SVU- International Journal of Veterinary Sciences, 4 (4): 56-67, 2021.
Print ISSN: 2535-1826 **Online ISSN: 2535-1877**

Research Article Open Access

Harmful effects of Mocha (Cappucinno) as instant coffee drinks on kidney and heart of Wister albino rats

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Abstract

To date, coffee drinks lies as the second largest trade widely consumed in a major variety of formats attributed to its easy preparation and desirable taste. There were growing concerns detected the correlation between coffee beverage consumption and variant clinical manifestations on the drinkers. The based study was designed to elucidate the potential aggravating influences derived from instant coffee drinks. Forty adult male albino rats within average age 3 months and body weight 120-150 gm divided into 4 groups. Group (1); used as control. While, group 2, 3 & 4 orally administrated coffee of Mocha (Cappucinno) type at 0.726, 1.44 and 2.16 ml, respectively daily for 7 weeks. After the experiment time out, blood samples were withdrawn for evaluation of kidney function tests and lipid profiles. In Addition, kidney and heart tissues collected for histopathology. From a biochemical point of view, Cappucinno was resulted in a significant increase in urea, creatinine and uric acid besides significant elevation in blood glucose, cholesterol and triglycerides levels when compared with control. Histopathologically, necrosis with inflammation of the renal tubules and myocardium were detected, in addition to congestion with dilatation of the blood vessels. It could be concluded that consumption of Cappuccino as instant coffee drinks conducted deleterious effect on the drinkers.

Keywords: Cappucinno, harms, heart, histopathology, kidneys.

DOI: 10.21608/svu.2021.87388.1135 Received: July 25, 2021 Accepted: November 1, 2021 Published: December 9, 2021 *Corresponding Authors: Zeinab Al-Amgad E-mail: zizi_1283@yahoo.com

Citation: Al-Amgad and Mahdy, Harmful effects of Mocha (Cappucinno) as instant coffee drinks on kidney and heart of Wister albino rats. SVU-IJVS 2021, 4 (4): 56-67.

Competing interest: The authors have declared that no competing interest exists.

Introduction

According to a survey, coffee constitutes the most popular worldwide among beverages (Frary et al., 2005). To the best of our knowledge, ingestion of coffee is popularly regarded as beneficial to variant health issues. Coffee is considered the distinct source of caffeine globally consumed. The amount of caffeine consumed is the stalk size, product type and production ways dependent on (Elmaadawy et al., 2015). However, several epidemiological studies implied coffee made consumers more prone to chronic hazards.

Kidneys and heart are assumed as two important organs in body; since it was functioned together for a keep body healthy. As when one influenced, the other was too. Heart affected the health of kidneys; on the contrary, kidneys also affect the health of heart. Furthermore, heart failure is highly associated with in patients undergone chronic kidney disease (Segall et al., 2014).

Prolonged caffeine consumption develops diverse physiological consequences; prominent kidney diseases are attributed. Caffeine act as a risk factor in the exacerbation of cysts in people suffered from autosomic dominant polycystic kidney (Belibi et al., 2004).

Furthermore, some researchers have provoked the close relationship between coffee drinks and cardiovascular issues. Coffee consumption turned out to be a significant indicator for heavy consumption suffered hypertension (Palatini et al., 2016). Coffee consumption for more than one time daily is consequences with an increase in the blood pressure attributed to enclosed bioactive compounds. Guidelines instructed a relationship between heavy coffee drinkers and detrimental effects on total cholesterol, total triglyceride, and low-density lipoprotein (LDL) cholesterol (Jee et al., 2001). Chown et al. (2001) and Keijzers et al. (2002) revealed that the consumption of a higher amount of coffee than usual resulted in block in the glucose tolerance. Also, it has propounded that high coffee consumption induced acute myocardial infarction, and stroke (Mostofsky et al., 2010). Moreover, many previous reviews investigated the aggravated risks; fibrosis/cirrhosis of the tissues concerning the daily consumption of more than three cups of instant coffee drinks (Leung et al., 2011).

The current work was designed to attract an attention to considerable influence of consumption of cappuccino drink on kidney, heart, blood glucose levels and lipid profiles in albino Wister rats.

Material and Methods

Materials:

1. Experimental animals:

The current study was carried at Department of Nutrition and Food Science, Faculty of Specific Education. South Valley University. Forty adult male albino rats with an average age of 3 months and body weight of 120-150 gm were used in the present study. The rats were obtained from Laboratory Animal House belonging to Giza Governorate, Egypt. The rats were well examined immediately upon their arrival to the laboratory. The animals were maintained under good ventilation and standard conditions (temperature of 23±3° C, relative humidity of 60-70%, and a 12-hour light/ dark cycle). Adequate commercial pellets ad libitum and water were supplied. Histopathological analysis was done at Laboratory of Pathology and Clinical Pathology, Faculty of Veterinary Medicine, South Valley University.

2. Instant coffee drink:

The Cappucinno beverage was purchased from a local market belonging to Qena governorate, Egypt. It was subjected to chemical analysis as the following composition shown in Table 1.

Table 1. The composition of acrylamide, caffeine, cholorogenic acid and caffeic acid presented in Mocha (Cappucinno) coffee using GC/MS-MS analysis.

Parameters	Mocha (Cappuccino)	LSD at 0.05
Acrylamide (ppm)	69.3± 1.07	3.409
Caffeine (mg/g)	48.78± 1.10	2.258
Chlorogenic acid (µg/g sample)	273.70	167.343
Caffeic acid (mg/g sample)	660.42± 24.16	80.700

Methods:

1- Experimental design:

Forty adult male Albino rats within 3 months-old average age and body weight 120-150 gm obtained from Laboratory Animal House belonging to Giza, Egypt. All animals were allowed to acclimatize inside a well-ventilated room and fed on standard diet during the experiment period. The rats were randomly classified into 4 groups, each one containing ten (10 rats for each) adult albino rats as following:

Group1: It served as control; it was supplied standard diet and distilled water for 7 weeks.

Mocha treated groups (Mo. 1, 2 & 3) were orally exposed to different doses of Cappucinno beverage according to Lestari et al. (2017) as follow:

Mocha 1: The rats orally administrated Mocha type (**Mo. 1**) at dose 0.72 ml daily for 7 weeks,

Mocha 2: The rats orally received Mocha type (**Mo. 2**) at dose 1.44 ml daily for 7 weeks, and

Mocha 3: The rats orally ingested Mocha type (**Mo. 3**) at dose 2.16 ml daily for 7 weeks.

During the experiments, all animals were daily examined for any clinical signs or mortalities.

2- Sampling:

2.a. Blood samples:

After the experiment run out, all animals were sacrificed for blood samples withdraw under general anesthesia for biochemical examination. The blood samples let go at room temperature to clot; the serum was segregated at 3000 rpm for 10 minutes using centrifuge. The resultant sera were collected in clean Epindorf's tubes and preserved at -20° C until biochemical assay according to Schermer (1967).

2.b. Organs collection:

Kidney and heart from all existing groups were incised and examined for any post-mortem changes. Representative samples were taken then fixed in 10% neutral buffered formalin for histopathological examinations.

2.c. Biochemical analysis:

Biochemical analysis had been estimated by spectrophotometer using standard test kits.

2.c.1. Assessment of kidney function tests in serum:

Enzymatic colorimetric method used for measurement of urea, creatinine and uric acid levels in blood serum by Bio-diagnostic kits (Dokki, Giza, Egypt) described by Fawcett and Scott (1960), Bartles et al. (1972), and Barham and Trinder (1972), respectively.

2.c.2. Assessment of glucose and lipogram in serum:

Glucose determination in serum was assessed according to Trinder (1969). Moreover, cholesterol and triglyceride levels

were calorimetrically determined according to Richmond (1973) and Fassati and Prencipe (1982), respectively.

3- Histopathological examination:

Kidney and heart specimens were dissected and fixed in 10% neutral buffered formalin, followed by dehydration in ascending grades of alcohol (70, 80, 90 & 100), cleared in xylene and embedded in paraffin wax according to Bacha and Bacha (2000). Sections about 4- 5µm thickness were prepared for Harries hematoxylin and eosin (H & E.) stain for the microscopical screening as described by Larson et al. (2011).

4 - Statistical analysis:

The results were statistically analyzed used one-way analysis of variance (ANOVA) using SPSS (Statistical Package for Social Sciences) according to Borenstein et al. (1997). The data were in form of Mean ± Standard Deviation. The resultant values were significantly different when P <0.05 in comparison with control.

Results:

1. Biochemical analysis:

1.a- Effect of Mocha on serum urea level (mg/dl):

Results recorded in Table 2, Fig. 1 revealed non-significant changes in urea level between Mo. (1) & (2) groups comparison with control negative. Mo. (3) group showed significant increase (P<0.05) in urea values when compared with control group.

1.b- Effect Mocha on serum creatinine level (mg/dl):

Level of creatinine was significantly increased (P<0.05) in Mo. (3) when compared with control. However, other groups exhibited non-significant difference when compared with control group as shown in Table 2, and Fig. 2.

1.c- Effect of Mocha on serum uric acid level (mg/dl):

The obtained data displayed that uric acid level recorded significant increase (P<0.05) in Mo. (1), (2) & (3) groups that detected when compared with control group as shown in Table 2, and Fig. 3.

Table 2. The effect of instant coffee drinks on kidney function tests including urea (mg/dl), creatinine (mg/dl), and uric acid (mg/dl) of albino rats of control group and Mocha treated groups. (Mean± SD)

Parameters	Urea (mg/dl)	Creatinine (mg/dl)	Uric Acid (mg/dl)
Control	41.76±0.21^{b}	0.81±0.04^{b}	2.1±0.07^{b}
Mo. (1)	41.4±0.42^{b}	0.84±0.01^{b}	3.2±0.04^{a}
Mo. (2)	45.8±0.3^{b}	0.90±0.03^{b}	4.2±0.03^{a}
Mo. (3)	50.7±0.64^{a}	2.0±0.01^{a}	6.1±0.23^{a}

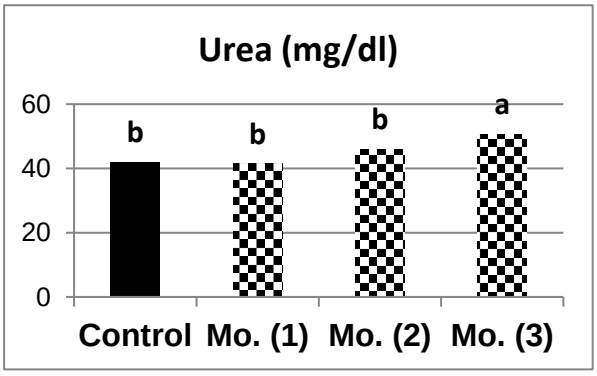

Fig. 1. The effect of instant coffee drinks on serum urea (mg/dl) of albino rats of control group and Mocha treated groups.

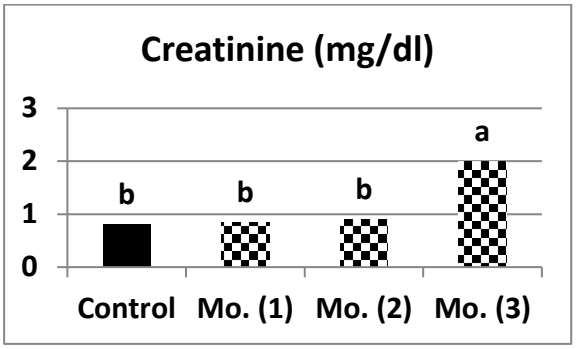

Fig. 2. The effect of instant coffee drinks on serum creatinine (mg/dl) of albino rats of control group and Mocha treated groups.

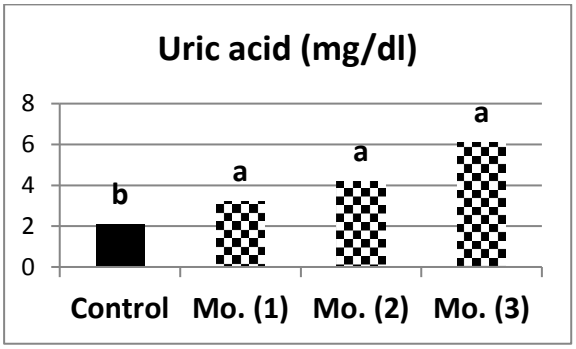

Fig. 3. The effect of instant coffee drinks on serum uric acid (mg/dl) of albino rats of control group and Mocha treated groups.

1.d- Effect of Mocha on serum glucose level:

Data of glucose level which shown in Table 3 and Fig. 4 exhibited significant increase ($P<0.05$) in Mo. (1), (2) & (3) groups when compared with control group.

2. Effect of Mocha on lipid profile:

2.a- Effect of Mocha on serum total cholesterol level (mg/dl):

Total cholesterol level detected higher mean level ($P<0.05$) among Mo. (2) & (3) groups as compared with control one. Whilst, non-significant differences were observed in Mo. (1) group in comparison with control group as shown in Table 3 and Fig. 5.

2.b- Effect of Mocha on serum triglyceride level (mg/dl):

Triglyceride values as in Table 3 and Fig. 6 recorded a noticeable elevation ($P<0.05$) in Mo. (2) & (3) groups in comparison with control.

Table 3. The effect of instant coffee drinks on serum glucose level (mg/dl) and lipid profile including Total cholesterol (TC) (mg/dl), Triglyceride (TG) (mg/dl) of albino rats of control group and Mocha treated groups. (Mean± SD).

Parameters	Glucose (mg/dl)	TC (mg/dl)	TG (mg/dl)
Control	123.4±2.07^{b}	109.3±3.1^{b}	144.1±0.97^{b}
Mo. (1)	128.7±0.7^{b}	115.6±1.14^{b}	147.8±1.64^{b}
Mo. (2)	139.0±1.05^{a}	136.8±0.83^{a}	160.4±1.48^{a}
Mo. (3)	152.3±0.95^{a}	152.6±0.83^{a}	174.2±1.92^{a}

Means in the same column with different letters are significantly different when $P<0.05$.

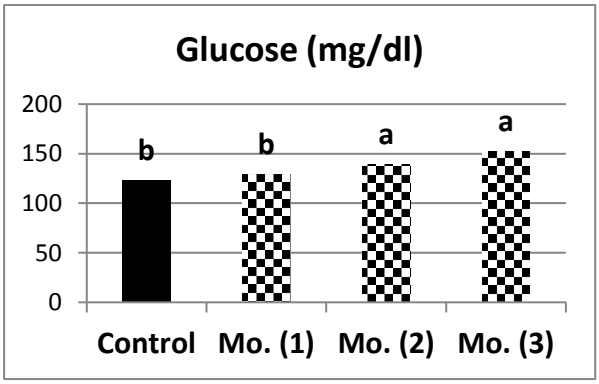

Fig. 4. The effect of instant coffee drinks on serum glucose (mg/dl) of albino rats of control group and Mocha treated groups.

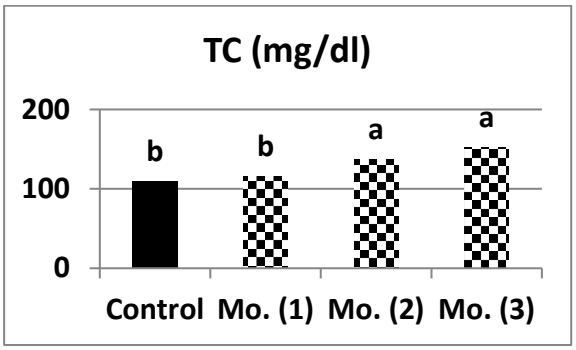

Fig. 5. The effect of instant coffee drinks on serum total cholesterol (mg/dl) of albino rats of control group and Mocha treated groups.

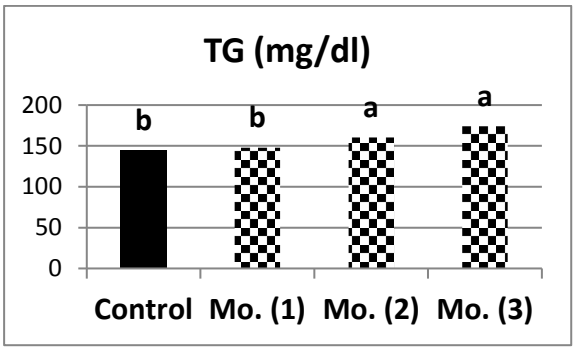

Fig. 6. The effect of instant coffee drinks on serum total triglycerides (mg/dl) of albino rats of control group and Mocha treated groups.

3. Histopathological results:

3.a- Macroscopic appearance:

Kidney of the control group displayed normal appearance within normal sizes and color. While, Mo. treated groups showed severe congestion with dark color of the kidney.

Heart of the control group appeared normally sizes and shapes. However, Mo. treated groups of exhibited congestion in the blood vessels.

3.b- Microscopic results:
Kidney:

As expressed in Table 4, Figs. 7 & 8; kidney of the control group showed normal histological structure. Since, it consisted of glomerulus occupied renal corpuscle, convoluted tubules, and collecting ducts (Fig. 7 a-b). Kidney of Mo. (1) detected interstitial cells infiltration, precipitation of protein casts inside the tubules in addition congestion in the blood vessels (Fig. 8 a), also extensive fibrosis with fibrous tissues infiltration and focal inflammatory cells infiltration was detected (Fig. 8 b). Kidney of Mo. (2) displayed severe fibrous tissues infiltration with interstitial cells infiltration mainly lymphocytes (Fig. 8 c). In addition, severe congestion of the renal blood vessels with cystic dilatation of the renal tubules was implied (Fig. 8 d). As for kidney of Mo. (3) suffered necrobiotic changes resulted in sloughing of the epithelial lining tubules, as well congestion and dilatation of the blood vessels (Fig. 8 e), aggressive degree of the renal fibrosis with fibrous tissues infiltration (Fig. 8 f).

Table 4. Histopathological scores of kidney and heart of control and Mocha exposed groups were divided into absent, (-), mild (+), moderate (++), and severe (+++) according to severity of lesions.

Lesions/Group		Control	Mo. (1)	Mo.(2)	Mo.(3)
Renal necrosis	**Kidney**	-	+++	+++	+++
Renal fibrosis		-	++	+++	++
Interstitial cells infiltration		-	+++	++	+++
Casts precipitation		-	+++	+++	+++
Congestion and dilatation of the blood vessels		-	++	+++	+++
Myocardium necrosis	**Heart**	-	++	++	+++
Cytoplasmic vacuolation		-	++	++	+++
Interstitial cells infiltration		-	++	++	+++
Congestion and dilatation of the blood vessels		+	+++	+++	+++

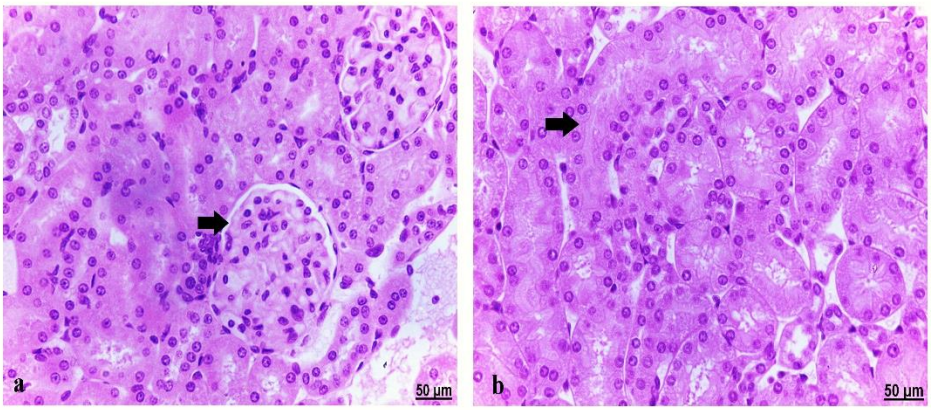

Fig. 7. Photomicrograph of kidney of the control group showing normal histological structure. It is composed of normal renal corpuscle comprising glomerulus within Bowman's capsule **(a)**, as well proximal and distal convoluted tubules **(b).** (H& E., bar= 50µm)

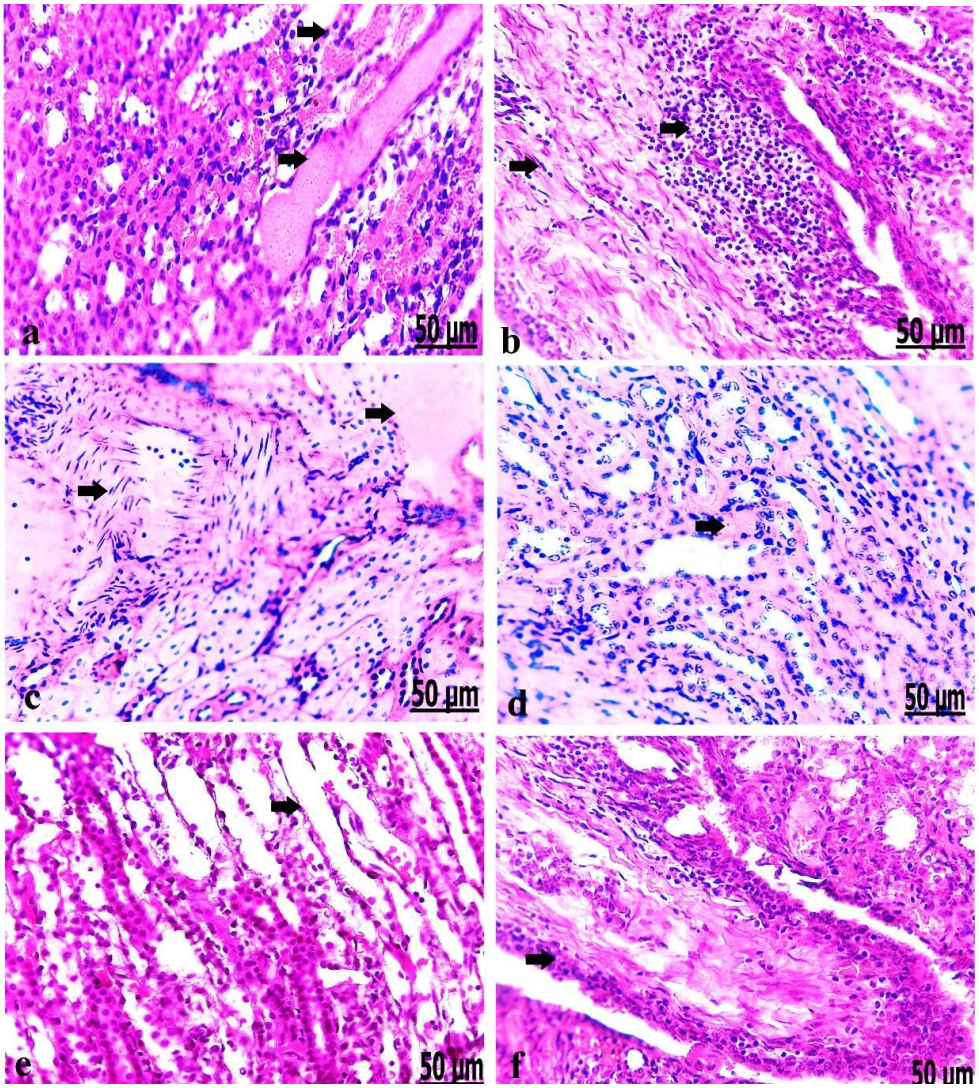

Fig. 8 (a-f): Photomicrograph of kidney treated groups. Kidney of **Mo. (1)** showing interstitial cells infiltration, proteinous cast precipitation inside the tubules in addition congestion in the blood vessels of the renal tubules **(a)**, extensive fibrosis with fibrous tissues infiltration and focal inflammatory cells infiltration **(b)**. Kidney of **Mo. (2)** showing severe fibrous tissues infiltration with interstitial cells infiltration mainly lymphocytes **(c)**, severe congestion of the renal blood vessels with cystic dilatation of the renal tubules **(d)**. Kidney of **Mo. (3)** showing necrosis and sloughing of the epithelial lining tubules with congestion and dilatation of the blood vessels **(e)**, high degree of the renal fibrosis with fibrous tissues infiltration **(f)**. (H& E., bar= 50µm)

Heart:

As shown in Table 4, Figs. 9 & 10; heart of the control group showed normal histological structure of myocardium with normal striation (Fig. 9 a-b). However, heart of Mo. (1) group showed necrosis of the muscle fibers in addition congestion in the blood vessels (Fig. 10 a), furthermore cytoplasmic vacuolation and focal inflammatory cells infiltration were elucidated (Fig. 10 b). Heart of Mo. (2) detected interstitial cells infiltration mainly lymphocytes (Fig. 10 c), necrosis of the muscle's fibers (Fig. 10 d). Heart of Mo. (3) displayed severe congestion and dilatation in the blood vessels with thickening of the wall (Fig. 10 e), extensive necrosis and destruction of the muscle fibers (Fig. 10 f).

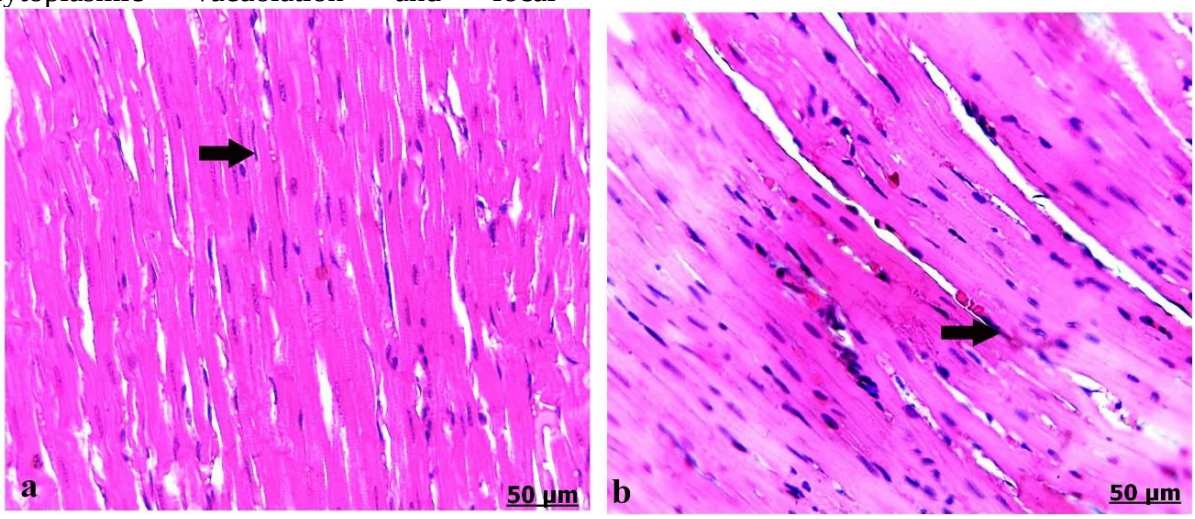

Fig. 9. Photomicrograph of heart of the control group showing normal histological structure of heart comprising normal muscle fibers striation. (H& E., bar= 50µm).

Discussion

The present work was conducted the deteriorating impacts derived from coffee consumption with reasonable evidence on renal and cardiovascular issues. Chronic consumption of coffee beverages (cappuccino) induced significant increase in urea, creatinine and uric acid growing by increase the concentration. Coffee constituents could disturb the excretory function of kidney resulted in detrimental renal effect in the body. Announced investigators have recorded that heavy exposure to sugar sweetened beverages (SSBs) in coffee was correlated with higher uric acid in the serum (Shih et al., 2019).

Adolescents consuming high amounts of SSBs or coffee tend to high values of serum uric acid correlated to higher component of fructose. Moreover, acrylamide (ACR) in coffee exactly implied renal dysfunction associated with impairing the secretory mechanism with a rise in serum uric acid levels at low doses. Whilst in higher doses, ACR may inhibit urate absorption producing a urate diuresis (Khalil, 2005). Kidney possesses an efficient binding capacity with ACR three times more significant in conjugating ACR than brain (Alturfan et al., 2011). Nicely, presence of ACR in kidney was capable of increase metabolism of glycidamide by the CYP450 pathway.

A significant increase in blood glucose concentration after cappuccino ingestion as previously described in our results. Coffee consumption hinders glucose regulation with resultant hyper insulinemia (Bidel et al., 2006). Furthermore, a significant increase in the mean glucose level interpreted with glucose homeostasis alterations through diminish in glucose uptake into skeletal muscle, thereby

producing elevations in the blood glucose level attributed to the caffeine (Ihim et al., 2019).

An increase in total cholesterol level was detected after coffee administration. Higdon and Frei (2006) exhibited that ill-deficient coffee filtration is a significant source of diterpenes involving cafestol and kahweol which critical to cholesterol rise. Also, coffee was immediately induced elevated total cholesterol level (Cai et al., 2012).

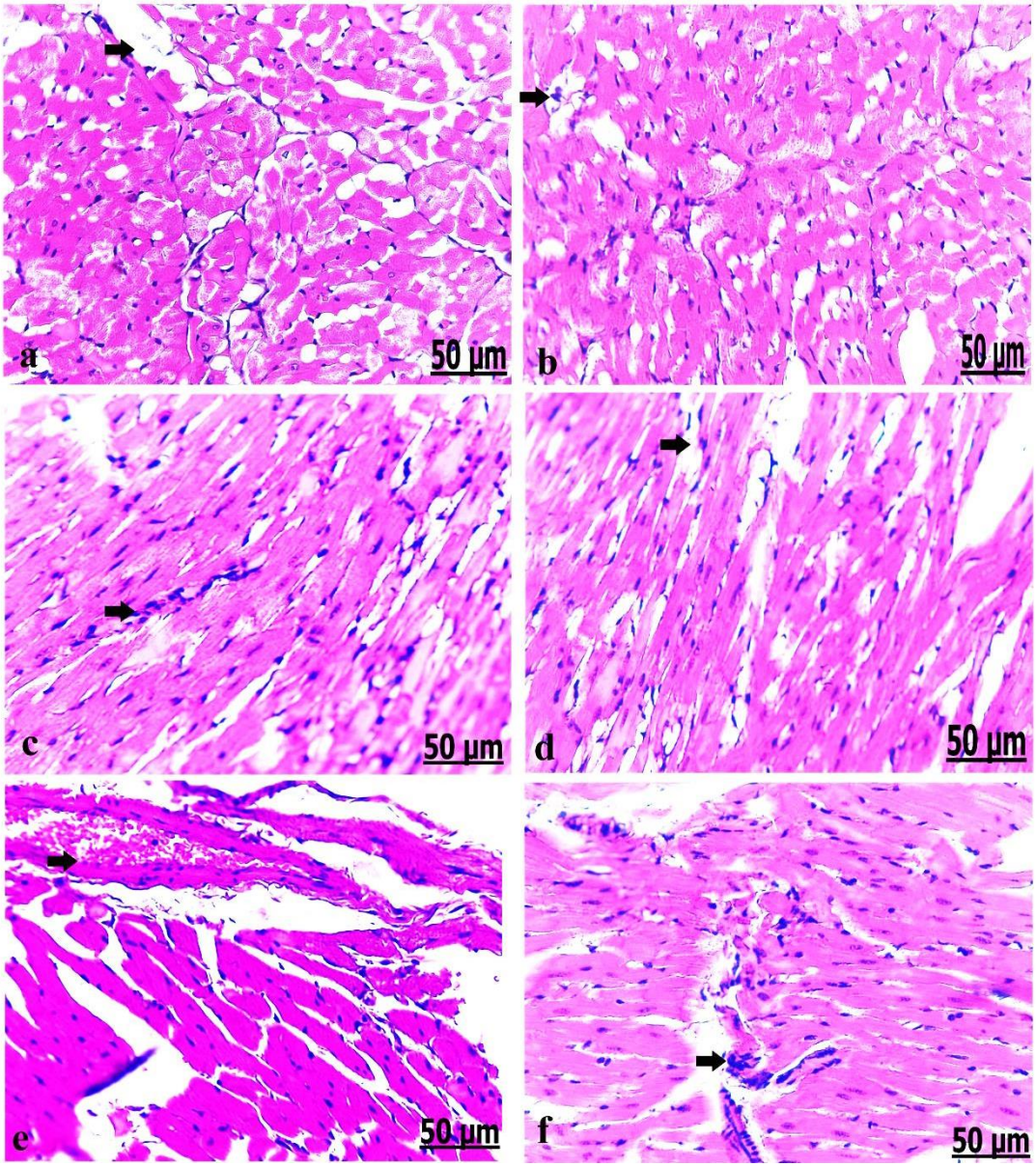

Fig. 10. Photomicrograph of heart of **Mo** treated groups. Heart of **Mo (1)** showing necrosis of the muscle fibers in addition congestion in the blood vessels **(a),** with cytoplasmic vacuolation and focal inflammatory cells infiltration **(b)**. Heart of **Mo (2)** showing interstitial cells infiltration mainly lymphocytes **(c)**, necrosis of the muscle fibers **(d)**. Heart of **Mo (3)** showing severe congestion and dilatation in the blood vessels with thickening of the wall **(e)**, extensive necrosis and destruction of the muscle fibers **(f)**. (H& E., bar= 50µm).

Concerning histopathological findings, necrobiotic changes with inflammatory focused areas in kidney and heart were progressed, moreover, congestion and

dilatation of the blood vessels. The higher administration of coffee provoked damaged kidney (Islam et al., 2016). Additionally, quality of coffee affected by temperature; since roasted Liberica coffee caused renal histological changes involving cytoplasmic degenerative changes, renal swelling and tubular necrosis (Widodo et al., 2019).

Caffeine occupied the major compound in coffee; caffeine exerts some deviating efforts on the cardio-vasculature (Papamichael et al., 2005)**.** Hanna and Abd Elmonem (2014) discussed that the consumption of instant coffee possesses non-dairy creamer elucidate a crucial potential impact on the heart. Propounded histological disruptions related to greedily coffee consumers comprise myocardial infarction and inflammation **(**Mostofsky et al., 2010).

Acknowledgment

Authors are thankful for Department of Pathology and Clinical Pathology, Faculty of Veterinary Medicine, South Valley University that allow performing of histopathological examination in the Department laboratory.

Conflicts of interest

The authors declare that there is no conflict of interest.

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