



BioBacta



Journal of Bioscience and Applied Research

www.jbaar.org



## Nephrotoxicity associated with Orlistat in normal and obese female rats

Ehab Tousson, Ahmed Massoud, Attyat Salem, Shahenda A. Fatoh

Department of Zoology, Faculty of Science, Tanta University, Tanta, Egypt.

Email: [shahenda66anter@yahoo.com](mailto:shahenda66anter@yahoo.com)

DOI: [10.21608/jbaar.2018.148655](https://doi.org/10.21608/jbaar.2018.148655)

### Abstract

Obesity is a global health concern associated with high morbidity and mortality. Therapeutic strategies include synthetic drugs and surgery, which may entail high costs and serious complications. Orlistat is a pancreatic lipase inhibitor licensed for the treatment of obesity. The current study was carried out to elucidate the modulating effect of Orlistat against obesity-induced kidney toxicity in female rats. A total of 50 female rats were divided into five groups (G1, Control; G2, Orlistat; G3, Obesity; G4, Co- treated Orlistat with Obesity; G5, Post- treated Obesity with Orlistat rat group). The current study revealed that a significant increase in serum urea, creatinine, while a significant decrease in the levels of sodium, potassium, calcium, and chloride ions levels in treated rats with Orlistat while a significant increase in serum urea, creatinine, sodium, potassium, and chloride ions levels in obesity group when compared with a control group. In contrast; a significant decrease in serum urea, Creatinine, sodium, potassium, and chloride ions levels in treated obese rats with Orlistat when compared with the obesity group. So; Orlistat-induced renal toxicity when used for the treatment of obesity and self-recovered obese rats is safe and better than the use of Orlistat in the treatment of obesity.

**Keywords:** Obesity, Orlistat, kidney functions, Electrolytes, Toxicity, Rat.

### 1 Introduction

Obesity is a chronic metabolic disease that is characterized by excess body fat which is generally accompanied by an increase in body weight and this happens when the body mass index (BMI) in an adult is greater than 30 units (Huang et al., 2015; Sánchez et al., 2018). Obesity has become a major health problem all over the world and is associated with substantial increases in morbidity, premature

mortality, impaired quality of life, and an increased risk of complications including insulin resistance, hypertension, dyslipidemia, diabetes mellitus, atherosclerosis, coronary heart disease, and certain cancers (Drew *et al.*, 2007; Wang et al., 2015; Campbell et al., 2016; Defo et al., 2017).

Anti-obesity medication or weight loss drugs are pharmacological agents that reduce or control weight by altering either appetite or absorption of calories.

Some anti-obesity drugs can have severe, even lethal side effects and these side effects are often associated with the medication's mechanism of action (Huang et al., 2015; Defo et al., 2017). Orlistat is an anti-obesity medication that is an inhibitor of gastric and pancreatic lipases that acts in the gastrointestinal tract (Amin et al., 2014). Orlistat when administered with fat-containing foods, it partially inhibits the hydrolysis of triglycerides, thus reducing the subsequent absorption of monoglycerides and free fatty acids, leading to a reduced-fat absorption by around 30% (Choussein et al., 2009). The current study was carried out to elucidate the modulation effect of orlistat against obesity-induced kidney toxicity in female rats.

## 2. Materials and Methods

### Orlistat Supplementation

Orlistat drugs were obtained from the pharmacy in Cairo, Egypt. Orlistat drug was marketed as a prescription under the trade name Xenical by Roche in most countries and is also known as tetrahydrolipstatin.

### Experimental animals

The experiment was performed on 50 male albino rats (*Rattus norvegicus*) weighing 120 g ( $\pm 10$ ) and of 10-12 weeks age. They were acquired from laboratory farms, Zoology Department, Faculty of Science, Tanta University, Egypt.

The rats were held in suitable plastic cages for one week before the experimental work for acclimation with a new room condition and maintained on a standard rodent diet and water available ad libitum. The temperature in the animal room was adjusted to  $23 \pm 2^\circ\text{C}$  with a relative humidity of  $55 \pm 5\%$ . The light was on a 12 h light/dark cycle. Animal procedures were performed following the Ethics Committee of the National Research Centre, Egypt.

### Experimental groups

A total of 50 rats were equally divided into five groups (10 rats each).

Group 1, Control group included rats received no treatment;

Group 2, Orlistat group included rats received (12 mg/kg) of orlistat daily dissolved in saline (1ml/kg) by intraperitoneal injection for four weeks (Amin et al., 2015).

Group 3, an obese group in which rats were fed on a high fat diet for six weeks. Also, saline solution was administered intraperitoneally to the rats of this group (1ml/kg).

Group 4, the post-treated group included obese rats that were treated with orlistat for four weeks.

Group 5, the Self-treated group included obese rats that left for another four weeks without receiving any treatment.

At the end of the experimental period, animals have fasted overnight and for clinical chemistry blood samples from each rat were collected from the eyes by retro-orbital puncture using blood capillary tubes without heparin as per requirement under mild ether anesthesia.

Blood samples were incubated at room temperature for 10 minutes and left to clot then centrifuged at 3000 r.p.m for 10 min and the serum was collected, serum was separated and kept in a clean stopper plastic vial at  $-80^\circ\text{C}$  until the analysis.

### Kidney functions and electrolytes in serum

Sera were used for the determination of urea and creatinine according to the methods described by Patton and Crouch (1977) and Henry et al. (1974), respectively. Sodium and potassium ions concentration were determined using the method of Henry et al. (1974).

### Statistical Analysis

Data were expressed as mean values  $\pm$  SE and statistical analysis was performed using one-way ANOVA to assess significant differences among treatment groups. The criterion for statistical significance was set at  $p < 0.05$  for the biochemical data.

## 3. Results

### Toxicity

Many side effects appeared on rats after Orlistat administration as loss of body weight, loss and changes in the colour of hairs, and bloody eyes, and many tumours appearing in a different locations in animals.

### Changes in relative body and kidney weights

Rat relative body and kidney weight levels in the obesity group were significantly increased when compared with the control. In contrast; a significant decrease in relative body weight level in treated rats with Orlistat when compared with control and obesity groups (Figures 1&2). On the other hand; a significant decrease in relative body weight level in treated obese rats with Orlistat when compared with obesity groups. In contrast; insignificant decreases were detected in relative kidney weight level in treated obese rats with Orlistat or self-treated obese rats when compared with obesity groups (Figures 1&2).

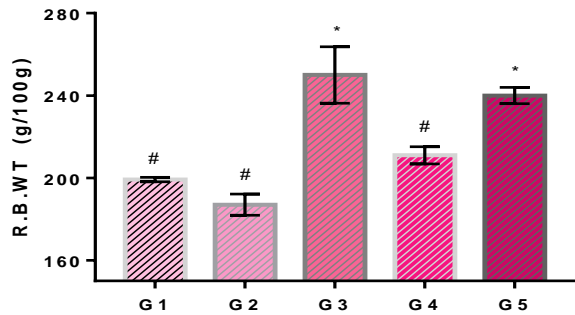


Figure 1: Changes in relative body weight levels in different groups. Values are expressed as means  $\pm$  SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. #Significantly different from obesity, \*Significantly different from control.

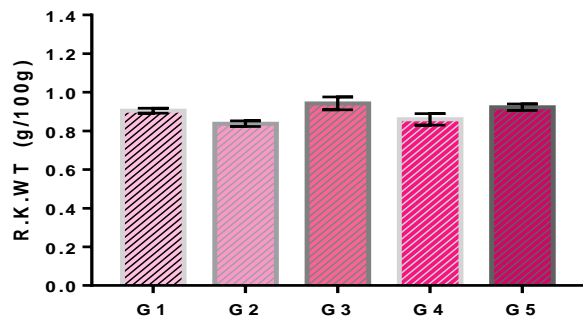


Figure 2: Changes in relative kidney weight levels in different groups. Values are expressed as means  $\pm$  SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. #Significantly different from obesity, \*Significantly different from control.

**Orlistat induces renal toxicity:**

Figures (3&4) showed a significant increase in serum creatinine and urea levels in treated rats with Orlistat when compared with control, also a significant increase in serum creatinine and urea levels in obese rats when compared with control. On the other hand; treatment of obese rats with Orlistat revealed significant changes in creatinine and urea levels when compared with the obese rat's group. Self-treated obese rats revealed an insignificant decrease in creatinine and urea levels when compared with obese or treated obese rats with Orlistat groups (Figures 3&4).

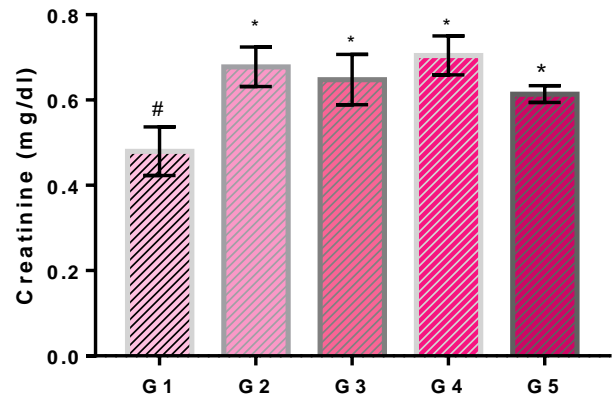


Figure 3: Changes in serum creatinine levels in different groups. Values are expressed as means  $\pm$  SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. # Significantly different from obesity, \*Significantly different from control.

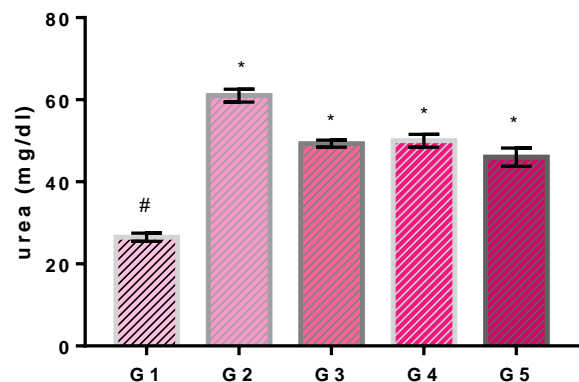
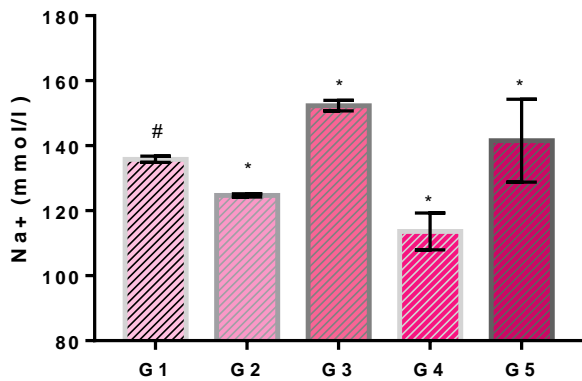


Figure 4: Changes in serum urea levels in different groups. Values are expressed as means  $\pm$  SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. # Significantly different from obesity, \*Significantly different from control.

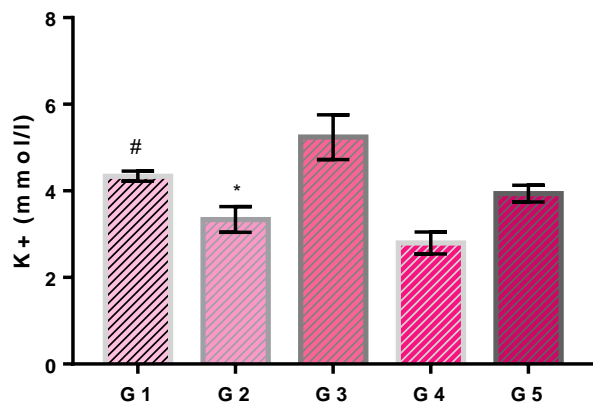
**Changes in electrolytes:**

Figures (5-8) showed the changes in electrolytes in different groups. a significant decrease in serum sodium, potassium, calcium, and chloride ions levels in treated rats with Orlistat when compared with control, in contrast, a significant increase in serum sodium, potassium, and chloride ions levels in obese rats when compared with control. On the other hand; calcium ions levels revealed significant changes between obese and control rats (Figures 5-8). A significant decrease in serum sodium, potassium,

calcium, and chloride ions in treated obese rats with Orlistat or in self-treated obese rats when compared with the obese rat's group (Figures 5-8).



**Figure 5:** Changes in serum sodium ions (Na<sup>+</sup>) levels in different groups. Values are expressed as means ± SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. #Significantly different from obesity, \*Significantly different from control.

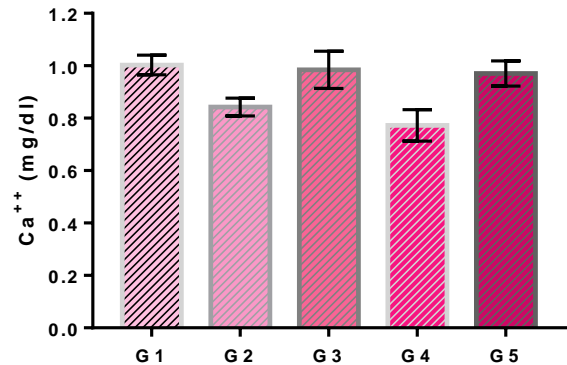


**Figure 6:** Changes in serum potassium ions (K<sup>+</sup>) levels in different groups. Values are expressed as means ± SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. # Significantly different from obesity, \*Significantly different from control.

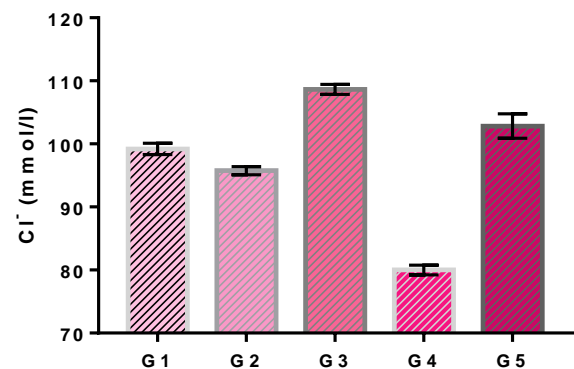
#### 4. DISCUSSION

Obesity is a global health concern associated with high morbidity and mortality. Obesity also increases the risk of diabetes, cardiovascular disorder, stroke, and colon cancer. The combination of these chronic diseases is called metabolic syndrome (Grundy 2004). However, renal risks induced by obesity, especially its role in the initiation and progression of

renal diseases, have only been recently recognized (Abrass 2004, Tang *et al.* 2012). Orlistat is a gastrointestinal lipase inhibitor used as an adjunct treatment of obesity. The current study was carried out to elucidate the modulation effect of orlistat against obesity-induced kidney toxicity, injury, and P53 immunohistochemical alterations in female rats.



**Figure 7:** Changes in serum calcium ions (Ca<sup>++</sup>) levels in different groups. Values are expressed as means ± SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. # Significantly different from obesity, \*Significantly different from control.



**Figure 8:** Changes in serum chloride ions (Cl<sup>-</sup>) levels in different groups. Values are expressed as means ± SE; n = 10 for each treatment group. G1, Control; G2, Orlistat; G3, obesity; G4, post-treated obesity with Orlistat; G5, Self-treated obesity. # Significantly different from obesity, \*Significantly different from control.

In the current study; a significant increase in serum creatinine and urea levels in obese rats when compared with control. Also, significant increases in

serum creatinine and urea levels in treated rats with Orlistat when compared with control rats indicating Orlistat induces renal toxicity.

On the other hand; treatment of obese rats with Orlistat revealed significant changes in creatinine and urea levels when compared with the obese rat's group. Self-treated obese rats revealed an insignificant decrease in creatinine and urea levels when compared with obese or treated obese rats with Orlistat groups. Courtney *et al.* (2007); Singh *et al.* (2007) and Weir *et al.* (2011) reported an association between orlistat and acute kidney injury. In 2007, Singh and colleagues provided the initial suggestion of orlistat-induced AKI [Singh *et al.* 2007]. In a patient with underlying stage III chronic kidney disease (CKD; among other comorbidities), initiation of orlistat treatment coincided with an increase in serum creatinine concentration, an increase in urine oxalate concentration, and the presence of calcium oxalate crystals in the lumen of the renal tubules. These findings resolved 1 month after discontinuation of orlistat.

Courtney and colleagues reported similar findings in a 55-year old woman with diabetic nephropathy (Courtney *et al.* 2007). After 5 months of orlistat therapy, the patient's estimated glomerular filtration rate (eGFR) declined from a baseline of 66 ml/min/1.73 m<sup>2</sup> to 12 ml/min/1.73 m<sup>2</sup> with a documented accumulation of calcium oxalate crystals in the renal parenchyma. In 2007, McLaughlin and Macdougall reported a case series of 33 obese patients with CKD (stages III or IV) taking orlistat for more than 6 months (MacLaughlin and Macdougall, 2007).

Weir *et al.* (2011) reported that; orlistat induced acute kidney injury in humans. In obese people, increased serum creatinine was observed, suggesting that obesity caused elevation in renal function test and produced proteinuria, concomitantly with other risk factors such as hypertension, diabetes, and dyslipidemia (Matsushita *et al.*, 2009). Renal triglyceride accumulated in obese rats accompanied by hypoalbuminemia and elevated blood urea nitrogen.

In the current study; a significant decrease in serum sodium, potassium, calcium, and chloride ions levels in treated rats with Orlistat when compared with control, in contrast, a significant increase in serum sodium, potassium, and chloride ions levels in obese rats when compared with control. On the other hand; a significant decrease in serum sodium, potassium, calcium, and chloride ions in treated obese rats with Orlistat or in self-treated obese rats when compared with the obese rats group. Weisinger *et al.* (1974) reported that massive obese patients developed

nephrotic range proteinuria. Obesity that is related to glomerulopathy is considered an increasing cause of end-stage renal disease. Potential mechanisms by which obesity affects renal physiology include altered renal hemodynamics, insulin resistance, hyperlipidemia, activation of the renin-angiotensin-aldosterone system, inflammation, and oxidative stress. Also, increases in both glomerular filtration rate and renal plasma flow are observed in both obese subjects and animals (Chagnac *et al.*, 2000).

## 6. References

- Abrass, CK (2004): Overview: Obesity: what does it have to do with kidney disease? *J Am Soc Nephrol.*, 15: 2768-2772.
- Amin KA, Galaly SR, Hozayen WG, Ramadan SM. (2014): Effects of orlistat and herbal mixture extract on renal function and oxidative stress. Biomarkers in a Rat Model of High Fat Diet. *International Journal of Biochemistry Research & Review* 4(2): 173-192.
- CAMPBELL KJ, HESKETH KD, MCNAUGHTON SA, BALL K, MCCALLUM Z, LYNCH J, CRAWFORD DA. (2016): THE EXTENDED INFANT FEEDING, ACTIVITY AND NUTRITION TRIAL (INFANT EXTEND) PROGRAM: A CLUSTER-RANDOMIZED CONTROLLED TRIAL OF AN EARLY INTERVENTION TO PREVENT CHILDHOOD OBESITY. *BMC*, 16: 166. DOI: 10.1186/s12889-016-2836-0
- Chagnac A, Weinstein T, Korzets A, Ramadan E, Hirsch J, Gafer U. (2000): Glomerular hemodynamics in severe obesity. *American Journal of Renal Physiology*, 278(5): 817-822.
- CHOUSSEIN S, MAKRI A, DASKALOPOULOU S. (2009): EFFECT OF ANTI-OBESITY MEDICATIONS IN PATIENTS WITH TYPE 2 DIABETES MELLITUS. *DIABETES, OBESITY AND METABOLISM* DOI:10.1111/J.1463-1326.2008.01026.X
- Courtney AE, O'Rourke DM, Maxwell AP. (2007): Rapidly progressive renal failure associated with successful pharmacotherapy for obesity. *Nephrol. Dialysis Transplant* 22: 621-623
- Defo PB, Wankeu-Nya M, Ngadjui E, Bonsou Fozin GR, Kemka FX, *et al.* (2017): Palm Oil Diet-Induced Obesity Impairs Male Rat Reproductive Performance. *Ann. Reprod Med. Treat.*, 2(2): 1012.
- Drew MD, Borgeson TL, Thiessen DL. (2007): A review of processing of feed ingredients to

- enhance diet digestibility in finfish. *Anim. Feed Sci. Technol.*, 138 (2): 118-136
- GRUNDY SM. (2004): OBESITY, METABOLIC SYNDROME, AND CARDIOVASCULAR DISEASE. *THE JOURNAL OF CLINICAL ENDOCRINOLOGY & METABOLISM*, **89(6)**: 2595–2600.
- Henry RJ. (1974): *Creatinine Measurements with Colorimetric Method: Clinical Chemical Principles and Techniques*. 2nd ed. New York: Harper and Row Publishers, p. 525.
- Huang CJ, McAllister MJ, Slusher AL, Webb AE, Mock JT, Acevedo E. (2015): Obesity-related oxidative stress: the impact of physical activity and diet manipulation. *Sports medicine-open* 1 (1): 32.
- MacLaughlin H., Macdougall I.C. (2007): Rapidly progressive renal failure associated with successful pharmacotherapy for obesity. *Nephrol Dialysis Transplant* 22: 2403–2405.
- Matsushita K, Yasuda G, Shouda M, Umemura S. (2009): Evaluation of renal function and protein urea body mass health examination in young obese Japanese people. *Clinical and Experimental Nephrology*. 13(4):316-324.
- Patton C, Crouch SR (1977): Spectrophotometric and kinetics investigation of the Berthelot reaction for the determination of ammonia. *Analytical Chemistry* 49: 464–468.
- SANCHEZ J, MORALES H, ALVAREZ P, FRUSCH J, ET AL. (2018): THE OBESITY IMPACT ON FERTILITY. *J. DIABETES METAB. DISORD. CONTROL*, 5(3):103–105.
- Singh A, Sarkar SR, Gaber LW, Perazella MA. (2007): Acute oxalate nephropathy associated with orlistat, a gastrointestinal lipase inhibitor. *Am. J. Kidney Dis.*, 49: 153–157.
- Tang J, Yan H, Zhuang S. (2012): Inflammation and oxidative stress in obesity-related glomerulopathy. *Int. J. Nephrol.*, 2012: 1-11.
- Wang ZJ, Zhou YJ, Galper BZ, Gao F, Yeh RW, Mauri L. (2015): Association of body mass index with mortality and cardiovascular events for patients with coronary artery disease: a systematic review and meta-analysis. *101(20):1631-8*. DOI: 10.1136/heartjnl-2014-307119.
- Weir MA, Beyea MM, Gomes T, Juurlink DN, Mamdani M, Blake PG, et al. (2011): Orlistat and acute kidney injury: an analysis of 953 patients. *Arch. Intern. Med.*, 171: 703–704.
- Weisinger JR, Kempson RL, Eldridge FL, Swenson RS. (1974): The nephrotic syndrome: a complication of massive obesity. *Annals of Internal Medicine*, 1974;81(4):440–447, doi: 10.7326/0003-4819-81-4-440.