Egyptian Journal of Zoology (EJZ)
Publisher: The Zoological Society of A. R. Egypt
DOI: 10.21608/ejz.2025.401440.1133

p-ISSN: 1110-6344
e-ISSN: 2682-3160

RESEARCH ARTICLE

# COMBINED EFFECT OF CURCUMIN AND PIPERINE ON OBESITY-INDUCED HEMATOPOIETIC, HEPATIC, AND RENAL DYSFUNCTION IN MALE WISTAR RATS

**Hanan Rashwan[1]; Nahed Mansour[1]; Eman O. Mohamed[1*]; Aida A. Hussein[2]**

[1]Zoology Department, Faculty of Science, Arish University, North Sinai, Egypt
[2]Zoology Department, Faculty of Science, Suez University, Suez, Egypt

**Article History:**
Received: 7 July 2025
Accepted: 31 August 2025

Published Online:
5 September 2025

**Keywords:**
Curcumin
Kidney toxicity
Liver toxicity
Obesity
Piperine

***Correspondence:**
Eman Mohamed
Zoology Department
Faculty of Science
Arish University
North Sinai, Egypt
E-mail:
eman.osama@sci.aru.edu.eg

## ABSTRACT

One of the adverse effects of obesity is hematopoietic toxicity and vital organs injury. This research sets out to determine whether or not obese rats might avoid damage to their erythrocytes, liver, and kidneys by taking curcumin "CUR" (100 mg/kg of body weight of the rats) and piperine "PIP" (5 mg/kg of body weight of the rats), *via* stomach tube, daily, for eight weeks. Alanine and aspartate aminotransferases, alkaline phosphatase, and γ-glutamyl transferase activities, platelet and white blood cell counts, as well as total bilirubin and creatinine level, in the blood rose sharply ($P \leq 0.05$) after high-fat diet consumption, but albumin level fell sharply ($P \leq 0.05$). Obese rat also showed macrocytic normochromic anemia. Obese rats' livers and kidneys showed several abnormalities upon histological analysis. Livers of the high-fat diet group showed dilated central veins with hemorrhage and dilated vascular sinuses, enlarged hepatocytes with numerous large-vessel fatty livers, activated Kupffer cells, and aggregates of inflammatory cells. Renal cortical sections from the high-fat diet group showed dilated glomeruli, congestion of intertubular vessels, and edematous degeneration of some lining epithelial cells. Supplementation with CUR and PIP alleviated significantly ($P \leq 0.05$) obesity-related hematopoietic toxicity, as well as liver and kidney damage. Therefore, these results may recommend daily intake of CUR and PIP to prevent obesity-related health risks and liver and kidney toxicity.

## INTRODUCTION

Worldwide, obesity ranks as the fifth leading cause of death, posing a serious public health issue[1]. It is directly associated with chronic diseases such as diabetes, cardiovascular disorders, and chronic kidney disease[2-4]. According to the WHO, nearly 650 million individuals are obese out of 2 billion overweight people[3]. Obesity results from an imbalance between calorie intake and expenditure, influenced by genetic, behavioral, and metabolic factors[4]. This metabolic imbalance leads to systemic inflammation and oxidative stress, damaging several vital organs[5]. Obesity is a major risk factor for metabolic syndrome, type 2 diabetes mellitus, and nonalcoholic fatty

liver disease (NAFLD)[6-8]. The liver is highly affected in obesity, as fat accumulation impairs its metabolic functions, leading to inflammation and NAFLD[9].

Curcumin (CUR) is a herbal powder of the *Curcuma longa* (turmeric) rhizome. It has been widely used as a spice and a coloring agent. It is a major ingredient of curry powder. CUR along with its compounds (curcuminoids) are yellowish pigments of turmeric and it's known for its influence as an antioxidant and anti-inflammatory component[10]. In addition, CUR plays a vital role in combating obesity by preventing the accumulation of fats in the body resulting from the consumption of high- fat diet (HFD), and by contributing to the reduction of fatty liver[11].

Piperine (PIP) is the main component of the most used spice "black pepper (*Piper nigrum*)"[12]. PIP is known for its pungency taste and recent studies have shown that it significantly enhances metabolism, absorption, and the bioavailability of other substances such as curcumin, increasing its effect by 2000% in human and 154% in rat's models. PIP also plays a role in weight loss in a healthy way by preventing or reducing hyperlipidemia caused by HFDs[12].

Eventually CUR gained attention for their role in treating obesity with minimal side effects. However, its poor oral bioavailability limits its therapeutic use[13]. Co-administration with PIP significantly enhances CUR bioavailability and efficacy[14,15]. Therefore, the aim of this study was to evaluate the efficacy of CUR-PIP combination treatment against obesity-induced hematological, hepatic, and renal disorders in male Wistar rats using biochemical and histological approaches.

## MATERIAL AND METHODS

### Natural products and chemicals

CUR and PIP supplementation capsules (catalogue number: 078826) were purchased from Puritan's Pride (Cairo, Egypt). Dimethyl sulfoxide (DMSO) was purchased from El-Gomhoria Company for Chemicals, Mansoura, Egypt.

### Animals and research subjects

In this study, male Wistar albino rats "*Rattus norvegicus*" (180–200 g) were used. All these rats came straight from Egyptian Institute of Serology and Vaccine Production (Helwan, Cairo, Egypt). The rats were kept in stainless steel cages in a well-ventilated animal facility with typical climatic conditions, including a room temperature of 23±2°C, a 12-hour light/dark cycle, and a humidity level of 40±5% with free access to food and water. The animals were allowed to acclimate to the new environment for one week and then randomly divided into five groups (six rats per group). Group I was the control group, where rats were fed a standard diet (8% fat, 75% carbohydrate, 17% protein). Group II was administered 5% DMSO, as a vehicle, through a stomach tube at a dose of 0.1 mL/100 g body weight[15]. Group III was orally administered CUR+PIP as reported by Antona *et al*.[16]: CUR (100 mg/kg of body weight) and PIP (5 mg/kg of body weight) dissolved in 5% DMSO, through a stomach tube. In group IV, participants were given a high-fat diet (HFD) consisting of 60% fat, 23% carbohydrates, and 17% protein[17]. In group V, participants were given the same HFD along with the same amounts of CUR+PIP. All animals received the corresponding treatments daily for 8 weeks.

### Ethical approval

All experimental protocols were approved by the Ethics Committee for Animal Research of the Faculty of Science, Arish University (approval number: ARU010). All methods were carried out in accordance with relevant guidelines and regulations. All methods are reported in accordance with ARRIVE guidelines (https://arriveguidelines.org).

### Samples collection

The rats were anesthetized at the conclusion of the eight-week study period according to Wellington *et al*.[18]. The used dosage of anesthesia (ketamine/xylazine) was 0.1 ml per 100 g of body weight. The procedure

consisted of drawing two blood samples from each rat: one without anticoagulant and one with sodium citrate. In order to prepare the serum for biochemical analysis, blood without anticoagulant was left to coagulate at room temperature, then was spun at a speed of 850 ×*g* for a duration of 15 minutes. After that, the separated serum was stored in Eppendorf tubes with labels at a temperature of –20°C. Following that, the rats were dissected, and their internal organs (kidney and liver) were separated before being washed with saline solution. In order to conduct histological study, sections of liver and kidney, were embedded in a neutral formalin solution that contained twenty percent.

**Determination of complete blood count (CBC)**

After the animals' blood samples were collected, blood analysis was immediately performed using citrated blood by an automated hematology analyzer DH-36 (Sysmex Corporation, Kope, Japan), which measures comprehensive blood counts, including red blood corpuscles (RBCs), hemoglobin content (Hb), hematocrit value (HCT%), mean cell volume (MCV), mean cell hemoglobin (MCH), and MCH concentration (MCHC), in addition to the total white blood cells count (WBCs), and platelet count (PLT).

**Determination of liver function and creatinine**

Following the colorimetric methods[19-22]; the albumin level and the following enzyme activities in serum were measured: alanine aminotransferase (ALT), aspartate amino-transferase (AST), γ-glutamyl transferase (GGT), and alkaline phosphatase (ALP). The kits used for these analyses were obtained from Bio-diagnostic Co. (Dokki, Giza, Egypt). In the case of bilirubin levels, a colorimetric kit was acquired from Diamonds Diagnostic Co. (Holliston, MA, USA)[23]. Creatinine levels in the blood were tested colorimetrically utilizing a kit acquired from BioMed (Cairo, Egypt)[24].

**Histological studies**

Dehydration of fixed kidney and liver tissues was accomplished using an increasing con-centration of ethyl alcohol. Following this, the tissues were cleared in xylene and inside a paraffin matrix. Following the protocol provided, sections were prepared and stained using hematoxylin and eosin (H&E)[25]. Light microscopy was then used for examination. Semiquantitative scoring for histological changes in the range "0-4" were assigned; 0: no changes, 1: minimal or small changes presented in <5% of the tissue, 2: mild changes presented in 5-20% of the tissue, 3: moderate changes presented in 20-40% of the tissue, 4: marked changes presented in >50% of the tissue compared to control group as previously described[26].

**Statistical analysis**

The statistical analysis was performed using GraphPad Prism 5.0 (GraphPad Software Inc., San Diego, CA, USA). The statistical significance was determined using one-way analysis of variance (ANOVA) and Tukey's test with a significance level of $P \leq 0.05$. The data were presented as mean ± standard error (SE) (n = 6).

**RESULTS**

**Effect of CUR+PIP on hematology of obese rat**

The HFD group showed significant decreases ($P \leq 0.05$) in the Hb content (42.6%), RBCs (51.1%), HCT (42.5%), In contrast, there was a substantial rise ($P \leq 0.05$) in WBCs (185.9%), PLT (133.5%), MCV (19.6%), and MCH (20.2%). On the other hand, the WBCs (–42.8%), PLT (–34.5), MCV (–9.0%), and MCH (–11.3%). decreased significantly ($P \leq 0.05$) in the HFD+CUR+PIP group compared with HFD group, but there was a notable rise in Hb (22.6%), RBCs (34.8%), and HCT (25.2%), No significant changes were observed between groups in the MCHC values. At the same time, neither the CUR+PIP nor the DMSO groups showed any discernible differences in the above parameters as compared to the control group (Table 1).

**Table 1:** Complete blood count (CBC) in control and different treated rat groups.

| | | Control | DMSO | CUR+PIP | HFD | HFD+CUR+PIP |
|---|---|---|---|---|---|---|
| Hb (mg/dL) | Mean±SE | 16.32±0.38 | 16.78±0.37 | 16.60±0.35 | 9.36±0.37[a] | 11.48±0.39[ab] |
| | % of [1] change | | +2.8 | +1.7 | –42.6 | –29.7 |
| | % of [2] change | | | | | +22.6 |
| RBCs ($10^6$/mL) | Mean±SE | 10.46±0.65 | 10.14±0.45 | 10.26±0.47 | 5.12±0.43[a] | 6.90±0.33[ab] |
| | % of [1] change | | –3.1 | –1.9 | –51.1 | –34.0 |
| | % of [2] change | | | | | +34.8 |
| HCT (%) | Mean±SE | 52.20±1.77 | 52.92±0.86 | 51.58±0.55 | 30.04±0.92[a] | 37.60±1.35[ab] |
| | % of [1] change | | +1.4 | –1.2 | –42.5 | –28.0 |
| | % of [2] change | | | | | +25.2 |
| MCV (fL) | Mean±SE | 50.28±2.12 | 53.02±1.75 | 50.70±2.52 | 60.16±4.41[a] | 54.74±2.13[ab] |
| | % of [1] change | | +5.4 | +0.8 | +19.6 | +8.9 |
| | % of [2] change | | | | | –9.0 |
| MCH (pg) | Mean±SE | 15.64±0.86 | 15.90±1.65 | 16.24±0.87 | 18.80±1.74[a] | 16.66±0.59[ab] |
| | % of [1] change | | +1.7 | +3.8 | +20.2 | +6.5 |
| | % of [2] change | | | | | –11.4 |
| MCHC (g/dL) | Mean±SE | 31.26±0.55 | 31.68±0.68 | 32.06±0.59 | 31.10±0.58 | 30.48±0.18 |
| | % of [1] change | | +1.3 | +2.6 | –0.5 | –2.5 |
| | % of [2] change | | | | | –2.0 |
| WBCs ($10^3$/µL) | Mean±SE | 8.82±0.67 | 8.98±0.51 | 8.76±0.60 | 25.22±1.24[a] | 14.42±1.22[ab] |
| | % of [1] change | | +1.8 | –0.7 | +185.9 | +63.5 |
| | % of [2] change | | | | | –42.8 |
| PLTs ($10^3$/µL) | Mean±SE | 366.2±30.7 | 383.2±27.4 | 374.6±15.4 | 855.2±41.5[a] | 560.4±55.8[ab] |
| | % of [1] change | | +4.6 | +2.3 | +133.5 | +53.0 |
| | % of [2] change | | | | | –34.5 |

a: significant change on comparing different groups with control group ($P \leq 0.05$), b: significant change on comparing HFD+CUR+PIP with HFD group ($P \leq 0.05$), [1]: % of change to control group, [2]: % of change to HFD group. CUR: curcumin, DMSO: dimethyl sulfoxide, Hb: hemoglobin content, HCT: hematocrit value, HFD: high-fat diet, MCH: mean cell hemoglobin, MCHC: MCH concentration, MCV: mean cell volume, PIP: piperine, PLT: platelet count, WBCs: white blood cells count, RBCs: red blood corpuscles, SE: standard error.

**Effect of CUR+PIP on liver and kidney functions of obese rat**

The findings showed that in comparison with the control group, the HFD group had substantially greater value ($P \leq 0.05$) of blood ALT (88.9%), AST (83.2%), ALP (68.9%), GGT (101.0%), total bilirubin (701.2%), and creatinine (299.5%), but substantially lower levels of albumin (66.5%, $P \leq 0.05$, Table 2). No significant changes were seen in the above parameters between the control group and the CUR+PIP or DMSO groups. However, there were significant decreases ($P \leq 0.05$) in blood ALT, AST, ALP, GGT, total bilirubin, and creatinine, as well as significant increases in albumin (91.4%) in the HFD+CUR+PIP group compared with HFD group (Table 2).

**Effect of CUR+PIP on hepatic histological changes of obese rat**

In H&E-stained liver sections, normal

arrangement of hepatic cords around prominent central veins, normal blood sinusoids, hepatocytes, and normal Kupffer cells were seen in the control, DMSO, and CUR-PIP groups (Figure 1A-C). In contrast, livers from the HFD group showed dilated central veins with hemorrhage, dilated vascular sinuses, hepatocellular swelling with numerous large vessel steatosis, activated Kupffer cells, and aggregations of inflammatory cells (Figure 1D). However, livers from obese rats treated with CUR-PIP showed improved liver architecture represented by the appearance of central vein, vascular sinus, Kupffer cells, and hepatocytes, with little microvascular fatty degeneration (Figure 1E). Scoring of the hepatic histological changes demonstrated that administration of CUR-PIP significantly reduced hepatic steatosis by ~64%, lobular inflammation by ~55%, and hepatocyte ballooning by ~50% compared to the HFD group ($P \leq 0.05$) (Table 3).

**Table 2:** Liver and kidney functions in serum of control and different treated rat groups.

| | | Control | DMSO | CUR+PIP | HFD | HFD+CUR+PIP |
|---|---|---|---|---|---|---|
| ALT (U/L) | Mean±SE | 111.8±2.8 | 108.8± 4.7 | 104.2±4.5 | 211.2±6.8[a] | 153.2±5.6[ab] |
| | % of [1] | | –2.7 | –6.8 | +88.9 | +37.0 |
| | change [2] | | | | | –27.5 |
| AST (U/L) | Mean±SE | 138.8±4.7 | 135.4±2.6 | 134.8±4.7 | 254.4±7.5[a] | 187.4±4.8[ab] |
| | % of [1] | | –2.4 | –2.9 | +83.3 | +35.0 |
| | change [2] | | | | | –26.3 |
| ALP (U/L) | Mean±SE | 342.6±13.7 | 340.2±7.7 | 336.4±11.4 | 578.8±16.4[a] | 416.2±13.2[ab] |
| | % of [1] | | –0.7 | –1.8 | +68.9 | +21.5 |
| | change [2] | | | | | –28.1 |
| Total bilirubin (mg/dL) | Mean±SE | 0.638±0.055 | 0.650±0.064 | 0.710±0.095 | 5.112±0.468[a] | 2.774±0.340[ab] |
| | % of [1] | | +1.9 | +11.3 | +701.3 | +334.8 |
| | change [2] | | | | | –45.7 |
| GGT (U/L) | Mean±SE | 57.0±4.3 | 55.90±2.70 | 54.0±2.1 | 114.6±5.4[a] | 78.3±4.6[ab] |
| | % of [1] | | –2.0 | –5.3 | +101.0 | +37.3 |
| | change [2] | | | | | –31.7 |
| Albumin (mg/dL) | Mean±SE | 9.8±0.3 | 10.1±0.4 | 10.3±0.3 | 3.3±0.2[a] | 6.3±0.4 [ab] |
| | % of [1] | | +2.8 | +5.1 | –66.5 | –36.0 |
| | change [2] | | | | | +91.4 |
| Creatinine (mg/dL) | Mean±SE | 0.854±0.069 | 0.650±0.059 | 0.716±0.095 | 3.412±0.198[a] | 1.946±0.117 [ab] |
| | % of [1] | | –23.9 | –16.2 | +299.5 | +127.9 |
| | change [2] | | | | | –43.0 |

a: significant change on comparing different groups with control group ($P \leq 0.05$), b: significant change on comparing HFD+CUR+PIP with HFD group ($P \leq 0.05$), [1]: % of change to control group, [2]: % of change to HFD group, ALT: alanine aminotransferase, ALP: alkaline phosphatase, AST: aspartate aminotransferase, CUR: curcumin, DMSO: dimethyl sulfoxide; GGT: γ-glutamyl transferase, HFD: high-fat diet, PIP: piperine, SE: standard error.

**Effect of CUR+PIP on renal histological changes of obese rat**

Normal glomeruli, intact brush boundaries on proximal convoluted tubules, and the absence of interstitial gaps in distal convoluted tubules were seen in kidney

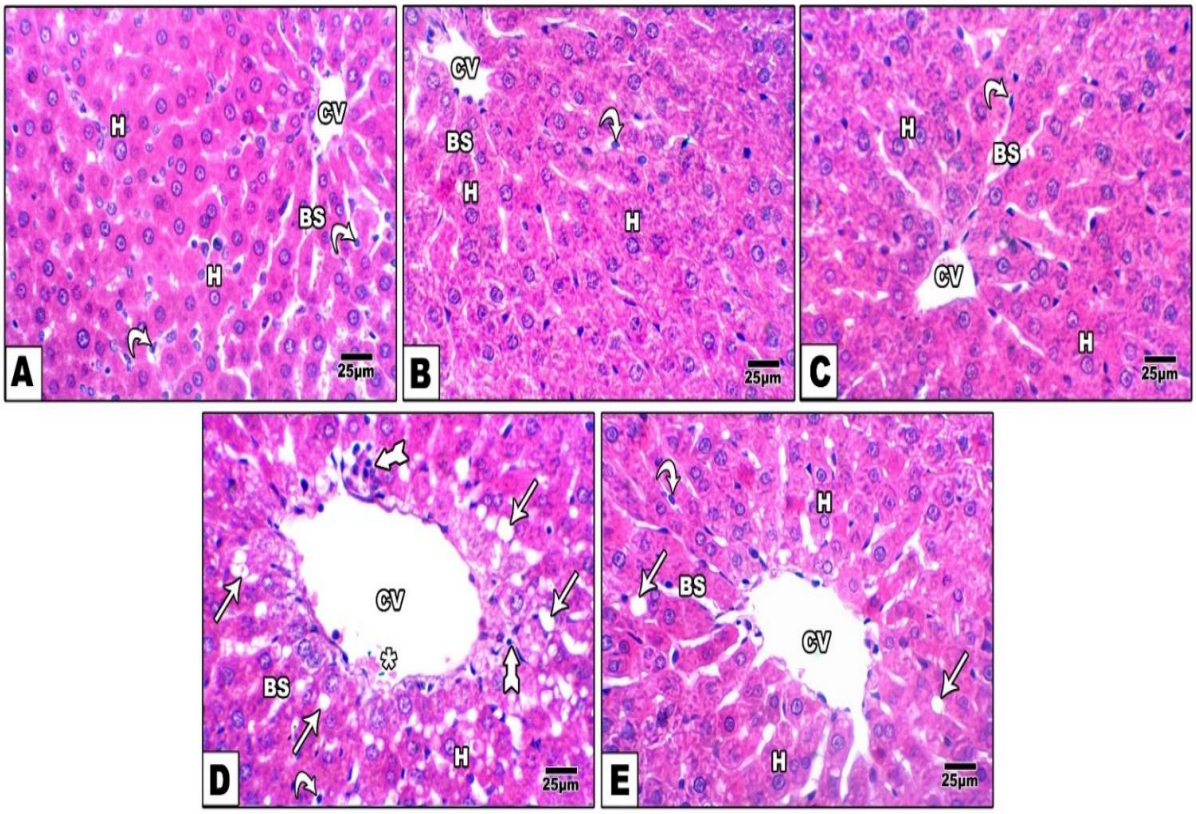


**Figure 1:** Photomicrograph of hematoxylin and eosin (H&E) stained liver section in control **(A)**, DMSO **(B)**, and curcumin plus piperine **(C)** groups showed normal arrangement of hepatic cords around distinct central vein (CV) with normal blood sinusoids (BS), hepatocytes (H), and normal kupffer cells (curved arrow). Liver sections from HFD group **(D)** showed dilated central vein (CV) with hemorrhage (asterisk) and dilated blood sinusoids (BS), ballooning of hepatocytes (H) with many macrovasicular steatosis (arrow), activated kupffer cells (curved arrow), and inflammatory cells aggregation (tailed arrow). However, HFD-fed rats treated with curcumin plus piperine **(E)** showed improvement in liver architecture represented in appearance of central vein (CV), blood sinusoids (BS), kupffer cells (curved arrow), and hepatocytes (H) with few microvasicular steatosis (arrow). Original magnification ×400, scale bar = 25 µm.

**Table 3:** Histopathological score of liver lesions observed in the different treated rat groups.

| Hepatic lesions | Control | DMSO | CUR+PIP | HFD | HFD+CUR+PIP |
|---|---|---|---|---|---|
| Steatosis | 0 | 0 | 0 | 1.83±0.16[a] | 0.66±0.21[b] |
| Lobular inflammation | 0 | 0 | 0 | 1.50±0.22[a] | 0.67±0.20[b] |
| Hepatocyte ballooning | 0 | 0 | 0 | 1.66±0.21[a] | 0.83±0.30[b] |

Histopathological changes in liver were evaluated using a semiquantitative scoring system. a: significant change on comparing different groups with control group ($P \leq 0.05$), b: significant change on comparing HFD+CUR+PIP with HFD group ($P \leq 0.05$). CUR: curcumin, DMSO: dimethyl sulfoxide; HFD: high-fat diet, PIP: piperine.

sections from the control, DMSO, and CUR-PIP groups (Figures 2A-C). However, renal cortical sections from the HFD group showed dilated glomeruli, congested inter-tubular vessels, and edematous degeneration in the epithelial cells lining some proximal convoluted tubules (Figure 2D) Nevertheless, when CUR-PIP were taken together, the symptoms of enlarged glomeruli, normal proximal convoluted tubules having intact brush borders, normal distal convoluted tubules with no interstitial space (Figure 2E). Scoring of the renal histological changes demonstrated that administration of CUR-PIP alleviated the histopathological changes that induced by HFD, as tubular damage was reduced by about 88%, congestion was totally prevented (100% reduction), and glomerular involvement decreased by about 45% compared to the HFD group ($P \leq 0.05$) (Table 4).

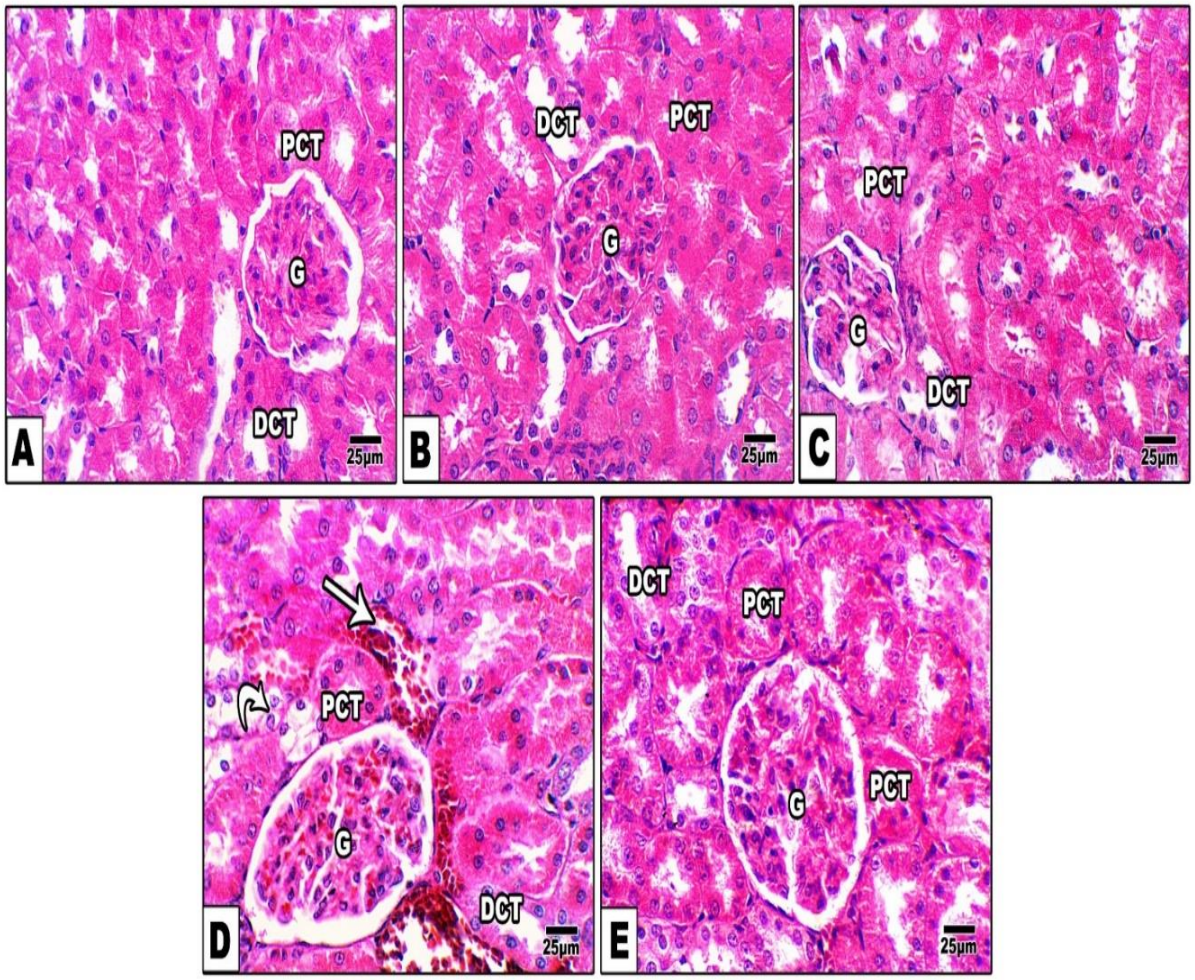


**Figure 2:** Photomicrograph of hematoxylin and eosin (H&E) stained kidney sections in control **(A)**, DMSO **(B)**, and curcumin plus piperine **(C)** groups showed normal glomeruli (G), normal proximal convoluted tubules (PCT) having intact brush borders, and normal distal convoluted tubules (DCT) with no interstitial space. Renal cortical sections from HFD group **(D)** showed enlarged glomeruli (G), congested intertubular blood vessels (arrow), hydropic degeneration in epithelial cells lining few PCT (curved arrow). However, renal cortical sections from HFD + curcumin + piperine group **(E)** showed enlarged glomeruli (G), normal proximal convoluted tubules (PCT) having intact brush borders, and normal distal convoluted tubules (DCT) with no interstitial space. Original magnification ×400, scale bar = 25 µm.

**Table 4**: Histopathological score of renal lesions observed in the different treated rat groups.

| Renal lesions | Control | DMSO | CUR+PIP | HFD | HFD+CUR+PIP |
|---|---|---|---|---|---|
| Tubular damage | 0 | 0 | 0 | 1.33±0.21[a] | 0.16±0.15[b] |
| Congestion | 0 | 0 | 0 | 1.16±0.14[a] | 0[b] |
| Glomerular involvement | 0 | 0 | 0 | 1.20±0.16[a] | 0.66±0.21[b] |

Histopathological changes in kidney were evaluated using a semiquantitative scoring system. a: significant change on comparing different groups with control group ($P \leq 0.05$), b: significant change on comparing HFD+CUR+PIP with HFD group ($P \leq 0.05$). CUR: curcumin, DMSO: dimethyl sulfoxide; HFD: high-fat diet, PIP: piperine.

## DISCUSSION

The buildup of extra fat in the body is known as obesity, and it is a significant public health concern, which poses risks to the health of various organs[27]. Diabetes, hypertension, and a number of malignancies are just a few of the illnesses linked to obesity[27]. HFDs that cause obesity to force the body to develop and increase pro-inflammatory cytokines, such as interleukin (IL)-1 and tumor necrosis factor (TNF)-α, and reduce the anti-inflammatory adiponectin. IL-1 contributes with synthesizes and secretes C-reactive protein (CRP) when its levels are elevated in the liver. CRP is evidence of having systemic inflammation[27,28]. As a result from generating and progression of inflammation, the levels of reactive oxygen species (ROS) elevated and nitric oxide (NO) decreased leading to oxidative stress that causes dysfunctional cells, vessels, and organs[28].

Changes in hematological parameters, including the increase in WBCs, RBCs, and platelet counts shown in the current study, are associated with obesity through various mechanisms related to oxidative stress and inflammation[29]. Hyperlipidemia in obesity might heighten oxidative stress by increasing free radical and ROS production leading to damage both plasma membranes and cytoplasmic components, as well as hemolysis of RBCs and a reduction in circulating RBCs[29]. In the current study, hematological tests of HFD group revealed a significant increase in MCV, while MCH values were nearly within the normal range. Alongside reductions in Hb, RBCs, and HCT all of which indicated the development of macrocytic normochromic anemia, which could be attributed to liver dysfunction associated with obesity. A rise in platelet counts with increased inflammatory responses are other issues of obesity, which in turn promote platelet aggregation and thrombus formation[30]. It is also probable that IL-6 plays a pivotal role in obese subject, as it increases thrombopoietin and stimulates megakaryocytopoiesis *via* its synergistic effects with other proinflammatory cytokines; thrombopoietin might potentially come from visceral adipose tissue as well[30]. In addition, inflammation is a hallmark of obesity caused by a high-fat diet, which contributes to increase the WBCs count and CRP, promoting cardiovascular disorders[30,31]

The liver is an organ that plays a key role in nearly all biochemical pathways involved in growth, disease control, nutrient transport, and regulating internal homeostasis[32]. As fat storage increases, the liver begins to undergo oxidative membrane damage, leading to the leakage of endogenous enzymes (ALT, AST, ALP, and GGT) into the circulation[32]. Serum ALT, AST, ALP, GGT, and total bilirubin activity were significantly elevated, while albumin was significantly decreased, in rats given

a high-fat diet in the current study, indicating that the hepatocytes in these animals were not functioning properly. This suggests that the rats may have liver injury and hepatotoxicity due to hyperlipidemia and fatty liver. The histological observation of the liver of HFD-fed rats revealed a dilated central vein with hemorrhage, dilated vascular sinuses, enlarged hepatocytes with numerous macrovascular fatty liver, activated Kupffer cells, steatosis in a diffused manner all over the hepatocytes associated with focal necrosis and hepatocellular ballooning, and aggregation of inflammatory cells. Wardani *et al*.[33] also reported similar changes in hepatic histology in obese rats. In the current study, obese rats given CUR-PIP had protective effects against liver damage and improved liver structure and functions. Other scientists reported that phosphatidylserine- and PIP-containing CUR phytosomes improved liver functions in patients with NAFLD[14]. In addition, CUR+PIP alleviated dyslipidemia, oxidative stress, and inflammation and protected from liver damage in obese rats[16]. Besides Chang *et al*.[34] has declared that obese mice, which treated with CUR for 10 weeks have shown significant decrease in fatty liver and small fat cells, and exhibited a 36% lower average liver fat infiltration score comparing to HFD group. Whereas, in our study, HFD-rats that received CUR-PIP have shown improvements at histological level of liver by about 50–64% compared to the HFD group.

Kidney disease is mostly caused by dietary and lifestyle factors[35]. Obesity increases the risk of kidney disease by causing lipids to accumulate within the cells of the kidneys. Increased perirenal fat accumulation leads to renal lipotoxicity with renal lipid accumulation[35]. Lipotoxicity causes renal dysfunction primarily through a decrease in mitochondrial membrane potential with ATP depletion, reduced renal tubular fluid flow, and decreased renal vascular elasticity[36]. Renal function may be evaluated by measuring serum creatinine and blood urea nitrogen (BUN)[36]. Obese rats in the HFD group had much higher blood creatinine in the current study that may reflect a reduction in the glomerular filtration. The histological observation of the kidneys of HFD-fed rats revealed glomerular hypertrophy, congestion of inter-tubular vessels, and hydropic degeneration of epithelial cells lining some proximal convoluted tubules. Other scientists also reported similar changes in the renal histology of the obese rats[36,37]. On the other hand, administration of CUR-PIP to HFD-fed rats showed significant improvement in renal histology and function. Other scientists reported the renoprotective activity of CUR+black pepper in obese rats[38]. There is not enough studies to declare or mentioned any scoring of renal lesion because of obesity and compare it to the scoring after treatment with CUR-PIP. However, in our study the findings have shown that the combined administration of CUR-PIP has developed modulation for tubular damage and glomerular involvement, with complete prevention of vascular congestion.

As curcumin exhibits remarkable effect as antioxidant, anti-inflammatory, antitoxic, and antiobesity component, and it is recommended to be consumed by people who suffer from obesity or at risk of developing it in order to prevent or delay any further health risks such as organs' dysfunction[10]. Also consumption black pepper that is the most using spice with its main component piperine has been proven that it has an exceptional role in reducing the risk factors that related to obesity-complications, as it has outstanding revers effect on liver lipid accumulation at HFD consumers when administrated orally and thus it can serve as a protective trail for healthy weight loss[39]. Also, it had been recorded that curcumin cannot be utilized alone and get that significant effect of its compound and this is due to its poor bioavailability as it gets rapid metabolism in liver however the effect of its bioavailability is raised by about (2000%) when combined with piperine[40]. Combing curcumin plus piperine also showed remarkable influence

on blood dyslipidemia compared to the effect of curcumin consumption alone[12]. Not only the curcumin has the remarkable effect to maintain the kidney function, as it reduced the amount of HFD that induces the renal damage by enhancing antioxidative defensive mechanisms in the liver and the kidney, but also it leads to increase the antioxidant enzymes and indicates less neuroinflammation[34].

In conclusion, disorders of the blood, liver, and kidneys of male Wistar rats occurred as a result of a high-fat diet that is consumed over an extended period (8 weeks). In addition, these risk variables were considerably reduced when CUR-PIP were taken together. So, to counteract the health concerns associated with obesity including the hematotoxicity, hepatic toxicity, and renal toxicity without adverse effects, CUR-PIP taken regularly may be a good treatment alternative.

## CONFLECT OF INTEREST

Authors declare that there is no significant financial or non-financial conflict of interests of the authors.

**Funding**

The authors did not receive any grant, fund, or other assistance to prepare this paper.

## AUTHORS' CONTRIBUTIONS

N.M., H.R. and A.A.H. provided the original idea for the work, organized the study, and drafted the manuscript. E.O.M. carried out animal experiments, collected the data, and performed the analysis. E.O.M. and H.R. both contributed to the study design, interpreted the results, and revised the manuscript. All authors have read and approved the final manuscript.

## REFERENCES

[1] Safaei, M.; Sundararajan, E. A.; Driss, M. *et al*. (2021). A systematic literature review on obesity: understanding the causes & consequences of obesity and reviewing various machine learning approaches used to predict obesity. Comput Biol Med, 136: 104754 (DOI: 10.1016/j.compbiomed.2021.104754).

[2] Pakhare, M. and Anjankar, A. (2024). Critical correlation between obesity and cardiovascular diseases and recent advancements in obesity. Cureus, 16(1): e51681 (DOI: 10.7759/cureus. 51681).

[3] Welsh, A.; Hammad, M.; Piña, I. L. *et al*. (2024). Obesity and cardiovascular health. Eur J Prev Cardiol, 31(8): 1026-1035.

[4] Omer, T. (2020). The causes of obesity: an in-depth review. Adv Obes Weight Manag Control, 10(3): 90-94.

[5] Bernier, M.; Mitchell, S. J.; Wahl, D. *et al*. (2020). Disulfiram treatment normalizes body weight in obese mice. Cell Metab, 32(2): 203-214.e4.

[6] Zhu, T.; Chen, Q.; Chen, H. *et al*. (2024). Independent and interactive associations of heart rate and obesity with type 2 diabetes mellitus: a population-based study. J Diabetes, 16(4): e13529 (DOI: 10.1111/1753-0407.13529).

[7] González-Arceo, M.; Aguirre, L.; Macarulla, M. T. *et al*. (2024). Effect of *Gracilaria vermiculophylla* Macroalga on non-alcoholic fatty liver disease in obese rats. Antioxidants (Basal), 13(3): 369 (DOI: 10.3390/ antiox13030369).

[8] Huang, X.; Ji, Q.; She, C.-Y. *et al*. (2023). Ophiopogonin D ameliorates non-alcoholic fatty liver disease in high-fat diet induced obese mice by improving lipid metabolism, oxidative stress and inflammatory response. Exp Ther Med, 26(3): 418 (DOI: 10.3892/ etm.2023.12116).

[9] Ulla, A.; Alam, M. A.; Sikder, B. *et al*. (2017). Supplementation of *Syzygium cumini* seed powder prevented obesity, glucose intolerance, hyperlipidemia and oxidative stress in high carbohydrate high fat diet-induced obese rats. BMC Complement Altern Med, 17: 289 (DOI: 10.1186/s12906-017-1799-8).

[10] Hassan, M. H.; Awadalla, E. A.; Abd El-Kader, A. E.-K. M. *et al.* (2022). Antitoxic effects of curcumin against obesity-induced multi-organs' bio-chemical and histopathological ab-normalities in an animal model. Evid Based Complement Alternat Med, 2022: 9707278 (DOI: 10.1155/2022/ 9707278).

[11] Shao, W.; Yu, Z.; Chiang, Y. *et al*. (2012). Curcumin prevents high fat diet induced insulin resistance and obesity *via* attenuating lipogenesis in liver and inflammatory pathway in adipocytes. PLoS ONE, 7(1): e28784 (DOI: 10.1371/journal.pone.0028784).

[12] Tu, Y.; Sun, D.; Zeng, X. *et al*. (2014). Piperine potentiates the hypo-cholesterolemic effect of curcumin in rats fed on a high fat diet. Exp Ther Med, 8(1): 260-266.

[13] Scazzocchio, B.; Minghetti, L. and D'Archivio, M. (2020). Interaction between gut microbiota and curcumin: a new key of understanding for the health effects of curcumin. Nutrients, 12(9): 2499 (DOI: 10.3390/nu1209 2499).

[14] Cicero, A. F. G.; Sahebkar, A.; Fogacci, F. *et al*. (2020). Effects of phytosomal curcumin on anthro-pometric parameters, insulin resistance, cortisolemia and non-alcoholic fatty liver disease indices: a double-blind, placebo-controlled clinical trial. Eur J Nutr, 59: 477-483.

[15] El-Habibi, E. M.; El-Wakf, A. M. and Mogall, A. (2013). Efficacy of curcumin in reducing risk of cardiovascular disease in high fat diet-fed rats. J Bioanal Biomed, 5(3): 066-070.

[16] Antona, M. E.; González, P. M.; Ramos, C. *et al*. (2021). Curcumin exerts a protective effect against obesity and liver injury induced by an atherogenic diet. Funct Foods Health Dis, 11(12): 673-689.

[17] Jiang, M.; He, J.; Sun, Y. *et al*. (2021). Leptin induced TLR4 expression *via* the JAK2-STAT3 pathway in obesity-related osteoarthritis. Oxid Med Cell Longev, 2021: 7385160 (DOI: 10.1155/2021/7385160).

[18] Wellington, D.; Mikaelian, I. and Singer, L. (2013). Comparison of ketamine–xylazine and ketamine–dexmedetomidine anesthesia and intra-peritoneal tolerance in rats. J Am Assoc Lab Anim Sci, 52(4): 481-487.

[19] Reitman, S. and Frankel, S. (1957). A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. Am J Clin Pathol, 28: 56-63.

[20] Szasz, G. (1969). A kinetic photo-metric method for serum γ-glutamyl transpeptidase. Clin Chem, 15(2): 124-136.

[21] Belfield, A. and Goldberg, D. M. (1971). Normal ranges and diagnostic value of serum 5′-nucleotidase and alkaline phosphatase activities in infancy. Arch Dis Child, 46(250): 842-846.

[22] Doumas, B. T.; Watson, W. A. and Biggs, H. G. (1971). Albumin standards and the measurement of serum albumin with bromcresol green. Clin Chim Acta, 31(1): 87-96.

[23] Kaplan, A.; Rubaltelli, F. F.; Hammer-man, C. *et al*. (1984). Bilirubin. In: Clinical Chemistry (Kaplan, A. and Peace, A. L., eds), pp. 1238-1241. C.V. Mosby Co., St Louis, Mo, USA.

[24] Toora, B. D. and Rajagopal, G. (2002). Measurement of creatinine by Jaffe's reaction—determination of concentra-tion of sodium hydroxide required for maximum color development in standard, urine and protein free filtrate of serum. Indian J Exp Biol, 40(3): 352-354.

[25] Suvarna, K. S.; Layton, C. and Bancroft, J. D. (2019). Bancroft's Theory and Practice of Histological Techniques. Elsevier, Amsterdam, Netherlands.

[26] Schafer, K. A.; Eighmy, J.; Fikes, J. D. *et al*. (2018). Use of severity grades to characterize histopathologic changes. Toxicol Pathol, 46(3): 256-265.

[27] Blüher, M. (2025). An overview of obesity-related complications: the epidemiological evidence linking body weight and other markers of obesity to adverse health outcomes. Diabetes Obes Metab, 27 (Suppl 2): 3-19.

[28] Ellulu, M. S.; Patimah, I.; Khaza'ai, H. *et al*. (2017). Obesity and inflammation: the linking mechanism and the complications. *A*rch Med Sci, 13(4): 851-863.

[29] Nagamma, T.; Konuri, A.; Nayak, C. D. *et al*. (2019). Dose-dependent effects of fenugreek seed extract on the biochemical and haematological parameters in high-fat diet-fed rats. J Taibah Univ Med Sci, 14(4): 383-389.

[30] Purdy, J. C. and Shatzel, J. J. (2021). The hematologic consequences of obesity. Eur J Haematol, 106(3): 306-319.

[31] Maysami, S.; Haley, M. J.; Gorenkova, N. *et al*. (2015). Prolonged diet-induced obesity in mice modifies the inflammatory response and worsens stroke outcomes. J Neuroinflammation, 12: 140 (DOI: 10.1186/s12974-015-0359-8).

[32] Oriquat, G.; Masoud, I. M.; Kamel, M. A. *et al*. (2023). The anti-obesity and anti-steatotic effects of chrysin in a rat model of obesity mediated through modulating the hepatic AMPK/mTOR/ lipogenesis pathways. Molecules, 28(4): 1734 (DOI: 10.3390/molecules 28041734).

[33] Wardani, H. A.; Rahmadi, M.; Ardianto, C. *et al*. (2019). Development of nonalcoholic fatty liver disease model by high-fat diet in rats. J Basic Clin Physiol Pharmacol, 30(6): (DOI: 10.1515/jbcpp-2019-0258).

[34] Chang, G.-R.; Hsieh, W.-T.; Chou, L.-S. *et al*. (2021). Curcumin improved glucose intolerance, renal injury, and nonalcoholic fatty liver disease and decreased chromium loss through urine in obese mice. Processes, 9(7): 1132 (DOI: 10.3390/pr9071132).

[35] Soliman, N. and Keshk, W. (2012). Effect of porpoise oil on kidney of rats receiving high-fat diet. Bull Egypt Soc Physiol Sci, 32(2): 43-56.

[36] Pengrattanachot, N.; Cherngwelling, R.; Jaikumkao, K. *et al*. (2020). Atorvastatin attenuates obese-induced kidney injury and impaired renal organic anion transporter 3 function through inhibition of oxidative stress and inflammation. Biochim Biophys Acta Mol Basis Dis, 1866(6): 165741. (DOI: 10.1016/j.bbadis.2020.165741)

[37] Rojas, J. M.; Bolze, F.; Thorup, I. *et al*. (2018). Effect of diet-induced obesity on toxicological parameters in the polygenic Sprague-Dawley rat model. Toxicol Pathol, 46(7): 777-798.

[38] Mohamed, R. R. A. R. (2022). Study the effect of combining curcumin and black pepper in different proportions on obese diabetic rats. J Res Fields Spec Educ, 8(43): 1399-1424.

[39] Wang, X.; Zhang, Y.; Zhang, L. *et al*. (2022). Piperine attenuates hepatic steatosis and insulin resistance in high-fat diet-induced obesity in Sprague-Dawley rats. Nutr Res, 108: 9-21.

[40] Shoba, G.; Joy, D.; Joseph, T. *et al*. (1998). Influence of piperine on the pharmacokinetics of curcumin in animals and human volunteers. Planta Med, 64(4): 353-356.

# التأثير المُشترك للكركمين والبيبيرين على اختلالات وظائف الدم والكبد والكُلى الناجمة عن السمنة لدى ذكور جرذان ويستار

حنان رشوان[1]، ناهد منصور[1]، إيمان محمد[1]، عايدة حسين[2]
[1]قسم علم الحيوان، كلية العلوم، جامعة العريش، شمال سيناء، جمهورية مصر العربية
[2]قسم علم الحيوان، كلية العلوم، جامعة السويس، السويس، جمهورية مصر العربية

أحد التأثيرات السلبية للسمنة هو سُمية الدم وإصابة الأعضاء الحيوية. يهدف هذا البحث إلى تحديد ما إذا كان بإمكان الجرذان السمينة تجنب تلف كريات الدم الحمراء والكبد والكُلى عن طريق تناول الكركمين (100 ملجم/كجم من وزن الجرذ) والبيبيرين (5 ملجم/كجم من وزن الجرذ)، عبر أنبوب معدة، يوميًا، لمدة ثمانية أسابيع. ارتفعت أنشطة ألانين وأسبارتات أمينوترانسفيراز، والفوسفاتيز القلوي، وجاما جلوتاميل ترانسفيراز، وعدد الصفائح الدموية وخلايا الدم البيضاء، بالإضافة إلى مستوى البيليروبين الكلي والكرياتينين في الدم بشكل حاد ($P\leq0.05$) بعد تناول نظام غذائي غني بالدهون، ولكن انخفض مستوى الألبومين بشكل حاد ($P\leq0.05$). كما أظهرت الجرذان السمينة فقر دم كبير الكريات سوي الصبغة. وأظهرت أكباد وكُلى الجرذان السمينة العديد من التشوهات عند التحليل النسيجي. أظهرت أكباد المجموعة التي اتبعت نظامًا غذائيًا غنيًا بالدهون توسعًا في الأوردة المركزية مع نزيف، وتوسعًا في الجيوب الوعائية، وتضخمًا في خلايا الكبد مع وجود العديد من الكبد الدهني في الأوعية الدموية الكبيرة، وتنشيطًا لخلايا كوبفر، وتجمعات من الخلايا الالتهابية. وأظهرت مقاطع القشرية الكُلوية للمجموعة التي اتبعت نظامًا غذائيًا غنيًا بالدهون توسعًا في الكبيبات، واحتقانًا في الأوعية بين الأنابيب، وتنكسًا وذميًا لبعض الخلايا الظهارية المبطنة. وقد خفف تناول مكملات الكركمين والبيبيرين من سُمية الدم المرتبطة بالسمنة، بالإضافة إلى تلف الكبد والكُلى، بشكل ملحوظ إحصائيًا ($P\leq0.05$). لذلك، قد توصي هذه النتائج بتناول مكملات الكركمين والبيبيرين يوميًا للوقاية من المخاطر الصحية المرتبطة بالسمنة وسُمية الكبد والكُلى.