

Effects of Soybean Diets Supplemented with Cardamom, Green Coffee and Olive Leaves on Liver Functions and Antioxidant Enzymes in Acute Renal Failure Rats

Ashraf Abd El- Aziz Abd El-Megid *, Seham A.M. Tharwat*,
And Eman Mohamed Sabry Abd El-Aziz El-Ashmawy**

*Department of Nutrition and Food Science. Faculty of Home
Economics, Helwan University, Egypt.

**Graduate student, Nutrition and Food Science Dept., Faculty of
Home Economics, Helwan University, Egypt.

ABSTRACT

This study aimed to investigate the effects of a soybean protein diet (SD), BD and a mixture between SD and Casein (1:1w/w), supplemented with 5% cardamom (Car), green coffee (GC) and olive leaves (OL), or a 5% mixture of them, on total protein, albumin and globulin, liver enzymes, and antioxidant enzymes in acute renal failure rats (ARFR). A total of 78 adult male albino rats of the Spragu Dawley strain weighing 200 ± 10 g were used in this study. The rats were divided into two main groups. The first main group (6 rats) was fed on basal diet (BD) and used as a control negative group (-ve). The second main group (72) rats were injected with glycerol to induce acute renal failure. These were divided into (12) subgroups. One of them (6 rats) was fed on (BD) and used as the (+ve) control group. The other subgroups (11 subgroups) were fed on (SD) or (BD) or mixed protein (1:1) from soybean and casein, supplemented with 5% from (Car) , (GC) (OL), or 5% mixture between them for (8 weeks). At the end of the experiment, rats were anesthetized with ether before being sacrificing after fasting overnight. The liver and kidney were removed, then taken and weighted. Blood samples were collected, left to clot, then serum was separated. The serum total protein, albumin and globulin, antioxidant enzymes glutathione peroxidase (GSH-PX), superoxide dismutase (SOD) and catalase (CAT) and liver enzymes activities aspartate aminotransferase (AST), alanine amine transferase (ALT) and alkaline phosphates (ALP) levels were determined. The obtained results indicated that glycerol injection induced acute renal failure (ARF) led to a significant decrease in BWG% and a significant increase in liver weight% BWG%, while there was no significant difference in FI. Results also revealed that

the (+ve) control group recorded a significant decrease in serum, total protein, albumin, globulin, total protein, and antioxidant enzymes (GSH-PX, SOD & CAT), while induced a significant increase in liver enzymes (AST, ALT and ALP) levels as compared to the (-ve) control group. Our results concluded that dietary treatments with 5% Car, GC or OL (SD) or BD supplement or mixed between (SD plus casein) (1:1 w/w) supplemented with 5% of (Car, GC or OL) or 5% mixture of (Car, GC or OL) induced a significant increase in TP, Alb and antioxidant enzymes (GSH-Px, SOD and CAT), while leading to a significant decrease in liver enzymes (AST, ALT and ALP). The best result induced a significant improvement induced by mixed soybean plus casein (1:1 w/w) supplemented with 5% mixture from (Car, GC & OL). This combination can be used to reduce the side effects of kidney and liver toxicity. Therefore, combination treatment with (1:1 w/w) soybean and casein supplemented with a 5% mixture of (Car, GC and OL) may be beneficial for liver and kidney disease can also be used for protection against kidney and liver toxicity.

Keywords : Soybean diets, Cardamom, Green coffee, Olive leaves, Liver functions, Antioxidant enzymes and Acute renal failure

INTRODUCTION:

Liver plays an essential role in the metabolism of foreign compounds entering the body. Human beings are exposed to these compounds through environmental exposure, consumption of contaminated food during exposure to chemical substances in the occupational environment. All these compounds produce a variety of toxic manifestations. (Athar et al., 1997) Glycerol injections are used to induce acute renal failure (ARE) in rats due to rhabdomyolysis and myoglobin release, resulting in ischemic injury and nephrotoxicity.

Sharma and Singh, (2014) demonstrated that the discovery of natural antioxidants has risen exponentially. The principal candidates in this discovery process are medical plants. There are many spices and seeds that contain a high proportion of antioxidants concentrated in the form of flavonoids, which are derivatives of polyphenols that may contribute to counteracting the **oxidation pressure caused by toxic compounds in the human body and work to neutralize them (Steinmetz and Potter, 1996).** Cardamom

is an herbaceous perennial belonging to the family Zingiberaceae. The dried fruit is used either whole or in ground form as a flavoring agent and in medicinal preparation for ingestion for flatulence (**Leela et al., 2008**). Cardamom has been employed in traditional medicinal plants used against kidney and urinary disorders (**Ballabh et al., 2008**). Some studies showed that extracts and their constituents from cardamom also possess hepatoprotective activity (**Kandasamy et al., 2010**) and antioxidative effects (**Das et al., 2012**).

Cardamom contains flavonoids and polyphenols that exhibit strong antioxidant activity, protecting the liver from oxidative stress by neutralizing free radicals. Studies have demonstrated that cardamom extracts can prevent liver damage caused by toxins. For example, Kandasamy et al. (2010) highlighted its ability to safeguard liver cells, while its anti-inflammatory properties help in reducing liver inflammation and preventing fibrosis **Kandasamy, (2010)**.

Coffee is a commonly consumed beverage comprising a complex mixture of compounds, including caffeine, chlorogenic acid, and diterpenes (**Ludwig et al., 2014**). These have a range of in vivo properties, including anti-inflammatory, antioxidants, and antifibrotic effects (**Ponte, 2002**). Epidemiologic studies indicate that coffee may protect against liver, neurologic, cardiovascular, and metabolic diseases (**Poole et al., 2017**). As a functional food with antioxidant properties, coffee reduces the incidence of cancer, diabetes, and liver diseases (**Jeszka-Skowron et al., 2016**).

The consumption of green coffee has been associated with lower levels of liver enzymes, such as ALT and AST, which are indicators of liver damage, suggesting its role in maintaining healthy liver function and preventing non-alcoholic fatty liver disease (NAFLD) **Ludwig, (2014)**.

Olive (*Olea europaea* L) leaves are rich in phenolic compounds such as oleuropein, rutin, luteol and epigenin, triterpenes and chalcones (**Pereira et al., 2007**). It has been shown that olive leaf extract has prominent protective effects against methotrexate-induced hepatotoxicity (**ElAzim , 2014**).Olive leaf extracts also have anti-inflammatory and anti-apoptotic effects, contributing to liver protection against drug-induced hepatotoxicity **ElAzim, (2014)**.

Cardamom, green coffee, and olive leaves provide significant benefits for liver health through their antioxidant, anti-inflammatory, and hepatoprotective properties. These natural substances protect against oxidative stress, inflammation, and toxin-induced liver damage, making them valuable in maintaining liver function and preventing liver diseases

Elgebaly et al., (2018) reported that olive oil and olive leaves extract can significantly prevent fluoxetine-induced hepatotoxicity and reduce inflammation, oxidative stress, and apoptosis in rats. Therefore, this study aimed to investigate the effects of soybean diets (SD), (BD) and mixed (SD) plus casein (1:1w/w) supplemented with 5% Car, GC, and OL or 5% mixture from them on liver functions and antioxidant enzymes of rats suffering from glycerol toxicity.

MATERIALS AND METHODS:

Material: Casein, vitamins, minerals, cellulose, and choline chloride were purchased from El-Gomhoria Company, Cairo, Egypt. Soybeans were obtained from the Agricultural Research Center in Cairo, Egypt. Cardamom, green coffee, and olive leaves were obtained from Agricultural Herbs and Medicinal Plants, Cairo, Egypt. Kits for biochemical analysis were obtained from Alkan for Pharmaceutical and Chemical Dokki, Egypt.

Rats: Seventy-eight adult male albino rats (Spragu Dawley strain) weighing 200 ± 10 were purchased from the Helwan farm of experimental animals, Ministry of Health and Population, Helwan, Cairo, Egypt.

Methods:

Experimental Design:

After acclimation to a basal diet (BD) for one week, rats were classified into two main groups. The first main group ($n = 6$) fed on (BD) as a control negative group (-ve). The second main group (72 rats) was injected with glycerol (50% weight/volume) in 0.9% saline at 5 ml/kg to induce acute renal failure according to the method described by **Maree et al., (1994)**.

The positive injected groups: the first group was fed a basal diet (BD), the second group served as a control group and was fed a soy protein diet (SD), while the remaining groups were fed either a basal diet (BD) or a soy protein diet (SD), and/or diets containing a

combination of casein and soy beans as sources of protein (1–1) supplemented with 5% (Car), (GC), and olive leaves (OL). The diet consumed was recorded every week. At the end of the experiment, the rats were fasted overnight, and then the rats were anesthetized with pentobarbital sodium (40 mg/kg) and sacrificed. Blood samples were collected from the aorta of all rats. The blood samples were centrifuged, and serum was separated by centrifugation of the blood sample. Then it was kept frozen at -20 °C until the analysis. The liver of each rat was removed from rats by careful dissection, washed in a saline solution (0.9%), dried using filter paper, and independently weighed.

Biochemical Analysis:

The analysis of serum proteins followed the method described by **Gomal et al. (1949)**, serum albumin was determined following the protocol outlined by **Doumas and Biggs (1971)**, The serum globulin level was determined by subtracting the albumin level from the total protein concentration, while aspartate aminotransferase AST and alanine aminotransferase ALT activities were measured according to **Henry (1974)**, and alkaline phosphatase levels were determined as per **Belfield and Goldberg (1971)**.

Determination of Glutathione peroxidase (GSH-Px) and superoxide dismutase (SOD) .

- Liver tissue samples:

At the end of the experimental period, rats were sacrificed. The liver specimen was quickly removed and weighted, then perfused with cold saline to exclude the blood cells and then blotted on filter paper; and stored at -20°C. Briefly, liver tissues were cut, weighed and minced into small pieces, homogenized with a glass homogenizer in 9 volumes of ice-cold 0.05 mM potassium phosphate buffer (pH7.4) to make 10% homogenates. The homogenates were centrifuged at 5,000 r.p.m for 15 minutes at 4°C, then the supernatant was used for the determination of superoxide dismutase (SOD) and Glutathione peroxidase (GSH-Px) activities were measured according to the methods described by (**Beauchamp and Fridovich, 1971 and Paglia & Valentine, 1967**). respectively.

Statistical Analysis:

The biochemical and biological data for each group were analyzed using descriptive statistics (mean and SD) and one-way ANOVA to assess group differences. Significant results were identified with a P-value threshold of < 0.05 . All analyses were conducted using SAS software (SAS, 2004).

RESULTS AND DISCUSSION:

Impact of soybean diets supplemented with cardamom, green coffee, or olive leaves on feed intake, body weight gain%, and liver weight/BW% of rats suffering from acute renal failure

Table (1) illustrates the effect of soybean diet (SD), basal diet (BD), and mixed (1:1w/w) of them supplemented with Car, GC and OL on feed intake FI, body weight gain% (BWG%), and liver weight / BW% of rats suffering from glycerol toxicity (ARF). Results revealed that (+ve) groups recorded a non-significant difference in FI as compared to the (-ve) control group, while body weight gain % results for the (-ve) group indicated a significant increase in BWG% as compared to the (+ve) control group. Concerning all (+ve) groups fed on all treatments (SD), (BD) or mixed (SD plus casein) supplemented with 5% Car, GC or OL and mixed 5% from them induced a significant decrease ($P < 0.05$) in BWG% and liver weight/body weight% as compared to (+ve) group fed on (BD) only.

Our results revealed that the (+ve) group fed on mixed soybean plus casein (1:1 w/w) supplemented with 5% mixed (Car, GC, and OL) recorded a significant $P < 0.05$ decrease in (BWG%) and liver weight / BWG% (LW / BWG%) as compared to the positive control group (+ve) fed only on (BD). While liver weight / BWG% of group fed on mixed soybean plus casein (1:1 w/w) supplemented with 5% mixed (Car, GC, and OL) recorded a significant $P < 0.05$ decrease in liver weight / BWG% (LW / BWG%) as compared to the positive control group (+ve) fed only on (BD), and other treated groups.

Our results agree with **Fouque et al., (1992)**, who demonstrated that not only protein restriction but also modification in the type of protein consumed has favorable effects on renal health because not all proteins are equal in their biological value. Also, soybeans are relatively enriched in isoflavone, rich in phenols, which have been reported to exhibit antioxidant activity.

Effect of soybean diets supplemented with cardamom, green coffee, and olive leaves on protein status of rats suffering from acute renal failure

Table 2 shows the levels of total protein (TP), albumin (AL), and globulin (GL) in the negative control group compared to the positive control group. The negative control group recorded levels of 7.180 ± 0.262 , 3.453 ± 0.081 g/l, and 3.726 ± 0.185 , respectively, whereas the positive control group recorded levels of 5.970 ± 0.210 , 2.116 ± 0.005 , and 3.853 ± 0.215 g/l, respectively. The results reveal a significant decrease ($P\leq 0.05$) in all parameters in the positive control group, which suffered from acute renal failure induced by glycerol injection, compared to the negative control group.

The positive group fed a soybean diet (SD) showed a significant increase in the levels of total protein (TP), albumin (AL), and globulin (GL) compared to the positive control group (+C) fed a basal diet (BD). Additionally, all positive groups fed a BD supplemented with cardamom (Car), green coffee (Gc), and olive leaves (OL) recorded a significant increase in TP, AL, and GL levels compared to the positive control group fed only a BD.

All positive groups fed a soybean diet (SD) supplemented with 5% of cardamom (Car), green coffee (GC), and olive leaves (OL) showed a significant increase in total protein (TP), albumin (AL), and globulin (GL) levels compared to positive groups fed a basal diet (BD) supplemented with Car, GC, and OL. The best improvement was observed in the positive groups fed a mixed diet of soybean and casein (1:1 w/w) supplemented with a 5% mixture of Car, GC, and OL. These groups recorded a significant increase in TP and AL levels compared to the positive groups fed BD, while the globulin level showed no significant difference compared to the negative control group.

The results of the current study revealed that a mixture of soybean diet (SD) and casein (1:1 w/w), both with and without supplementation with cardamom (Car), green coffee (GC), and olive leaves (OL), significantly ameliorated the toxic effects of glycerol in rats with acute renal failure (ARF). Our findings agree with **Stephenson et al. (2005)**, who demonstrated that various phytochemicals in foods, particularly soybeans and soybean

products, have shown significant benefits. In cases of type 1 diabetes, the use of soy protein has been shown to reduce the glomerular filtration rate (GFR) and proteinuria.

Effect of soybean diets supplement with cardamom, green coffee and olive leaves on liver enzymes of rats suffering from acute renal failure

Regarding the effect of soybean diets supplemented with cardamom, green coffee, and olive leaves on liver enzymes in rats suffering from acute renal failure (ARF), the results presented in Table 3 illustrate the levels of aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP). The study revealed that the liver enzyme levels (AST, ALT, and ALP) in the positive control group were significantly higher compared to the negative control group. Specifically, the positive control group recorded levels of 163.506 ± 11.000 for AST, 77.166 ± 4.150 for ALT, and 250.00 ± 4.00 for ALP, while the negative control group recorded levels of 80.840 ± 2.542 for AST, 30.700 ± 3.092 for ALT, and 100.323 ± 5.110 for ALP. The positive control group showed a significant increase ($P < 0.05$) in AST, ALT, and ALP enzymes compared to the negative control group. The results revealed that positive groups fed only a soybean diet (SD) showed a significant decrease in liver enzymes (AST, ALT, and ALP) compared to the positive control group fed only a basal diet (BD). Additionally, positive groups fed a BD supplemented with 5% cardamom (Car), green coffee (GC), or olive leaves (OL) recorded a significant decrease in AST, ALT, and ALP levels compared to the positive control group fed only a BD. Furthermore, positive groups fed an SD supplemented with 5% Car, GC, and OL showed a significant decrease in liver enzyme levels compared to those in the positive control groups fed BD with the same level of supplementation.

The current study shows that the positive group injected with glycerol to induce acute renal failure exhibited decreased levels of total protein and albumin, and increased levels of AST, ALT, and ALP enzymes. This indicates liver cell damage in rats with glycerol-induced acute renal failure. Our results agree with **Kamel et al. (2011)**, who found that a significant decrease in total protein and

albumin, along with increases in ALT, AST, and ALP, indicated liver cell damage in diabetic rats.

The current study revealed that positive groups fed a mixed soybean and casein diet (1:1 w/w) supplemented with a 5% mixture of cardamom (Car), green coffee (GC), and olive leaves (OL) showed the best improvement. These groups had a significant increase in total protein and albumin levels, while also recording a significant decrease in liver enzyme levels (AST, ALT, and ALP). Our results suggest that the synergistic effect observed may be attributed to the wide range of bioactive components present in these combinations.

Stephenson et al. (2005) demonstrated that various phytochemicals derived from foods are available for the treatment of diabetes mellitus. Among these, soybean and soy products have shown a significant impact on patients with chronic kidney disease. In cases of type 1 diabetes, replacing animal protein with soy protein has been found to reduce the glomerular filtration rate and proteinuria. **Zhao and Brinton. (2007)** highlighted that soybeans and soybean products, which are relatively enriched in isoflavones, are of particular interest as a significant dietary protein source. Furthermore, soy is rich in phenols, which have been reported to exhibit antioxidant activity (**Zang et al., 2014**). In the current study, supplementation with cardamom at a 5% level induced a significant improvement. Our results align with those of **Elguindy et al. (2016)**, who reported beneficial effects of cardamom on inflammatory factors such as IL-6, TNF- α , NF-KB, as well as serum AST and ALT levels.

Regarding the potential effects of cardamom, our findings are consistent with those of **El-Segey et al. (2007)**, who showed that cardamom, along with clove, exhibits hepatoprotective effects as evidenced by significantly lower liver enzyme activity.

Our findings align with **Hamzaa and Osman (2012)**, who demonstrated that the combined effects of coffee and cardamom significantly improved liver function by reducing damaging effects. Additionally, our results are consistent with **Higdon and Frei (2006)**, who indicated that elevated serum alanine aminotransferase (ALT) activity is a specific marker of hepatic injury, and consumption of coffee or caffeine can decrease the risk of abnormally elevated ALT levels. Moreover, increasing coffee consumption has been inversely associated with liver enzyme concentrations,

including ALT, aspartate aminotransferase (AST), and gamma-glutamyl transferase (GGT). **Ruhl and Everhart (2005)** highlighted that green coffee contains antioxidant constituents such as chlorogenic acid, ferulic acid, caffeic acid, as well as compounds like caffeine, trigonelline, and phenylalanine, which possess antioxidant properties. Among these antioxidants, green coffee primarily contains isomers of caffeoylquinic acid and caffeic acid (**Henning et al., 2004**).

Regarding the effects of olive leaves, our study's results are consistent with those of **Hamad (2015)**, who concluded that olive leaf extract has a high phenol content and exhibits potent antioxidant activity. Hamad also reported significant effects on liver damage induced by CCl₄ administration, including improvements in serum ALT and AST levels, as well as an increase in serum total antioxidant capacity.

Effect of soybean diets supplement with cardamom, green coffee and olive leaves on antioxidants enzymes of rats suffering from acute renal failure

Table 4 presents the effects of basal diet (BD), soybean diet (SD), a 1:1 mixture of BD and SD supplemented with 5% of cardamom (Car), green coffee (GC), or olive leaves (OL), and a 5% mixture of these supplements on antioxidant enzyme (AOE) levels in rats with acute renal failure. The results showed that the AOE levels in the positive group (+ve) significantly decreased ($P < 0.05$) compared to the negative control group (-ve), which recorded levels of 0.264 ± 0.013 ng/g liver for glutathione peroxidase (GSH-Px), 0.271 ± 0.009 U/g liver for superoxide dismutase (SOD), and 0.194 ± 0.005 mmol/g liver for catalase (CAT). In contrast, the negative control group recorded levels of 0.518 ± 0.011 , 0.407 ± 0.014 , and 0.402 ± 0.014 for GSH-Px, SOD, and CAT, respectively.

Conversely, our results indicated that the positive group (+ve) fed a soybean diet (SD) showed a significant increase ($P < 0.05$) in antioxidant enzyme levels (GSH-Px, SOD, and CAT) compared to the positive group fed a basal diet (BD), which recorded levels of 0.297 ± 0.006 , 0.292 ± 0.006 , and 0.222 ± 0.012 , respectively, for GSH-Px, SOD, and CAT, while the BD-fed group recorded levels of 0.264 ± 0.013 , 0.271 ± 0.009 , and 0.194 ± 0.005 , respectively. Furthermore, all positive groups fed an SD supplemented with 5%

cardamom (Car), green coffee (GC), or olive leaves (OL) showed a significant increase in all antioxidant enzyme levels (GSH-Px, SOD, and CAT) compared to the positive group fed a BD with 5% of the same supplementation (Car, GC, and OL). The greatest improvement in increased levels of antioxidant enzymes was observed in the positive group fed a mixed diet of soybean protein and casein (1:1 w/w) supplemented with a 5% mixture of Car, GC, and OL.

Our findings are consistent with those of **Fouque et al. (1992)**, who emphasized that not all proteins have equal biological value, and highlighted that soybeans are relatively enriched in isoflavones and phenols, which exhibit antioxidant activity. Regarding supplementation with a 5% mixture of cardamom (Car), green coffee (GC), and olive leaves (OL), our results align with **Brewer (2011)**, who noted that cardamom contains significant amounts of phenolic and flavonoid components with potential biological activity. The major constituents of cardamom include α -terpinyl acetate, α -terpineol, 1,8-cineole, and limonene, which are believed to have effects on metabolic syndrome.

Regarding green coffee, our study's results are consistent with those of **Baeza et al. (2014)**, who concluded that green coffee beans contain significant amounts of polyphenolic antioxidants, including chlorogenic, caffeic, ferulic, and n-coumaric acids. The antioxidant capacity of green coffee was observed to be closely associated with the content of chlorogenic acid, making green coffee a natural source of antioxidants.

Olive leaves are a rich source of polyphenolic compounds, particularly oleuropein, known for its antioxidant and antihypertensive properties. Olive leaves contain numerous active constituents with medicinal value, including water-soluble antioxidants and anti-inflammatory properties (**Taamalli et al., 2012**).

Our findings align with **Rafieian-Kopaei (2013)**, who emphasized that oxidative stress plays a significant role in kidney and liver damage by increasing oxidant production when there is a lack of endogenous antioxidant defense. Antioxidants from medicinal plants have been shown to mitigate oxidative-induced kidney and liver damage by reducing lipid peroxidation and enhancing the antioxidant defense system's scavenging capacity.

Conclusion:

"In conclusion, a soybean diet mixed with casein (1:1 w/w) supplemented with a 5% mixture of cardamom, green coffee, and olive leaves produces a significant array of beneficial compounds, particularly polyphenols and flavonoids. These compounds have therapeutic value and can modify the toxic effects of glycerol-induced acute renal failure, improving liver and kidney function. They may also offer protection against liver and kidney toxicity."

Table (1): Biological Effect of soybean diets supplemented with cardamom, green coffee and olive leaves on feed intake, body weight gain % and liver weight / body weight % of rats suffering from acute renal failure.

Groups		Parameters	Feed intake g/day/each rat	Body weight gain %	Liver weight/ body weight%
Control (-ve) fed on basal diet (BD)			18.343 ^a ± 0.478	35.695 ^a ± 3.458	3.005 ^d ± 0.139
Control (+ve) fed on basal diet			17.970 ^a ± 0.374	24.229 ^b ± 1.669	4.335 ^a ± 0.118
Acute renal failure group fed on	Soybean diet (SD)		17.816 ^a ± 0.407	19.872 ^c ± 0.573	3.855 ^b ± 0.105
	BD supplement with 5% cardamom		18.060 ^a ± 0.542	17.130 ^d ± 1.052	3.735 ^b ± 0.175
	SD supplement with 5% cardamom		17.873 ^a ± 0.219	13.995 ^{efg} ± 0.743	3.505 ^c ± 0.128
	Diet supplement with mixed diet casein and soybean protein (1:1) with 5% cardamom		18.00 ^a ± 0.501	15.670 ^{de} ± 0.283	3.411 ^c ± 0.132
	BD supplement with 5% green coffee		17.846 ^a ± 0.736	16.168 ^{de} ± 0.647	3.700 ^b ± 0.122
	SD supplement with 5% green coffee		17.910 ^a ± 0.238	12.552 ^g ± 1.155	3.400 ^c ± 0.170
	Diet supplement with mixed diet casein + soybeans (1:1) with 5% green coffee		18.00 ^a ± 0.260	14.166 ^{efg} ± 0.148	3.210 ^c ± 0.105
	BD supplement with 5% olive leaves		17.880 ^a ± 0.363	16.450 ^d ± 0.609	3.711 ^b ± 0.105
	SD supplement with 5% olive leaves		18.060 ^a ± 0.187	13.288 ^{fg} ± 0.217	3.403 ^c ± 0.113
	Diet supplement with mixed diet casein + soybeans (1:1) with 5% olive leaves		17.940 ^a ± 0.721	14.891 ^{def} ± 0.359	3.300 ^c ± 0.150
	Diet supplement with mixed diet casein + soybeans (1:1) with 5% Mix. of tested materials		17.730 ^a ± 0.238	12.179 ^g ± 0.357	3.100 ^d ± 0.103

LSD: Least significant differences ($P < 0.05$). BD: Basal Diet SD: Soybean Diet
Mean values in each column with same letters are not significantly different.

Table (2): Effect of soybean diets supplemented with cardamom, green coffee and olive leaves on protein status of rats suffering from acute renal failure

Groups	Parameters	Total protein	Albumin	Globulin
		g/l		
Control (-ve) fed on basal diet (BD)		7.180 ^{ab} ± 0.262	3.453 ^a ± 0.081	3.726 ^{bcd} ± 0.185
Control (+ve) fed on basal diet		5.970 ^g ± 0.210	2.116 ^j ± 0.005	3.853 ^{bcd} ± 0.215
Acute renal failure group fed on	Soybean diet (SD)	6.556 ^{ef} ± 6.326	2.466 ⁱ ± 0.076	4.090 ^a ± 0.191
	BD supplement with 5% cardamom	6.326 ^f ± 0.164	2.700 ^h ± 0.050	3.626 ^d ± 0.118
	SD supplement with 5% cardamom	6.796 ^{cde} ± 0.089	2.933 ^{fg} ± 0.056	3.863 ^{bc} ± 0.066
	Diet supplement with mixed diet casein and soybean protein (50%:50%) supplement with 5% cardamom	6.933 ^{bcd} ± 0.076	3.077 ^e ± 0.144	3.855 ^{bcd} ± 0.067
	BD supplement with 5% green coffee	6.760 ^{de} ± 0.045	3.043 ^{ef} ± 0.102	3.716 ^{bcd} ± 0.065
	SD supplement with 5% green coffee	7.040 ^{ab} 0.096	3.210 ^{cd} ± 0.044	3.829 ^{bcd} ± 0.063
	Diet supplement with mixed diet casein plus soybeans (50%:50%) 1:1 supplement with 5% green coffee	7.135 ^{ab} ± 116	3.275 ^{bc} ± 0.056	3.860 ^{bcd} ± 0.060
	BD supplement with 5% olive leaves	6.543 ^{ef} ± 0.125	2.900 ^g ± 0.070	3.643 ^{cd} ± 0.110
	SD supplement with 5% olive leaves	6.966 ^{bcd} ± 0.145	3.113 ^{de} ± 0.059	3.853 ^{bcd} ± 120
	Diet supplement with mixed diet casein + soybeans (50%:50%) supplement with 5% olive leaves	7.117 ^{ab} ± 1.141	3.223 ^{cd} ± 0.074	3.894 ^a ± 0.068
	Diet supplement with mixed diet casein + soybeans (50%:50%) supplement with 5% Mix. of tested materials	7.244 ^a ± 0.086	3.386 ^{ab} ± 0.049	3.858 ^{bcd} ± 0.051

LSD: Least significant differences ($P < 0.05$). BD: Basal Diet SD: Soybean Diet
Mean values in each column with same letters are not significantly different.

Table (3): Effect of soybean diets supplement with cardamom, green coffee and olive leaves on liver enzymes of rats suffering from acute renal failure.

Parameters		AST	ALT	ALP
		u/l		
Control (-ve) fed on basal diet (BD)		80.840 ^{fg} ± 2.542	30.700 ⁱ ± 3.092	100.323 ^k ± 5.110
Control (+ve) fed on basal diet		163.506 ^a ± 11.000	77.166 ^a ± 4.150	250.00 ^a ± 4.00
Acute renal failure group fed on	Soybean diet (SD)	146.186 ^b ± 5.010	69.373 ^b ± 3.137	207.470 ^b ± 6.013
	BD supplement with 5% cardamom	143.376 ^b ± 1.860	69.260 ^b ± 3.137	195.333 ^c ± 6.658
	SD supplement with 5% cardamom	108.826 ^d ± 4.899	58.080 ^{cd} ± 5.856	169.690 ^{de} ± 7.453
	Diet supplement with mixed diet casein and soybean protein (50%:50%) supplement with 5% cardamom	102.190 ^{de} ± 7.419	51.726 ^{def} ± 4.236	160.200 ^{efg} ± 6.050
	BD supplement with 5% green coffee	126.886 ^c ± 3.347	53.370 ^{de} ± 3.769	165.080 ^{ef} ± 4.957
	SD supplement with 5% green coffee	88.260 ^{fg} ± 3.609	44.558 ^{fgh} ± 3.094	151.053 ^{gh} ± 5.961
	Diet supplement with mixed diet casein plus soybeans (50%:50%) 1:1 supplement with 5% green coffee	88.734 ^g ± 4.197	42.109 ^{gh} ± 2.626	139.351 ^{ij} ± 3.688
	BD supplement with 5% olive leaves	132.846 ^c ± 2.719	61.383 ^c ± 1.890	174.840 ^d ± 5.249
	SD supplement with 5% olive leaves	96.689 ^{ef} ± 4.005	48.879 ^{efg} ± 3.117	159.510 ^{fg} ± 6.385
	Diet supplement with mixed diet casein + soybeans (50%:50%) supplement with 5% olive leaves	95.827 ^{ef} ± 4.581	47.492 ^{ef} ± 7.063	147.378 ^{hi} ± 4.436
	Diet supplement with mixed diet casein + soybeans (50%:50%) supplement with 5% Mix. of tested materials	82.746 ^g ± 2.559	38.095 ^h ± 2.548	130.943 ^j ± 3.281

LSD: Least significant differences ($P < 0.05$).

BD: Basal Diet

SD: Soybean Diet

Mean values in each column with same letters are not significantly different.

Table (4): Effect of soybean diets supplement with cardamom, green coffee and olive leaves on antioxidants enzymes of rats suffering from acute renal failure.

Parameters		Glutathione peroxidase (GSH-Px) ng/g Liver	Superoxide dismutase (SOD) U/g liver	Catalase (CAT) mmol/g liver
Groups				
	Control (-ve) fed on basal diet (BD)	0.518 ^a ± 0.011	0.407 ^a ± 0.014	0.402 ^a ± 0.014
	Control (+ve) fed on basal diet	0.264 ^k ± 0.013	0.271 ^f ± 0.009	0.194 ^g ± 0.005
Acute renal failure group fed on	Soybean diet (SD)	0.297 ^j ± 0.006	0.292 ^e ± 0.006	0.222 ^f ± 0.012
	BD supplement with 5% cardamom	0.331 ⁱ ± 0.007	0.325 ^d ± 0.013	0.238 ^e ± 0.007
	SD supplement with 5% cardamom	0.363 ^{f g} ± 0.009	0.344 ^c ± 0.012	0.252 ^{d e} ± 0.007
	Diet supplement with mixed diet casein and soybean protein (50%:50%) supplement with 5% cardamom	0.382 ^{d e} ± 0.009	0.365 ^b ± 0.007	0.263 ^{c d} ± 0.006
	BD supplement with 5% green coffee	0.354 ^{g h} ± 0.007	0.345 ^c ± 0.011	0.249 ^{d e} ± 0.007
	SD supplement with 5% green coffee	0.379 ^e ± 0.013	0.361 ^{b c} ± 0.010	0.271 ^c ± 0.006
	Diet supplement with mixed diet casein plus soybeans (50%:50%) 1:1 supplement with 5% green coffee	0.394 ^{c d} ± 0.004	0.376 ^b ± 0.004	0.272 ^c ± 0.007
	BD supplement with 5% olive leaves	0.340 ^{h i} ± 0.003	0.376 ^b ± 0.005	0.251 ^{d e} ± 0.006
	SD supplement with 5% olive leaves	0.373 ^{e f} ± 0.006	0.360 ^{b c} ± 0.008	0.269 ^c ± 0.008
	Diet supplement with mixed diet casein + soybeans (50%:50%) supplement with 5% olive leaves	0.400 ^c ± 0.007	0.375 ^b ± 0.008	0.279 ^{b c} ± 0.008
	Diet supplement with mixed diet casein + soybeans (50%:50%) supplement with 5% Mix. of tested materials	0.434 ^b ± 0.006	0.394 ^a ± 0.005	0.288 ^b ± 0.138

LSD: Least significant differences ($P < 0.05$).

BD: Basal Diet

SD: Soybean Diet

Mean values in each column with same letters are not significantly different

REFERENCE:

- Athar, M., Zakir, H. Z. S., & Hassan, N. (1997).** Drug metabolizing enzymes in the liver. In S. V. S. Rana & K. Taketa (Eds.), *Liver and environmental xenobiotics* (pp. 65–72). New Delhi: Narosa Publishing House.
- Baeza, G., Amigo-Benavent, M., Sarria, B., Goya, L., Mateos, R., & Bravo, L. (2014).** Hydroxycinnamic acids, but not caffeine, protect human HepG2 cells against oxidative stress. *Food Research International*, 62, 1038–1046.
- Ballabh, B., Chaurasia, O. P., Ahmed, Z., & Singh, S. B. (2008).** Traditional medicinal plants of cold desert Ladakh—used against kidney and urinary disorders. *Journal of Ethnopharmacology*, 118(2-3), 331–339.
- Beauchamp, C., & Fridovich, I. (1971).** Superoxide dismutase: Improved assays and an assay applicable to acrylamide gels. *Analytical Biochemistry*, 44, 276-287.
- Belfield, A., & Goldberg, D. M. (1971).** Normal ranges and diagnostic value of serum 5 nucleotidase and alkaline phosphatase activities in infancy. *Archives of Disease in Childhood*, 46, 842–846.
- Brewer, M. (2011).** Natural antioxidants: Sources, compounds, mechanisms of action, and potential applications. *Comprehensive Reviews in Food Science and Food Safety*, 10, 221–247.
- Das, L., Acharya, A., Berry, D. L., Sen, S., Williams, E., Permaul, E., Sengupta, A., Bhattacharya, S., & Saha, T. (2012).** Antioxidative effects of the spice cardamom against nonmelanoma skin cancer by modulating nuclear factor erythroid-2-related factor 2 and NF- κ B signaling pathways. *British Journal of Nutrition*, 108(6), 984–997.
- Doumas, B. T., & Biggs, H. G. (1971).** Albumin standard and measurement of serum albumin with bromocresol green. *Clinica Chimica Acta*, 31, 78.
- EIAzim, E. A. (2014).** Protective effects of olive leaf extract on methotrexate-induced hepatotoxicity in rats. *Journal of Medicinal Plant Research*, 8(12), 482-487.
- EIAzim, H. (2014).** Protective Effects of Olive Leaf Extract against Methotrexate-Induced Hepatotoxicity. Volume9, Issue6. 3362-3384.

- Elgebaly, H. A., Mosa, N. M., Allach, M., El-Massry, K. F., El-Ghorab, A. H., Al Hroob, A. M., & Mahmoud, A. M. (2018).** Olive oil and leaf extract prevent fluoxetine-induced hepatotoxicity by attenuating oxidative stress, inflammation, and apoptosis. *Biomedicine & Pharmacotherapy*, 98, 446–453.
- Elguindy, N. M., Yacout, G. A., & El Azab, E. F. (2016).** Cardamom (*Elettaria cardamomum*) protects against lipopolysaccharide-induced liver injury in rats. *Comparative Clinical Pathology*, 25(5), 1125-1135.
- El-Segey, O., Ab-Alla, A., & Abu-Al-Nman, S. (2007).** Experimental study of antioxidant and hepatoprotective effects of clove and cardamom in ethanol-induced hepatotoxicity. *Tanta Medical Sciences Journal*, 2(1), 27–36.
- Fouque, D., Laville, M., Boissel, J. P., & Zech, P. Y. (1992).** Controlled low protein diets in chronic renal insufficiency: Meta-analysis. *BMJ (Clinical Research Edition)*, 304(6821), 216-220.
- Gornall, A. C., Bardawill, C. J., & David, M. M. (1949).** Determination of serum protein. *Journal of Biological Chemistry*, 177, 715.
- Hamad, I. (2015).** Antioxidant activity and potential hepatoprotective effect of Saudi olive leaf extract. In *2nd International Conference on Advances in Environment, Agriculture and Medical Sciences (ICAEAM'15)*, June 11–12, 2015.
- Hamzaa, R. G., & Osman, N. N. (2012).** Using coffee and cardamom mixture to ameliorate oxidative stress induced in γ -irradiated rats. *Biochemistry & Analytical Biochemistry*, 1, 113.
- Henning, S. M., Niu, Y., Lee, N. H., Thames, G. D., Minutti, R. R., Wang, H., Go, V. L., & Heber, D. (2004).** Bioavailability and antioxidant activity of tea flavanols after consumption of green tea, black tea, or green tea extract supplement. *American Journal of Clinical Nutrition*, 80(5), 1558–1564.
- Henry, R. J. (1974).** Creatinine measurements with colorimetric method. In *Clinical Chemistry: Principles and Techniques* (2nd ed., p. 525). Harper & Row Publishers.
- Higdon, J. V., & Frei, B. (2006).** Coffee and health: A review of recent human research. *Critical Reviews in Food Science and Nutrition*, 46(2), 101–123.

Jeszka-Skowron, M., Sentkowska, A., Pyrzyńska, K., & Paz De Peña, M. (2016). Chlorogenic acids, caffeine content and antioxidant properties. *European Food Research and Technology*, 242, 1403–1409.

Kamel, Z. H., Daw, I., & Marzouk, M. (2011). Effect of *Cichorium endivia* leaves on some biochemical parameters in streptozotocin-induced diabetic rats. *Australian Journal of Basic and Applied Sciences*, 5, 387–396.

Kandasamy, C. S., Basil, M., Thasnim, P. S. S., Siva, K. R., Gopal, V., & Venkatnarayanan, R. (2010). Hepatoprotective activity of polyherbal formulation containing some indigenous medicinal plants in rats. *Research Journal of Pharmaceutical and Technological*, 3(3), 828–831.

Kandasamy, N. (2010). Hepatoprotective and Antioxidative Activities of Cardamom BENHA VETERINARY MEDICAL JOURNAL, VOL. 29, NO. 2:100-105

Leela, N. K., Prasath, D., & Venugopal, M. N. (2008). Essential oil composition of selected cardamom genotypes at different maturity levels. *Indian Journal of Horticulture*, 65, 366–369.

Ludwig, I. A. (2014). The Importance of Chlorogenic Acid in the Liver Protective Effects

Ludwig, I. A., Clifford, M. N., Lean, M. E. J., Ashihara, H., & Crozier, A. (2014). Coffee: Biochemistry and potential impact on health. *Food & Function*, 5(8), 1695–1717.

of Green Coffe *Journals Foods* Volume 13 Issue 14

Paglia, D. E., & Valentine, W. N. (1967). Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. *Journal of Laboratory and Clinical Medicine*, 70(1), 158-169.

Pereira, A. P., Ferreira, I. C., Marcelino, F., Valentão, P., Andrade, P. B., Seabra, R., Estevinho, L., Bento, A., & Pereira, J. A. (2007). Phenolic compounds and antimicrobial activity of olive (*Olea europaea* L. Cv. Cobrancosa) leaves. *Molecules*, 12(5), 1153–1162.

Ponte, S. (2002). The ‘latte revolution’? Regulation, markets and consumption in the global coffee chain. *World Development*, 30(7), 1099–1122.

Poole, R., Kennedy, O. J., Roderick, P., Fallowfield, J. A., Hayes, P. C., & Parkes, J. (2017). Coffee consumption and health: Umbrella review of meta-analyses of multiple health outcomes. *BMJ*, 359, j5024.

Rafieian-Kopaie, M. (2013). Medicinal plants for renal injury prevention. *Journal of Renal Injury Prevention*, 2(2), 63–65.

Ruhl, C. E., & Everhart, J. E. (2005). Coffee and caffeine consumption reduce the risk of elevated serum alanine aminotransferase activity in the United States. *Gastroenterology*, 128(1), 24–32.

SAS Institute Inc. (2004). *Statistical Analysis System, SAS Users Guide: Statistics*. Cary, NC: SAS Institute Inc.

Sharma, V., & Singh, M. (2014). Attenuation of N-nitrosodimethylamine-induced hepatotoxicity by *Operculina turpethum* in Swiss albino mice. *Iranian Journal of Basic Medical Sciences*, 17, 73–80.

Steinmetz, K. A., & Potter, J. D. (1996). Vegetables, fruit and cancer prevention: A review. *Journal of the American Dietetic Association*, 96(10), 1027–1039.

Stephenson, T. J., Setchell, K. D., Kendall, C. W., Jenkins, D. J., Anderson, J. W., & Fanti, P. (2005). Effect of soy protein-rich diet on renal function in young adults with insulin-dependent diabetes mellitus. *Clinical Nephrology*, 64(1), 1–11.

Taamalli, D. A., Román, E. B., Catalán, V. R., Torres, A. P., Sánchez, M., Herrero, E., Ibañez, E., Micol, V., Zarrouk, A. S., Carretero, A. F., & Gutiérrez, A. F. (2012). Preparation of polyphenol fine particles: Potent antioxidants by a supercritical antisolvent process using different extracts of *Olea europaea* leaves. *Food and Chemical Toxicology*, 50, 1817.

Zang, Y., Igarashi, K., & Yu, C. (2014). Anti-obese and anti-diabetic effects of a mixture of daidzin and glycitin on C57BL/6J mice fed with a high-fat diet. *Bioscience, Biotechnology, and Biochemistry*, 11, 1–7.

Zhao, L., & Brinton, R. D. (2007). WHI and WHIMS follow-up and human studies of soy isoflavones on cognition. *Expert Review of Neurotherapeutics*, 7, 1549–1564.

المستخلص العربي

تأثير وجبات الصويا المدعمة بالهيل ، القهوة الخضراء واوراق الزيتون على وظائف الكبد والانزيمات المضادة للاكسدة في الفئران المصابة بالفشل الكلوي الحاد

استهدفت هذه الدراسة معرفة تأثير وجبات فول الصويا وخليط من الصويا والكازين (١:١) المدعمة بنسبة ٥٪ من الهيل ، القهوة الخضراء واوراق الزيتون منفردة او ٥٪ خليط منها على المأخوذ اليومي من الطعام ، النسبة المئوية لمعدل الزيادة في وزن الجسم ونسبة وزن الكبد بالنسبة لوزن الجسم . ومعرفة تأثير ذلك على مستوى البروتين الكلى في سيرم الدم ، الاليومين ، الجلوبيولين انزيمات الكبد ، الانزيمات المضادة للاكسدة . وذلك في الفئران المحدث لها فشل كلوي حاد ومن اجل ذلك تم استخدام عدد ٧٨ فأر بالغ من الالينو من فصيلة اسبراجو دولي اوزانهم 200 ± 10 جم . تم تقسيم الفئران الى مجموعتين رئيسية المجموعة الرئيسية الأولى (٦ فئران) تم تغذيتها على الغذاء الأساس واستخدمت كمجموعة ضابطة سالبة . المجموعة الرئيسية الثانية (٧٢ فأر) ثم حقنها بالجليسرول لاحداث فشل كلوي حاد للفئران ثم تم تقسيمها الى (١٢) مجموعة تم استخدام (٦ فئران) كمجموعة ضابطة إيجابية تم تغذيتها على الغذاء الاساسي أما بالنسبة (١١ مجموعة) المصابة الأخرى فقد تم تغذيتها أما على غذاء الصويا او خليط الصويا والكازين (١:١) مدعما بنسبة ٥٪ إما من الهيل ، القهوة الخضراء او أوراق الزيتون او ٥٪ خليط (الهيل ، القهوة الخضراء واوراق الزيتون) وذلك لمدة ٨ أسابيع . وفي نهاية التجربة تم ذبح الفئران بعد صيام طوال الليل تم فصل الكبد وتم اجراء الوزن كما تم تجميع عينات الدم ثم فصل السيرم كما تم تقدير مستوى كلا من البروتين الكلى ، الاليومين ، الجلوبيولين انزيمات الكبد **AST, ALT, and ALP** ، وكذلك الانزيمات المضادة للاكسدة **GSH-PX (SOD & CAT)**. وقد أوضحت النتائج التي تم الحصول عليها ان الفئران المحدث لها إصابة بالفشل الكلوي الحاد عن طريق الحقن بالجليسرول . والتي تغذت على غذاء يحتوي على خليط فول الصويا والكازين بنسبة (١:١) والمدعم بنسبة ٥٪ من خليط (الهيل ، القهوة الخضراء واوراق الزيتون) قد أدت هذه التغذية الى عدم حدوث فروق معنوية في معدل المأخوذ من الطعام وذلك مقارنة بالكنترول الموجبة المغذاة على الغذاء الاساسي كذلك أدت الى انخفاض معنوي لوزن الجسم كذلك بالنسبة لوزن الكبد بالنسبة لوزن الكبد بالنسبة لوزن الجسم الكلى وذلك بالمقارنة بالكنترول الموجب المصاب المغذاة على الغذاء الاساسي بدون تدعم كذلك أظهرت التحاليل البيوكيميائية لسيرم الدم ان مجموعة الفئران الموجبة المصابة بالفشل الكلوي الحاد قد حدث لها انخفاض معنوي في مستوى كل عن البروتين الكلى ، الاليومين ، الانزيمات المضادة للاكسدة بينما حدث لها زيادة معنوي في مستوى انزيمات الكبد (**AST,ALT&ALP**) وذلك مقارنة

بمجموعة فئران الكنترول السالب (الغير مصابة) المغذاة على الغذاء الرئيسي (BD)، كذلك أظهرت نتائج هذه الدراسة ان مجموعات فئران التجارب المصابة بالفشل الكلوي الحاد والتي تم تغذيتها على فول الصويا بمفرده او الكازين المدعم بنسبة ٥٪ من (الهيل، القهوة الخضراء او الهيل وخليط منهم) كذلك المجموعة التي تغذت على خليط الصويا مع الكازين (١:١) والمدعم بنسبة ٥٪ من خليط (الهيل ، القهوة الخضراء و الصويا) حدوث تحسن معنوي أدى الى ارتفاع مستوى البروتين الكلى والاليومين كذلك ارتفاع مستوى الانزيمات المضادة للاكسدة (GSH – PX ,SOD,CAT) بينما أدى الى انخفاض مستوى انزيمات الكبد (AST, ALT &ALP). وتظهر نتائج هذه الدراسة ان خليط بروتين الصويا مع الكازين بنسبة (١:١) والتدعيم بنسبة ٥٪ بخليط (الهيل ، القهوة الخضراء و أوراق الزيتون) هذه التوليفة من الممكن ان تقلل التأثير الضار لاصابة الكبد والكلى لذا فان هذه التوليفة من الممكن ان تكون مفيدة كغذاء وقائي مساعد ضد سمية الكبد والكلى كذلك كغذاء علاجي للمرض المصابين بامراض الكبد والكلى.