pre and postoperatively.

Research design and methods:

Study Subjects:

The current prospective case-control study was conducted from April 2012 to December 2012 and this study was approved by the internal review board of Ain Shams University. All subjects provided written informed consent to undergo various examinations, and provide blood samples.

Our study was conducted on 50 subjects divided into 3 groups. **Group I:** 20 obese subjects with T2DM (BMI > 35) reviewed before and after Roux-en-y gastric bypass surgery; **Group II:** 20 lean subjects with T2DM (BMI<25 kg/m²) and **Group III:** 10 lean subjects as healthy controls. All patients in group 1 were on medical treatment, 9 of them were on insulin plus oral anti-diabetics while the remaining 11 were on oral anti-diabetics alone. All patients in group 2 were on medical treatment, 7 were on insulin plus oral anti-diabetics while the remaining 13 were on oral anti-diabetics alone. In group1, tests were done preoperative and three months after surgery.

All patients were recruited from the outpatient clinic of both the endocrinology and the bariatric and metabolic surgery departments of Ain Shams University Hospitals. All were subjected to full medical history emphasizing the duration of diabetes mellitus and the type of treatment, along with thorough clinical examination including blood pressure and anthropometric measurements (Height, weight, Body Mass Index (BMI) and Waist/Hip ratio). Subjects with history of impaired renal function, impaired liver function, heart failure, cancer, autoimmune disease, pregnancy, and alcohol or drug abuse were excluded. Also, patients using a dipeptidyl peptidase-4 inhibitor or a GLP-1 agonist therapy were excluded.

BMI was calculated as body weight in kilograms divided by the height in meters squared (kg/m^2) and waist circumference was measured at the highest point of the iliac crest at minimal respiration to the nearest 0.1 cm. Serum lipid concentrations were assayed using Quantitative Enzymatic Colorimetric Determination for total and HDL cholesterol and triglycerides in plasma (Stanbio Cholesterol Liquicolor, Procedure NO. 1010). LDL cholesterol was calculated using the Friedewald equation as follows: LDL-C = (Total cholesterol- HDL-C) + Triglycerides/5[9]. Fasting blood glucose (FBG) and 2hPP plasma glucose were measured using an automated glucose oxidase method using Behring Diagnostics Reagents (SVR Glucose Test; Behring, La Jolla, CA). HbA1c is assayed by Stanbio Procedure No.0350 “Quantitative colorimetric determination of Glycohemoglobin in blood”. Enzyme linked immunoassay (ELISA) was used for in vitro quantitative measurements of fasting plasma insulin (BioSource INS-EASIA Kit. Catalogue number: KAP1251). Insulin resistance was estimated by HOMA-IR and was defined as fasting serum insulin (μU/ml)×FPG (mmol/l)/22.5[10].

Sample Collection: Subjects were instructed to fast 8 hours, 7 ml of venous blood was collected by venipuncture under complete aseptic conditions, and then the subjects were instructed to continue fasting. After completing 14 hours of fasting, another 3 ml of venous blood were collected. The first sample was used for measurement of FPG, fasting insulin,

fasting GLP-1 and HbA_{1c}. The second sample was used for measurement of lipid profile (total cholesterol, HDL-C, LDL-C and triglycerides), and it was frozen at -20°C until assayed. Next, 75 gm glucose were ingested by all subjects after the second venipuncture then two hours later another 3 ml of venous blood were collected to measure two hours post prandial plasma glucose (2hPPG) and 2hPPGLP-1. Samples that were used for estimation of GLP-1 were stored at -70 C° for subsequent assay in plasma using DRG Glucagon like peptide-1 (Human,Rat and Mouse) ELISA (EIA-4141) kit, USA (Porstmann and Kiessig,1992)[11].

Surgical Technique: The technique used in this study was RYGB, which combines restrictive and malabsorptive mechanisms. A vertical gastric pouch (20–30 ml) was constructed with surgical staples in the lesser curvature of the stomach. Gastrojejunostomy adjustment was performed using a 32-G French tube. Reconstruction was performed by RYGB with an alimentary limb measuring 100 cm and a biliopancreatic limb of 50 cm from the ligament of Treitz.

Statistical Analysis:

Data analysis was performed using the SPSS program, v.12. Data were expressed as mean ± standard deviation (SD) for parametric data and as median and interquartile range (IQR) for non-parametric data respectively. Parametric data were analyzed using one-way analysis of variance (ANOVA) for the comparison of three groups. Pearson's correlation coefficient (r) test was used for correlating data. Independent-samples T test of significance was used when comparing between two groups. Statistical significance was detected at p value <0.05, while p <0.001 was accepted as highly significant.

Results:

The studied groups were matched regarding age and gender, and there was no statistically significant difference between group I and group II regarding diabetes duration and the use of anti-diabetic medications (Table 1). There was a highly statistical significant difference between group I and both group II and group III regarding BMI, while there was no statistical difference between (group II & group III). Regarding fasting and 2hPP GLP-1 levels, there were highly statistical significant differences among the three groups (table 1). Fasting GLP-1 was lower in obese diabetics than lean diabetics (4.33 ± 1.10 versus 5.21 ± 0.896 ng/ml, p=0.012), as well as 2hrsPP GLP-1 levels (4.94±1.17 versus 6.04±0.995 ng/ml, p= 0.006). All criteria of subjects included in the study are summarized in (table 1).

We also found a highly significant negative correlation between both fasting as well as 2 hours postprandial GLP-1 and all of the following: BMI, waist/ hip ratio, blood pressure, FPG, 2hPPG, HbA1c, fasting insulin, HOMA IR, total cholesterol, triglycerides and LDL-C, as well as the duration of diabetes mellitus. There was a highly significant positive correlation between fasting GLP-1 and HDL-C (Table 2).

Regarding the postoperative results, we found an increase in the fasting GLP-1 levels (from 4.3±1.1 to 4.53±1.02 ng/ml), not reaching statistical significance (p=0.095), while there was a highly statistical significant increase in the 2hPP GLP-1 level (from 4.9±1.2 to 9.79±1.99ng/ml, p<0.001). Also, there was a highly statistical significant decrease in BMI, waist/hip ratio, SBP, DBP, FPG, 2hPPG, HbA1c, fasting insulin, HOMA-IR, total cholesterol, triglycerides and LDL-C, while levels of HDL-C were increased significantly (Table 3).

BMI decreased significantly after surgery (from 47.3±4.2 to 42.10±3.06kg/m2 p<0.001). However, there was still a highly statistical significant difference between group I and both groups II and III. FPG, 2hPPG, fasting insulin, HOMA-IR, total cholesterol, triglycerides and LDL-C were significantly decreased to levels similar to healthy controls. After surgery, fasting GLP-1 levels were still low in comparison to the other 2 groups (4.53±1.02 vs. 5.21 ± 0.89, 6.14 ± 0.47 ng/ml, p < 0.001). The 2hPP GLP-1 levels were elevated to levels similar to healthy controls (9.79±1.999 vs. 9.32±0.97 ng/ml, p>0.05); (Table 4).

Our results also demonstrated a highly statistical significant reduction regarding the percentage of patients using anti-diabetic medications; as all patients (100%) were on treatment preoperatively, while only 40% were receiving anti-diabetic medications postoperatively. There was a highly significant decrease in use of insulin plus oral therapy from 45% to only 10%, as well as a reduction in the percentage of patients who use oral medications from 55% to 30% (Figure 1). There was also a statistical significant decrease in the use of anti-hypertensive and lipid lowering medications (Figure 2).

Table (1): Comparison among the three studied groups regarding the baseline results of different variables:

Variables		Group I (pre-op.)	Group II	Group III	ANOVA		Tukey's test	
		Mean ± SD	Mean ± SD	Mean ± SD	f	P-value	Comp.	P-value
Age (years)		46.10 ±4.84	45.45±5.48	44.60±4.50	0.299	0.743	-----	-----
SBP (mmHg)		138.75±7.59	127.25 ± 8.66	120.00±4.08	23.562	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.043*
DBP (mmHg)		90.75±4.06	82.50±7.695	75.50± 4.97	23.634	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.011*
BMI (kg/m²)		47.30±4.19	22.50±1.28	21.30±1.34	470.25	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.526
WHR		0.97±0.02	0.87±0.03	0.74±0.02	335.35	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	<0.001**
FPG (mg/dl)		145.85±19.19	136.40±17.12	85.30±6.60	46.843	<0.001**	I&II	0.181
							I&III	<0.001**
							II&III	<0.001**
2hPP.PG (mg/dl)		208.9±33.19	183.6±51.33	122.0±7.44	16.600	<0.001**	I&II	0.111
							I&III	<0.001**
							II&III	0.001*
HbA1c (%)		9.57±1.51	7.90±1.48	5.300±0.61	33.352	<0.001**	I&II	0.001*
							I&III	<0.001**
							II&III	<0.001**
Fasting Insulin (mU/ml)		20.90±6.84	13.00±2.13	5.96±0.53	38.199	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.001*
HOMA IR		7.83±3.61	4.46±1.28	1.26±0.21	25.564	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.004*
Total Cholesterol (mg/dl)		220.75±34.82	195.10±27.21	149.20±17.00	20.221	<0.001**	I&II	0.020*
							I&III	<0.001**
							II&III	0.001*
Triglycerides (mg/dl)		199.35±53.73	172.55±52.56	103.70±16.71	13.114	<0.001**	I&II	0.197
							I&III	<0.001**
							II&III	0.002*
HDL- Chol. (mg/dl)		34.10±9.36	39.25±5.02	59.20±7.269	38.799	<0.001**	I&II	0.085
							I&III	<0.001**
							II&III	<0.001**
LDL- Chol. (mg/dl)		173.92±26.82	152.74±18.05	116.62±15.15	23.522	<0.001**	I&II	0.009*
							I&III	<0.001**
							II&III	<0.001**
F GLP-1 (ng/ml)		4.33±1.10	5.21 ±0.896	6.14±0.47	13.250	<0.001**	I&II	0.012*
							I&III	<0.001**
							II&III	0.033*
2hPP. GLP-1 (ng/ml)		4.94±1.17	6.04 ±0.995	9.320±0.97	57.105	<0.001**	I&II	0.006*
							I&III	<0.001**
							II&III	<0.001**
Duration of DM (years)		4.78±1.86	4.38±1.597	---------	**T-Test**		**t**	0.730
							P-value	0.470
Gender N (%)	**Male** 16 (32%)	5 (25%)	7 (35%)	4 (40%)	**Chi-Square**		**X^2**	0.835
	Female 34 (68%)	15 (75%)	13 (65%)	6 (60%)			**P-value**	0.659
Antidiabetic medications N (%)	**Insulin+oral**	9 (45%)	7 (35%)	---------	**Chi-Square**		**X^2**	0.104
	Oral only	11 (55%)	13 (65%)				**P-value**	0.746

Variables		Group I (pre-op.)	Group II	Group III	ANOVA		Tukey's test	
		Mean ± SD	Mean ± SD	Mean ± SD	f	P-value	Comp.	P-value
Age (years)		46.10 ±4.84	45.45±5.48	44.60 ± 4.50	0.299	0.743	-----	-----
SBP (mmHg)		138.75±7.59	127.25±8.66	120.00±4.08	23.562	<0.00**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.043*
DBP (mmHg)		90.75±4.06	82.50±7.695	75.50 ± 4.97	23.634	<0.001**	I&II	<0.001**
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BMI (kg/m²)		47.30±4.19	22.50±1.28	21.30 ± 1.34	470.25	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.526
WHR		0.97±0.02	0.87±0.03	0.74 ± 0.02	335.35	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	<0.001**
FPG (mg/dl)		145.85±19.19	136.40±17.12	85.30 ± 6.60	46.843	<0.001**	I&II	0.181
							I&III	<0.001**
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2hPP.PG (mg/dl)		208.9±33.19	183.6±51.33	122.0± 7.44	16.600	<0.001**	I&II	0.111
							I&III	<0.001**
							II&III	0.001*
HbA1c (%)		9.57±1.51	7.90±1.48	5.300 ± 0.61	33.352	<0.001**	I&II	0.001*
							I&III	<0.001**
							II&III	<0.001**
Fasting Insulin (mU/ml)		20.90±6.84	13.00±2.13	5.96± 0.53	38.199	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.001*
HOMA IR		7.83±3.61	4.46±1.28	1.26± 0.21	25.564	<0.001**	I&II	<0.001**
							I&III	<0.001**
							II&III	0.004*
Total Cholesterol (mg/dl)		220.75±34.82	195.10±27.21	149.20±17.00	20.221	<0.001**	I&II	0.020*
							I&III	<0.001**
							II&III	0.001*
Triglycerides (mg/dl)		199.35±53.73	172.55±52.56	103.70±16.71	13.114	<0.001**	I&II	0.197
							I&III	<0.001**
							II&III	0.002*
HDL- Chol. (mg/dl)		34.10±9.36	39.25±5.02	59.20± 7.269	38.799	<0.001**	I&II	0.085
							I&III	<0.001**
							II&III	<0.001**
LDL- Chol. (mg/dl)		173.92±26.82	152.74±18.05	116.62±15.15	23.522	<0.001**	I&II	0.009*
							I&III	<0.001**
							II&III	<0.001**
F GLP-1 (ng/ml)		4.33±1.10	5.21 ±0.896	6.14 ± 0.47	13.250	<0.001**	I&II	0.012*
							I&III	<0.001**
							II&III	0.033*
2hPP. GLP-1 (ng/ml)		4.94±1.17	6.04 ±0.995	9.320 ± 0.97	57.105	<0.001**	I&II	0.006*
							I&III	<0.001**
							II&III	<0.001**
Duration of DM (years)		4.78±1.86	4.38±1.597	---------	**T-Test**		**t**	0.730
							P-value	0.470
Gender N (%)	**Male** 16 (32%)	5 (25%)	7 (35%)	4 (40%)	**Chi-Square**		**X^2**	0.835
	Female 34(68%)	15 (75%)	13 (65%)	6 (60%)			**P-value**	0.659
Antidiabetic medications N(%)	**Insulin+oral**	9 (45%)	7 (35%)	--------	**Chi-Square**		**X^2**	0.104
	Oral only	11 (55%)	13 (65%)				**P-value**	0.746

Group I: obese T2DM **Group II:** Lean T2DM **Group III:** Lean healthy control

Table (2): Correlation between fasting GLP-1, 2hsPP GLP-1 levels and the different variables:

Correlations				
Variables	**F GLP-1 (ng/ml)**		**2hPP GLP-1 (ng/ml)**	
	R	**P-value**	**r**	**P-value**
Age (years)	0.031	0.897	-0.002	0.994
Duration of diabetes(years)	-0.981	<0.001**	-0.693	<0.001**
SBP (mmHg)	-0.819	<0.001**	-0.745	<0.001**
DBP (mmHg)	-0.662	0.001*	-0.644	0.002*
BMI (kg/m²)	-0.818	<0.001**	-0.742	<0.001**
WHR	-0.754	<0.001**	-0.670	<0.001**
FPG (mg/dl)	-0.823	<0.001**	-0.731	<0.001**
2hPP.PG (mg/dl)	-0.792	<0.001**	-0.752	<0.001**
HbA1c (%)	-0.833	<0.001**	-0.760	<0.001**
Fastig insulin (mU/ml)	-0.832	<0.001**	-0.760	<0.001**
HOMA IR	-0.826	<0.001**	-0.756	<0.001**
Total Cholest (mg/dl)	-0.806	<0.001**	-0.748	<0.001**
Triglycerides (mg/dl)	-0.825	<0.001**	-0.752	<0.001**
HDL-Chol. (mg/dl)	0.807	<0.001**	0.721	<0.001**
LDL-Chol. (mg/dl)	-0.773	<0.001**	-0.726	<0.001**

***** Significant difference , ****** highly significant difference.

Table (3): Comparison between the preoperative and 3 months post-RYGP results in obese T2D:

Variables	**Pre-operative**	**Post-operative**	**Paired Differences**		**Paired t-test**	
	Mean ± SD	**Mean ± SD**	**Mean**	**SD**	**t**	**P-value**
SBP (mmHg)	138.8 ± 7.6	123.750 ± 8.252	15.000	2.294	29.240	<0.001**
DBP(mmHg)	90.8 ± 4.1	83.000 ± 8.176	7.750	5.250	6.601	<0.001**
BMI(kg/m²)	47.3 ± 4.2	42.100 ± 3.059	5.200	1.508	15.422	<0.001**
WHR	0.97±0.02	0.911 ± 0.029	0.060	0.016	17.245	<0.001**
FPG(mg/dl)	145.9 ± 19.2	98.250 ± 16.032	47.600	6.151	34.609	<0.001**
2hPP.PG(mg/dl)	208.9 ± 33.2	137.850 ± 14.908	71.050	20.715	15.339	<0.001**
HbA1c (%)	9.6 ± 1.5	7.110 ± 1.770	2.460	0.339	32.419	<0.001**
F. insulin (mU/ml)	20.9 ± 6.8	7.250 ± 1.997	13.650	4.902	12.453	<0.001**
HOMA IR	7.8 ± 3.6	1.829 ± 0.806	5.998	2.815	9.527	<0.001**
T.Cholest(mg/dl)	220.8 ± 34.8	173.250 ± 32.197	47.500	11.678	18.191	<0.001**
TG(mg/dl)	199.4 ± 53.7	143.950 ± 42.465	55.400	14.926	16.599	<0.001**
HDL-Chol.(mg/dl)	34.1 ± 9.4	48.050 ± 10.870	-13.950	2.685	-23.237	<0.001**
LDL-Chol.(mg/dl)	173.9 ± 26.8	135.000 ± 26.253	38.920	12.159	14.315	<0.001**
F GLP-1(ng/ml)	4.3 ± 1.1	4.525± 1.02	-0.195	0.150	-2.025	0.095
2hPPGLP1(ng/ml)	4.9 ± 1.2	9.785 ± 1.999	-4.845	0.842	-25.734	<0.001**

***** Significant difference, ****** highly significant difference.

Table (4): Comparison among the three studied groups 3 months after RYGB regarding the different variables:

Variables	Group I (post-operative)	Group II	Group III	ANOVA		Tukey's test	
	Mean ± SD	Mean ± SD	Mean ± SD	f	P-value	Comp.	P-value
SBP (mmHg)	123.75±8.25	127.25±8.66	120.00±4.08	2.987	0.060	I&II	0.340
						I&III	0.436
DBP (mmHg)	83.0±8.18	82.50±7.69	75.50±4.97	3.798	0.030*	I&II	0.976
						I&III	0.033*
BMI (kg/m²)	42.10±3.06	22.50±1.28	21.300±1.337	502.56	<0.001**	I&II	<0.001**
						I&III	<0.001**
WHR	0.91±0.03	0.87±0.03	0.74±0.02	124.25	<0.001**	I&II	<0.001**
						I&III	<0.001**
FPG (mg/dl)	98.25±16.03	136.40±17.12	85.30±6.60	49.327	<0.001**	I&II	<0.001**
						I&III	0.081
2hPP.PG (mg/dl)	137.85±14.91	183.60±51.33	122.00±7.44	14.125	<0.001**	I&II	<0.001**
						I&III	0.460
HbA1c (%)	7.11±1.77	7.90±1.45	5.30±0.61	10.333	<0.001**	I&II	0.220
						I&III	0.008*
Fasting Insulin (mU/ml)	7.25±1.99	13.00±2.128	5.96±0.53	67.133	<0.001**	I&II	<0.001**
						I&III	0.187
HOMA IR	1.83±0.81	4.462±1.28	1.26±0.21	52.370	<0.001**	I&II	<0.001**
						I&III	0.294
Total Chol. (mg/dl)	173.25±32.19	195.10±27.21	149.20±17.00	9.409	<0.001**	I&II	0.043*
						I&III	0.076
Triglyceride (mg/dl)	143.95±42.47	172.55±52.56	103.70±16.71	8.421	0.001*	I&II	0.106
						I&III	0.054
HDL- Chol. (mg/dl)	48.05±10.87	39.25±5.01	59.20±7.27	19.903	<0.001**	I&II	0.004*
						I&III	0.003*
LDL- Chol. (mg/dl)	135.00±26.25	152.74±18.05	116.62±15.15	10.003	<0.001**	I&II	0.030*
						I&III	0.077
F GLP-1 (ng/ml)	4.525±1.02	5.21±0.896	6.14±0.47	11.214	<0.001**	I&II	0.048*
						I&III	<0.001**
2hPP. GLP-1 (ng/ml)	9.79±1.999	6.04±0.995	9.32±0.97	35.529	<0.001**	I&II	<0.001**
						I&III	0.699

***** Significant difference, ****** highly significant difference.
Group I: obese T2DM Group II: Lean T2DM Group III: Lean healthy control

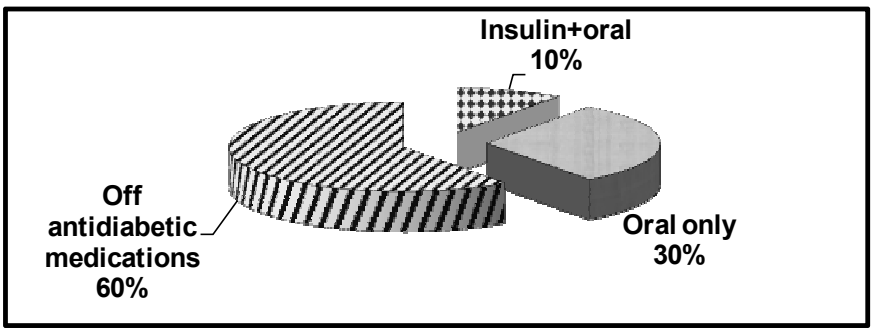

Figure (1): Descriptive figure for the percentage of usage of the anti-diabetic medications among obese T2DM 3 months post- RYGB surgery

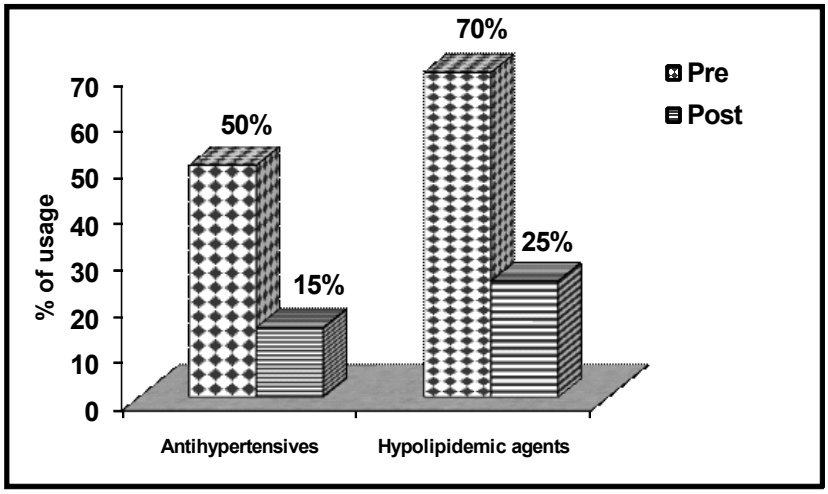

Figure (2): Comparison between the preoperative and postoperative use of antihypertensive and hypolipidemic medications in obese type 2 diabetics.

Discussion:

The results of our study showed a highly statistically significant decrease of the fasting and 2hPPGLP-1 levels among type 2 diabetic patients compared with those of control subjects. These results were in agreement with those of Toft-Nielsen et al.[12] and Vilsboll et al.[13] who reported lower postprandial GLP-1 levels in patients with type 2 diabetes compared with normal oral glucose–tolerant subjects. However, some authors pointed out that the GLP-1 levels were not reduced in patients with type 2 diabetes mellitus in comparison to control subjects[14,15].

We found that fasting and 2hPP GLP-1 levels among obese diabetic patients were significantly lower than those of lean diabetics. This was supported by our finding of a negative correlation between GLP-1 levels and body mass index, which was of a high statistical significance (p<0.001).These results were in agreement with those of Toft-Nielsen et al.[12] and Vollmer et al.[15] who found a decreased GLP-1 response to oral glucose load with increasing BMI. Furthermore, the fasting GLP-1 concentrations were found to be inversely related to BMI (P = 0.003) by Greenfield et al.[16] On the other hand, Yamaoka-Tojo et al.[17] found a statistically non significant positive correlation between fasting GLP-1 concentrations and BMI (P = 0.747), and in the study of Lee et al.[18] the postprandial GLP-1 levels were not correlated with BMI. Moreover, Kozawa et al.[19] showed that incretin secretion did not differ between Japanese obese and non-obese patients with type 2 diabetes. They explained their results as the decreased secretion of GLP-1 in Caucasian subjects with T2DM may be related to insulin resistance and obesity[20], but the obesity and insulin resistance in their patients did not reach levels sufficient to influence GLP-1 secretion. Alternatively, it may be that total GLP- 1 levels in Japanese subjects are naturally low compared with those in Caucasian subjects and not parallel to insulin secretion[19].

The results of our study showed a highly statistical significant negative correlation between both fasting and 2hPP GLP-1 levels and FBG, HbA1c, fasting insulin and HOMA IR (p<0.001). Zhang et al.[21] found that both fasting and postprandial GLP-1 levels were reduced in type 2 diabetic patients compared to subjects with normal glucose tolerance (P<0.005), and their levels were inversely proportional with the HOMA-IR. On the other hand, Lee et al.[18] examined Japanese newly diagnosed T2DM patients with relatively mild hyperglycemia (HbA1c levels of less than 7.5%), none of which were receiving any glucose-lowering medications. The authors demonstrated that, although GLP-1 was lower in the T2DM group than in the NGT group at 120 minutes in the OGTT, there were no statistical significant differences, and GLP-1 levels were not correlated with FPG, HbA1c or HOMA-IR. Although the cause and the mechanism for this discrepancy are not clear until now, the possible factors for it may be: course of disease, sample size and influence of treatment[21].

We also found that GLP-1 concentrations were inversely related to SBP, DBP, BMI, total cholesterol, LDL-cholesterol and triglyceride levels but directly related to HDL- cholesterol, all the previous correlations were of a highly statistical significant importance (p<0.001). These results were in line with Zhang et al.[21] who showed that the total fasting and postprandial GLP-1 levels were positively correlated with the HDL-C. Also, de Luis et al.[22] revealed a significant negative correlation among serum GLP-1 levels and the independent variables; waist-to-hip ratio, glucose, total cholesterol, and LDL-cholesterol. They revealed that obese patients with metabolic syndrome had lower mean GLP-1 levels than those without metabolic syndrome and they found that GLP-1 levels remained as a preventive factor to develop metabolic syndrome.

There was a postoperative highly significant decrease in systolic blood pressure, diastolic blood pressure, BMI, waist/hip ratio, FBG & 2hPP, HbA1c, fasting insulin and HOMA IR (p<0.001).These results were in agreement with those of Mor´inigo et al.[23] and Laferrère et al.[24] who reported that 6 weeks following RYGB, there was a significant decrease in the systolic blood pressure, diastolic blood pressure, BMI, waist circumference, fasting glucose, fasting insulin and HOMA-IR.

In our study, there was a postoperative increase in the fasting GLP-1 level but statistically insignificance, while there was a highly statistical significant increase in the 2hours postprandial GLP-1 levels. Our results were in agreement with those of Laferrère et al.[24] and Kashyap et al.[25] who showed that fasting GLP-1 was not altered by surgery, however, the postprandial GLP-1 response was increased (P<0.05). In addition, Umeda et al.[26] observed early changes occurring in the shape of GLP-1 postprandial curve thirty days after RYGB, and they suggested that the RYGB surgery induces early beneficial hormonal changes and is a very efficient surgical therapy for rapid glycemic control in obese patients with type 2 diabetes.

We found that 60% of patients have stopped their anti-diabetic medications, with a decrease in use of insulin plus oral therapy from 45% to only 10%, as well as a reduction in the percentage of patients who use oral anti-diabetics from 55% to 30%. These results were in agreement with Hall et al.[27] Who found that diabetes remission is achieved in 68.4% of obese subjects who underwent RYGB, a reduction in insulin ± oral anti-diabetics from 26.5% to 8.2% and reduction in oral anti-diabetics use from 73.5% to 23.5%. There was also a statistical significant decrease in the percentage of use of antihypertensive medications (from 50% to 15%) and lipid lowering medications (from 70% to 25%), (P<0.05). These findings were consistent with Mingrone et al.[28] demonstrated 75% diabetes remission, and they showed that the antihypertensive therapy was reduced in 80% of patients who underwent RYGB.

Conclusion:

RYGB surgery resulted in significant reduction in BMI and improvement in all metabolic parameters. 2hPP GLP-1 is low in diabetic patients. Postoperative 2hPP GLP-1 levels increased, possibly responsible for the metabolic benefits. Diabetes remission was noted in 60% of obese diabetic patients after surgery.

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