



## Evaluation of some hematological and biochemical parameters after bariatric surgery in overweight male dogs

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### Abstract

This study was conducted to determine the effect of bariatric surgery on some hematological and biochemical measurements in overweight male dogs and whether this type of surgery has complications. The study included 12 overweight male dogs (age 1.5-3 years) weighing 33-39 kg. The dogs were randomly divided into two groups, each containing six dogs. While the second group underwent gastric bypass surgery, the first group underwent gastric sleeve surgery. A blood sample was taken from each animal before surgery, as well as after 2, 4, 6, and 8 weeks, to evaluate some blood parameters such as (Red blood cell count, White blood cell count, Hemoglobin and Platelet count) and biochemical parameter such as (Iron, Vitamin B12, Vitamin D3, Calcium, Sodium, Potassium, and Chlorides) according to biochemical analysis. The iron, vitamin B12 and calcium levels decreased significantly in both groups, more in the second group than in the first group. However, there was no difference between the two groups in other biochemical tests, such as vitamin D3, sodium, potassium and chlorides. According to the blood test results, statistically significant differences were found in the white blood cell (WBC), red blood cell (RBC), hemoglobin (Hg), and platelets (PLT) before and after surgery but between the two groups, there were no differences. In conclusion, there was a significant decrease in the level of iron and vitamin B12, which is the cause of anemia in both groups, and there was a decrease in the level of calcium. The results showed that gastric bypass surgery caused more severe changes and more complications than gastric sleeve surgery.

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### Introduction

Excess weight is an abnormal and excess fat accumulation in different body parts (1). Obesity is now so prevalent in the world's population that it is overtaking malnutrition and infectious diseases as the leading cause of disease (2). Obesity is associated with an increased incidence of type 2 diabetes mellitus, cardiovascular disease, dyslipidemia, and hypertension, as well as an increased risk of atherosclerotic disease and mortality (3). For clinically obese patients, bariatric surgery is an increasingly accepted treatment (4). The most common and most effective way to treat morbid obesity is bariatric surgery (5). It leads to long-term weight loss, significant improvements in comorbid

conditions such as diabetes, hypertension, and obstructive sleep apnea, and reduced mortality (6). It is a form of weight loss surgery that causes weight loss through food restriction and malabsorption; in these bypass surgeries, the duodenum and a piece of the small intestine are excluded from the digestive system; a tiny gastric pouch is made as part of the RYGBP procedure. This pouch is joined to the jejunum area where food passes (7). Studies on humans have proven that such operations are usually accompanied by several blood and biochemical changes and cause serious complications (8).

The research aims to study the most essential hematological and biochemical changes that accompany these operations in dogs, on the one hand and the other hand,

to compare the two most common types of this operation, which are intestinal bypass surgery and gastric sleeve surgery, in terms of the effect on the hematological and biochemical changes.

## Materials and methods

### Ethical approval

Ethical approval was granted through the local committee on animal care and use at the College of Veterinary Medicine within the University of Mosul, number UM.VET.2023.098, dated 02/01/2023.

### Animals

Twelve overweight local breed male dogs weighing 33-39 kg and 1.5 to 3 years of age were used in this study. The dogs were randomly divided into two groups. The group consisted of six dogs in each group. The first group underwent gastric sleeve surgery, while the second group underwent gastric bypass surgery. A blood sample was taken from all dogs before surgery, as well as 2,4,6 and 8 weeks after surgery to evaluate some hematological parameters such as WBC, RBC, PLT, and Hg and biochemical parameters such as iron, vit B12, vit D3, calcium, sodium, potassium, and chlorides.

### Surgical procedures

The surgery was done under general anesthesia using (9,10). The ventral abdominal wall was prepared for aseptic surgery, and an incision of 10-15 cm was made, extending from the xyphoid cartilage to the umbilical region. Then two types of operation were done: sleeve gastrectomy was performed for each dog of the first group, while a gastric bypass operation was performed for each dog of the second group (8,11).

### Complete blood picture

Before the surgical procedure was completed, a blood sample was taken into account at time zero, as well as 2, 4, 6, and 8 weeks following the procedure. A blood sample was obtained from each animal, and a comparison was conducted between them. To avoid contaminating the sample, the collection site was prepared before the sample was collected from the jugular vein. 5ml of blood was collected, put into a glass tube with the anticoagulant ethylenediamine tetra acetic acid (EDTA), and brought to the laboratory for blood

analysis by Mythic 18 for complete blood picture measurement.

### Biochemical analysis

Every animal had a blood sample collected before the surgical operation and 2, 4, 6, and 8 weeks following the procedure. To avoid infection, the area where the blood collected was cleaned and prepared. 5ml of blood was collected from the jugular vein, put in a glass tube empty of anticoagulant, and sent to the laboratory for examination of iron level using a chemical reaction by Spin 120 (British) and vit B12 and D3 level using fully automatic immune assay by LIASON (Diasorin) Germany Fuji Film Corporation for electrolyte measurement company production in Japan.

### Statistical analysis

The data of experiments were expressed as mean±SE. The data were compared using one-way and two-way repeated measures (ANOVA). Significant differences were determined by Duncan's Multiple Range Test. Data were analyzed using Sigma Stat (Jandel scientific software V3.1), and P<0.05 was considered statistically significant (12).

## Results

### White blood cell count

The white blood cell examination results are shown in table 1, Where there was a significant increase in the number of white blood cells in the dogs of both groups, and there was no significant difference between the dogs of the two groups. This increase continued until four and six weeks after the operation when it returned to normal.

### Red blood cell count

The results of the red blood cell examination are shown in table 2, there was a significant decrease in the number of red blood cells in the dogs of both groups, and there was a considerable difference when comparing the first group with the second group, and this decrease continued until the eight weeks.

### Hemoglobin

The hemoglobin test results are shown in table 3, there was a significant decrease in hemoglobin levels in dogs of both groups after the surgery, and the decrease continued for six weeks after surgery.

Table 1: The mean of WBC measurement of the animals before and after surgical operation in two groups

Groups	WBC (10 <sup>3</sup> /μl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	6.8±0.5 a,A	18.4±1.2 a,B	14.2±0.8 a,C	11.4±0.5 a,D	8.4±0.2 a,A
Group 2	11.1±0.4 b,A	17.4±0.1 a,B	16.5±0.1 b,B	12.8±0.1 a,A	11.1±0.2 b,A

a, b the different small letters refer to significant variation between the rows at P≤0.05. A-D the different capital letters refer to a significant variation between the columns at P≤0.05.

Table 2: The mean of RBC measurement of the animals before and after surgical operation in two groups

Groups	RBC (10 <sup>3</sup> /μl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	5.2±0.08 a,A	4.2±0.1 a,B	5.1±0.1 a,A	5.1±0.1 a,A	5.2±0.1 a,A
Group 2	5.5±0.1 a,A	3.8±0.08 b,B	4.6±0.08 b,C	5.3±0.05 a,A	5.4±0.07 a,A

a, b the different small letters refer to significant variation between the rows at P≤0.05. A-C the different capital letters refer to a significant variation between the columns at P≤0.05.

Table 3: The mean of Hgb measurement of the animals before and after surgical operation in two groups

Groups	Hgb (g/dl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	13.0±0.2 a,A	9.9±0.4 a,B	9.2±0.2 a,B	12.9±0.2 a,A	13.2±0.2 a,A
Group 2	13.7±0.4 a,A	11.4±0.1 b,B	10.1±0.1 b,C	13.2±0.1 a,A	13.9±0.4 a,A

a, b the different small letters refer to significant variation between the rows at P≤0.05. A-C the different capital letters refer to a significant variation between the columns at P≤0.05.

### Platelets

The results of platelets are shown in table 4, the platelet count increased significantly after surgery, but there are no significant differences when compared between the two groups. The platelet returned to average values after six weeks.

### Iron

Table 5 shown both techniques iron levels before and after surgery. The result analysis showed a significant decrease in the average blood iron level in both groups compared to its level before the operation. While the iron level was 134.3±5.3 and 113.5±3.8 before the operation, and in the two groups respectively, its level in the blood began to decrease from the first week, and this decrease became more severe in the eighth week compared to the second week in

the first group the same result was showed in the second group. When comparing the reduction in iron levels between the two groups, no significant differences were observed in the decrease.

### Vitamin B12

The results of the vitamin B12 test for the first group showed a slight decrease in the vitamin level in the blood throughout the experiment period. However, this decrease was not significant, while the level of the vitamin declined significantly throughout the observation period in the dogs of the second group. The results also showed that there were no significant differences. The vitamin level was compared between the dogs underwent gastric sleeve surgery and the second group (Table 6).

Table 4: The mean of PLT measurement of the animals before and after surgical operation in two groups

Groups	PLT (10 <sup>3</sup> /μl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	180±2.7 a,A	329±26.6 a,B	375.5±29 a,C	261.6±23.9 a,D	235.8±16.5 a,D
Group 2	186.5±3.5 a,A	303.3±4.4 a,B	320.1±6 b,B	273.6±5.8 a,B	212.1±4.2 a,A

a, b the different small letters refer to significant variation between the rows at P≤0.05. A-D the different capital letters refer to a significant variation between the columns at P≤0.05.

Table 5: The mean of iron measurement of the animals before and after surgical operation in two groups

Groups	Iron (μg/dl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	134.3±5.3 a,A	105.8±7.2 a,B	100.3±6.6 a,B,D	95.1±7.4 a,B,D	87.6±6.3 a,C,D
Group 2	113.5±3.8 b,A	99.1±2.7 a,B	86.6±1.8 a,B,D	76.6±2.0 b,C,D	65.0±1.7 b,C

a, b the different small letters refer to a significant variation between the rows at P≤0.05. A-D the different capital letters refer to a significant variation between the columns at P≤0.05.

Table 6: The mean of B12 measurement of the animals before and after surgical operation in two groups

Groups	B12 (µg/dl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	280.6±13.4 a,A	251.3±8.6 a,B	227±2.9 a,C	214.5±1.3 a,C,D	204.8±1.6 a,D
Group 2	297.5±2.5 a,A	253.3±4.9 b,B	216.6±2.4 a,C	196.6±3.3 a,D	183.3±3.8 b,D

a, b the different small letters refer to significant variation between the rows at  $P \leq 0.05$ . A-D the different capital letters refer to a significant variation between the columns at  $P \leq 0.05$ .

### Vitamin D3

The results showed a significant decrease in the vitamin D3 level for both groups over the length of the experiment compared to the level before the operation. However, the two groups had no significant differences in the vitamin D3 levels. The results showed a substantial decrease in the vitamin D3 level for both groups over the length of the experiment compared to the level before the operation. However, the two groups had no significant differences in the vitamin D3 levels (Table 7).

### Calcium, Sodium, Potassium, and Chlorides

Tables 8-11, respectively show the levels of calcium, sodium, potassium and chlorides for dogs in the first and second groups. There was a fluctuation in the levels of these ions, and these changes were insignificant when comparing the first group that had gastric sleeve surgery with the second group that had gastric bypass surgery. There was no consistent change in the level when comparing the levels of these ions with their levels in dogs before the surgical operation.

Table 7: The mean of D3 measurement of the animals before and after surgical operation in two groups

Groups	D3 (µg/dl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	40±1.5 a,A	34.5±1.5 a,B	30.3±1.5 a,C	27±1.3 a,D	24±1.0 a,D
Group 2	37.6±0.7 a,A	33.3±0.8 a,B	29±1.0 a,C	25.8±0.6 a,C,D	22.6±0.6 a,D

a the similar small letters refer to no significant variation between the rows at  $P \leq 0.05$ . A-D the different capital letters refer to a significant variation between the columns at  $P \leq 0.05$ .

Table 8: The mean of calcium measurement of the animals before and after surgical operation in two groups

Groups	Calcium (mg/dl)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	9.6±0.09 a,A	9.3±0.06 a,B	9.3±0.06 a,B	9±0.05 a,C	8.8±0.08 a,C
Group 2	9.4±0.04 b,A	9±0.04 b,B	8.8±0.04 b,C	8.5±0.05 b,D	8.4±0.06 b,D

a, b the different small letters refer to significant variation between the rows at  $P \leq 0.05$ . A-D the different capital letters refer to a significant variation between the columns at  $P \leq 0.05$ .

Table 9: The mean of sodium measurement of the animals before and after surgical operation in two groups

Groups	Sodium (mmol/L)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	148.5±0.2 a,A	146.6±0.3 a,B	145.6±0.5 a,B	143.8±0.4 a,C	143.3±0.6 a,C
Group 2	148.8±0.3 a,A	148±0.4 b,B	147.5±0.4 b,B	146.6±0.3 b,B,C	146.5±0.5 b,B,C

a, b the different small letters refer to significant variation between the rows at  $P \leq 0.05$ . A-C the different capital letters refer to a significant variation between the columns at  $P \leq 0.05$ .

Table 10: The mean of potassium measurement of the animals before and after surgical operation in two groups

Groups	Potassium (mmol/L)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	4.9±0.01 a,A	4.8±0.04 a,A,B	4.7±0.05 a,B,C	4.5±0.08 a,C	4.5±0.08 a,C
Group 2	4.9±0.04 a,A	4.8±0.04 a,A	4.7±0.06 a,A	4.6±0.06 a,A	4.4±0.2 a,B

a the similar small letters refer to no significant variation between the rows at  $P \leq 0.05$ . A-C the different capital letters refer to a significant variation between the columns at  $P \leq 0.05$ .

Table 11: The mean of chlorides measurement of the animals before and after surgical operation in two groups

Groups	Chlorides (mmol/L)				
	Before surgery	2 weeks	4 weeks	6 weeks	8 weeks
Group 1	106.8±0.7 a,A	105±0.8 a,B	103.5±0.7 a,C,B	102.1±0.4 a,D,C	101.3±0.3 a,D
Group 2	107.1±0.6 a,A	106.5±0.7 a,A	105.6±0.6 b,A,B	104.5±0.3 b,B	104.3±0.4 b,B

a, b the different small letters refer to significant variation between the rows at  $P \leq 0.05$ . A-c the different capital letters refer to a significant variation between the columns at  $P \leq 0.05$ .

## Discussion

The most popular and highly efficient method for treating severe obesity is gastric bypass surgery (5). This is a form of weight reduction surgery that causes weight loss through both food restriction and malabsorption. In these bypass surgeries, the duodenum and a piece of the small intestine are excluded from the digestive system. A tiny gastric pouch is made as part of the RYGBP procedure. This pouch is joined to the jejunum area where food passes (7).

The results showing two weeks following the procedure, the WBC counts are excessively high 18.4 in group one and 17.4 in group two. Although elevated white blood cell counts don't necessarily signify a particular illness, they can indicate a problem, such as an infection, stress, inflammation, or trauma, given that the animals have just undergone surgery, where an elevated level of WBCs was expected. Fortunately, the white blood cell count returned to normal once these problems were resolved. For this reason, four, six-, and eight-weeks following surgery, there was a minor decrease in the white blood cell count, which indicates that the WBC count was returning to normal. This agrees with Estopa (13), Bilen (14) and Rubio *et al.* (15).

Before surgery, the animals' RBC count was within normal limits in group one and 5.5 in group two, but it fell below the normal level two weeks after the procedure, then the RBC return to normal level after four, six, eight weeks post-operatively this agrees with Estopa (13), Bilen (14) and Rubio *et al.* (15). The surgical procedure's unavoidable bleeding or blood loss caused the RBC count to decrease. This agrees with Estopa (13).

Before surgery, the animals' Hg was within normal limits 13 in group one and 13.7 in group two, but it fell below the normal level two, four weeks after the procedure, then the Hg return to normal level after six, eight weeks post-operatively this agrees with Estopa (13), Bilen (14) and Rubio *et al.* (15). The surgical procedure's unavoidable bleeding or blood loss caused the RBC count to decrease, which led to a decrease in Hg level. This agrees with Estopa (13).

before to surgery, the animals' PLT was within normal limits 180 in group one and 186 in group two. However, it increased after two to four weeks post-operatively but is still within the normal range, which agrees with Estopa (13), Bilen (14) and Rubio *et al.* (15). The animals had a large cut, and the operation was very invasive, so a high or elevated

platelet count was expected, which agrees with Estopa (13) and Rubio *et al.* (14).

The iron results showed an apparent decrease in the iron level in dogs that underwent gastric sleeve surgery, the same result was shown in dogs of group two of the Roux-en-Y gastric bypass. The percentage of iron begins to decrease gradually after the surgical operation for both groups. The decreases were more severe in the second group, the same result confirmed by Kwon *et al.* (16). Changes in the gut anatomical structure affect iron digestion and absorption, in addition, decreased hydrochloric acid secretion impedes the conversion of ferric iron to absorbable ferrous iron this agrees with Steenackers *et al.* (17), Roux-en-Y gastric bypass surgery was more invasive than the sleeve gastrectomy, which further contributes to the anemia and decreases iron levels as adverse effect, Salgado *et al.* findings confirm this theory (18). Also, the Roux-en-Y gastric bypass of the principal site of absorption in the duodenum and proximal jejunum may lead to the development of iron deficiency and anemia after surgery. This agrees with Ghanbari *et al.* (8). The results of the B12 showed that there was an apparent decrease in the level of vitamin B12 in the dogs of both groups after the operation, and the decrease was more pronounced in the second group of dogs that had a gastric bypass operation. This result was inconsistent with what was reported by Ghanbari *et al.* (8), as they proved that there was no difference in the level of vitamin B12 in the dogs of both groups after the operation between the two methods. The cause of vitamin B12 deficiency is due to the loss of the intrinsic factor that produces parietal cell mass. This loss of intrinsic factors is responsible for vitamin B12, and this consistent with Majumder *et al.* (19). In addition, Roux-en-Y gastric bypass surgery means the loss of the organ designated for absorbing vitamin B12, and this is consistent with what he mentioned with Majumder *et al.* (19). There is no difference in vitamin D3 deficiency between the two groups, and this was confirmed by Moore and Sherman (20). However, there was a decrease in D3 over the course of the experiment in each group because the absorption of vitamin D3 in the jejunum and ileum was confirmed by Tucker *et al.* and Bloomberg *et al.* (21,22). This area of absorption will be affected after bariatric surgery. This is the main cause of the decrease in vitamin D3 levels. Also, vitamin D3 malabsorption occurs due to bypassing certain parts of the gut during bariatric surgery. This agrees with Bloomberg *et al.* (22). Vitamin D

deficiency has been linked to subsequent hypocalcemia following bariatric surgery. This occurred during this experiment, supported by Becker *et al.* (23). There was a decrease in calcium levels in both groups, which agreed with Compher *et al* and Gemmel *et al.* (24,25). Calcium malabsorption occurs due to bypassing certain parts of the gut during bariatric surgery. This concurs with Bloomberg *et al.* (22). The decrease in calcium levels was more significant in the bypass group that changed course, and this agrees with Bernert *et al.* (26). that said calcium deficits are explained by a dietary intake and reduction in the absorption which usually takes place in the duodenum and the proximal jejunum this agree with Goode *et al.* (27). It is generally known that people undergoing bariatric surgery can suffer from calcium and vitamin D deficiency. Calcium is absorbed preferentially in the duodenum and proximal jejunum, whereas vitamin D is absorbed primarily in the jejunum and ileum (22). Some complications can happen following bariatric surgery, the most serious of which are electrolytes (sodium, potassium, chloride) and nutritional deficiencies. Hyponatremia is characterized by an incorrect antidiuretic hormone syndrome, elevated vasopressin levels, and adrenal insufficiency, as well as edema, diuretic usage, vomiting, and diarrhea (28).

## Conclusion

level of iron, vitamin B12 and calcium effect during gastric bypass surgery more than sleeve gastrectomy. Hematological parameters show changes in both groups, but other parameters show the same changes in both procedures.

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## Conflict of interests

The authors declare that there is no conflict of interest regarding the publication of this paper.

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## تقييم بعض المعايير الدموية والكيموحيوية بعد جراحة السمنة لدى ذكور الكلاب ذات الوزن الزائد

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### الخلاصة

أجريت هذه الدراسة لمعرفة تأثير جراحة السمنة على بعض القياسات الدموية والكيموحيوية لدى ذكور الكلاب ذات الوزن الزائد وما إذا كان لهذا النوع من الجراحة مضاعفات. شملت الدراسة ١٢ كلباً ذكراً زائد الوزن (عمرهم ١,٥-٣ سنوات) وتراوح أوزانهم بين ٣٣-٣٩ كيلوغرام. تم تقسيم الكلاب بشكل عشوائي إلى مجموعتين، تحتوي كل مجموعة على ستة كلاب. بينما خضعت المجموعة الثانية لعملية تحويل مسار المعدة، خضعت المجموعة الأولى لعملية تكميم المعدة. تم أخذ عينة دم من كل حيوان قبل الجراحة، وكذلك بعد ٢، ٤، ٦، ٨ أسابيع، لتقييم بعض مؤشرات الدم مثل (عدد خلايا الدم الحمراء، عدد خلايا الدم البيضاء، الهيموجلوبين وعدد الصفائح الدموية) ومؤشرات الكيموحيوية مثل (الحديد، فيتامين ب١٢، فيتامين د٣، الكالسيوم، الصوديوم، البوتاسيوم والكلوريد) وذلك حسب التحليل الكيموحيوي. انخفضت مستويات الحديد وفيتامين ب١٢ والكالسيوم بشكل ملحوظ في كلا المجموعتين، بشكل أكبر في المجموعة الثانية مقارنة بالمجموعة الأولى. ومع ذلك، لم يكن هناك فرق بين المجموعتين في الاختبارات الكيموحيوية الأخرى، مثل فيتامين د٣ والصوديوم والبوتاسيوم والكلوريد. وفقاً لنتائج فحص الدم، تم العثور على فروق ذات دلالة إحصائية في خلايا الدم البيضاء، وخلايا الدم الحمراء، والهيموجلوبين والصفائح الدموية قبل وبعد الجراحة، ولكن بين المجموعتين، لم تكن هناك اختلافات. وفي الختام، كان هناك انخفاض كبير في مستوى الحديد وفيتامين ب١٢، وهو سبب فقر الدم في كلا المجموعتين، كما حدث انخفاض في مستوى الكالسيوم. وأظهرت النتائج أن جراحة تحويل مسار المعدة تسببت في تغييرات أكثر حدة ومضاعفات أكثر من جراحة تكميم المعدة.