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How creamy coffee could potentially take a part in public health risks particularly in liver?

Zeinab Al-Amgad [1*] and Maha Mahdy [2]

[1]General Authority for Veterinary Services, Qena Veterinary Directorate, Qena, Egypt, [2]Home Economics Department, Nutrition and Food Science, Faculty of Specific Education, South Valley University, Qena, Egypt.

Abstract

Interestingly, coffee is accentuated as a widespread stimulant beverage. Rising concern inferred an approach between coffee imbibe and an array of public health conditions. The existing study was predicated to clarify the crucial influences of the prolonger term intake of coffee drinks. Forty adult male albino rats were divided into four groups of ten rats each. First group employed as control. However, 2nd, 3rd and 4th groups were orally exposed to ascending concentrations of Mocha (Cappucinno) at 0.72, 1.44 and 2.16 ml, respectively daily for 7 weeks. Blood samples were dragged for an assessment liver functions, level of carcinoembryonic antigen (CEA) and mean of total testosterone hormone (T.T.) using standard kits methods. Furthermore, liver biopsies were taken for the histological screening. From biochemical aspect, serum levels of aspartate aminotranferase (AST), alanine aminotranferase (ALT), and alkaline phosphatase (ALP) were significantly exceeded the mean levels in a comparison with control, besides level of T.T. was remarkably decreased as when compared with control. By cross sectional screening, liver was frankly distinguished by degenerative disorders with cytoplasmic vacuolation as well congested blood vessels. Based on these findings, it could be concluded that cappuccino declaredly implying a detrimental effect on the consumers was evidently boosted by the time and concentration.

Keywords:

Coffee, Histopathology, Liver enzymes, Public health.

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*Corresponding Author: Zeinab Al-Amgad E-mail: zizi_1283@yahoo.com

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Introduction

Instant coffee drinks comprising a class of non-natural manufactured additives like vegetables oils, stabilizers, and chemical sweeteners termed non-dairy creamers. Surrounding of coffee with such additives was believed to provide a bright and lightening appearance without diminishes from its usefulness. Unfortunately, an existence of non-dairy creamer in coffee brought harms and interferes with absorption of others beneficial compounds (Cutler, 2013).

The concept of liver is generally regarded as targeted locale for the xenobiotic biotransformation where well plays a considerable part for detoxification in body (Kamińsk and Wiaderkiewicz, 2007). An ingestion of coffee drinks unfavorably influenced the health status of the consumer (Jee et al., 2001). It is probably formed as a risk factor for chronic diseases (Palatini et al., 2016). Considering similarly effects, a series of arbitrators have attracted the attention to the relevance of coffee consumption and liver tendency. A provoke of consumption of 2 g coffee per day for a month had significantly dropped the activities of liver enzymes and bilirubin**,** worsens preexisting risk factors (Onuegbu et al., 2011).

According to scientific studies in respect to coffee, where declared that coffee play a crucial role in carcinogenesis aggravation (Nkondjock, 2009). As well and from investigation view, an actual exposure to 0.1% caffeine could be capable for the incidence of hepatocellular adenoma (Furtado et al., 2013). Ultimately, the inconsistency over coffee is assumed to subsistence of variable confounders like diterpenes, cafestol and kahweol that could modify various enzymes have the responsibilities for carcinogenic detoxification (Larsson and Wolk, 2007). Palm oil in coffee as one contaminant of non-dairy creamers constitutes a source of dietary oxidized fats implied liver toxicity (Imoisi et al., 2015).

Constituents in coffee unlike caffeine namely acrylamide are potentially carcinogenic and genotoxic. Subsequently, Food and Drug Administration (FDA) has graded acrylamide as group 2A induced destructive damage and tumors to organs (Baylin et al., 2006). As to, it was proved that an existence of acrylamide (ACR) contributed to hepatocellular deteriorations (Hamdy et al., 2017).

Regarding to various the published investigations, coffee exist an association with fertility disruption is up to the crucial effect of caffeine through immediately impressed backwardly on the germinative epithelium, or on hypothalamus-pituitary gonads indirectly tend to a pronounced deviation in sex hormone (Dias et al., 2015).

Therefore, the current work was planned to validate the considerable link of coffee intake and public health hazards; in particular liver.

Materials and methods

Materials:

a) Experimental animals:

The study was carried out by using forty (40) adult male Wister albino rats weighing 120-150 gm. It obtained from Laboratory Animal House, Giza, Egypt. The rats undergone inspection just arrival. The experimentation was done in the Laboratory belonging to Department of Nutrition and Food Science, Faculty of Specific Education, South Valley University, Qena, Egypt. The animals were placed under good environmental conditions during the maintained experiment. Moreover, sufficient standard *ad libitum* and water were supplied along over the exposure.

b) Instant coffee:

Brand of Cappuccino was purchased from a local market in Qena City, Qena governorate, Egypt. Cappuccino was subjected chemical analysis for fatty acids using GC/MS-MS analysis.

Methods:

1- Experimental protocol:

The protocol was performed on forty (40) adult male albino rats weighing 120±30 gm., where animals were classified in the justified manner into 4 groups; ten (10) animals /group as following:

Control group: It only supplied standard diet plus distilled water.

Mocha treated groups were daily subjected to series of duplicated concentrations of Mocha according to Lestari et al. (2017) as the next:

Mocha (1): The rats orally received 0.72 ml of cappuccino as lower concentration.

Mocha (2): The rats orally received 1.44 ml of cappuccino as medium concentration.

Mocha (3): The rats orally received 2.16 ml of cappuccino as higher concentration.

The experimental protocol was performed daily for 7 weeks. The animals were carefully observed for any abnormalities along over the experiment.

2- Sampling:

a- Blood samples:

After the experiment was over, venous blood collected under an anesthetized control for biochemical examination. Serum was obtained via the subsequent centrifugation at 3000 rpm for 10 minutes. The resultant sera were assembled in clean Epindorf's tubes and stored frozen at -20° C till a forthcoming biochemical assay.

b- Liver collection:

Liver biopsies were taken from the list groups; then it immediately checked for macroscopically. Samples followed by fixation in 10% neutral buffered formalin for the microscopical findings.

c- Biochemical analysis:

1. Assessment of liver functions tests:

a) Determination of serum AST and ALT:

Biodiagnostic enzymatic kits (Biodiagnostic Company, Giza, Egypt) were dedicated in the vitro for assessment of enzymatic activities of AST and ALT in serum in a usage of colorimetric method as described by Reitman and Frankel (1957).

B) Determination of serum ALP:

Standard biodiagnostic kits also were aimed for calculation of activity of ALP through the colorimetric method mentioned by Belfield and Goldberg (1971).

2. Assessment of tumor marker and sex hormone:

a) Determination of serum CEA and T.T.:

Further, the method of Patrono and Peskar (1987) was employed for follow up enzymatic evaluation of levels of CEA and T.T. through enzyme-linked immunosorbent assays (ELIZA) using SpectraMax340.

3- Histopathological examination:

Liver biopsies were consecutively processed in an ascending series of ethanol. Consequent frozen paraffin blocks were sectioned according to Bacha and Bacha (2000) and stained with Harries hematoxylin and eosin (H&E.) for microscopically as expressed by Larson et al. (2011).

4 - Statistical analysis:

The results were statistically performed via one-way analysis of variance (ANOVA) using SPSS (Statistical Package for Social Sciences) according to Borenstein et al. (1997). The resultant data were expressive in Mean ± Standard Deviation. Significant difference was statistically when $P<0.05$.

Results

1- Chemical composition:

Chemical compositions of some fatty acids as expressed in Table 1 were significantly increased ($P<0.05$) in the cappuccino drink in according to the known permissible limits.

2- Biochemical analysis:

A. Liver function biomarkers ALT, AST and ALP (IU/l):

Instant coffee drink caused hepatic deterioration which could be represented by elevation of liver function biomarkers. The activity of ALT, AST and ALP significantly increased after Instant coffee treatment compared to control, the mean levels of serum ALT, AST and ALP were significantly elevated (P<0.05) in Mo. (2) & Mo. (3) groups as in Table 2.

Table 1. The composition of some fatty acids presented in instant coffee drink.

Parameters	(gm/ sachet)	LSD at 0.05
Total saturated fat	3.755± 0.05	0.111
Palimitic acid (PA) C16:0	0.446 ± 0.009	0.024
Oleic acid (OA) C18:1	1.073± 0.01	0.033
Linoleic acid (LA) C18:2	0.111± 0.003	0.007

Table 2. Effect of instant coffee drink on serum liver functions involving aspartate aminotransferase (AST) (IU/l), alanine aminotransferase (ALT) (IU/l) and alkaline phosphate (ALP) (IU/l) of albino rats of control and Mocha exposed groups (Mo. 1, 2 & 3). (Mean± SD).

Parameters	AST (IU/l)	ALT (IU/l)	ALP (IU/l)
Control	172.8±12.4^{a}	39.58±1.5^{a}	156.2±6.7^{a}
Mo. (1)	177.6±4.2^{a}	41.6±0.7^{a}	158.2±1.9^{a}
Mo. (2)	196.8±3.5^{b+}	51.0±1.0^{b+}	173.4±4.8^{b+}
Mo. (3)	230.2±3.8^{b+}	64.2±0.83^{b+}	186.4±1.67^{b+}

Means in the same column with different litters are significantly different when P<0.05.
+ → referring to significant increase when compared with control when P<0.05.

A- Effect on serum CEA (ng/dl):

Regarding to results of CEA, the difference in the mean values non-significantly changed in Mocha treated groups at the variant concentrations when compared with control described in Table 3.

B- Effect on serum T.T. (pg/ml):

The mean level of T.T. hormone was significantly reduced (P<0.05) among Mo. (3) group when compared with control group as shown in Table 3.

3- Histopathological results:

a- Macroscopic features:

At gross findings, liver of the control group manifested normal features of the color, shape and consistency. As for Mocha received groups suffered from darkish discoloration and congestion.

b- The entire body and liver weight:

In line with previous results, Table 4 expressed that body weight was significantly increased (P<0.05) in Mo. (2) and Mo. (3) groups as compared with control group.

Subsequently, liver weight represented significant increase (P<0.05) among groups Mo. (2) and Mo. (3), Table 4.

Table 3. Effect of instant coffee drink on serum level of CEA (ng/dl) and T.T (pg/ml) of albino rats of control and Mocha exposed groups (Mo. 1, 2 & 3). (Mean± SD).

Parameters	CEA (ng/dl)	T.T. (pg/ml)
Control	1.02±0.17[a]	2.53±0.07[a]
Mo. (1)	1.02±0.17[a]	2.45±0.21[a]
Mo. (2)	1.04±0.05[a]	2.16±0.08[a]
Mo. (3)	1.3±0.07[a]	1.19±0.07[b-]

Means in the same column with different litters are significantly different when P<0.05.
- → referring to significant decrease when compared with control when P<0.05.

Table 4. Effect of instant coffee drink on the entire body weight and liver weight of albino rats of control and Mocha exposed groups (Mo. 1, 2 & 3). (Mean± SD).

Parameters	Body weight (gm)	Liver weight (gm)
Control	233.4±5.4[a]	7.4 ±0.22[a]
Mo. (1)	234.0±5.4[a]	7.73±0.15[a]
Mo. (2)	239.8±3.7[b+]	8.39±0.31[b+]
Mo. (3)	246.6±4.0[b+]	10.0±0.09[b+]

Microscopic results:
Liver:

In Table 5, Fig. 1 & Fig. 2, liver parenchyma of the control albino rats demonstrated normal histological criteria with well-defined architecture, comprising normally arranged hepatic cords, as well healthy vasculature view that was obviously composed of intact formed blood sinusoids, central veins and bile ducts (Fig. 1a & 1b). Contradictory the histological observations of Mo. received groups, it exhibited pronounced histological disruptions in form of necrosis and cytoplasmic vacuolation of the hepatocytes as observed in Mo. (1) group (Fig. 2a). Liver of Mo. (1) group also suffered congestion and inflammation in were clearly detected the portal area (Fig. 2b). Likewise, liver of Mo. (2) demonstrated hepatic cytoplasmic vacuolation (Fig. 2c), furthermore dilatation in blood vessels comprising portal area with inflammatory cells infiltration, and hepatic necrosis (Fig. 2d). Ultimately, Mo. (3) group displayed prominent signs of vacuolar degeneration characterized by cytoplasmic vacuolation; as well hyperactivity of kuppfer cells was noticed (Fig. 2e). Moreover, Mo. (3) group was distinguished by focal accumulation of inflammatory cells among hepatic tissues (Fig. 2f).

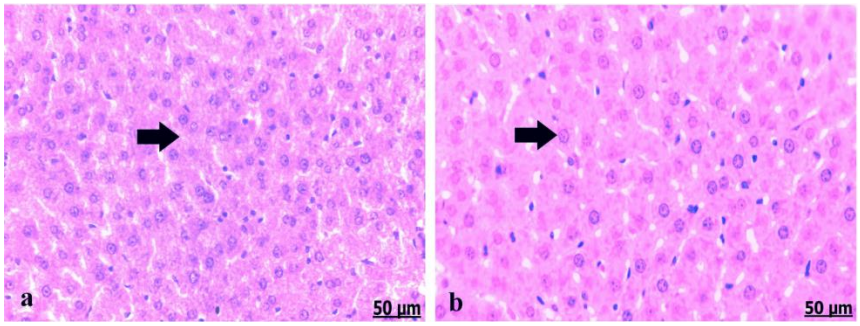

Fig. 1 (a-b). Liver of the control group showed normally arranged hepatic cords, besides normal vasculature. (H& E., bar= 50 µm).

Table 5. Histopathological scores of liver of albino rats of control and Mocha received groups classified according to severity of lesions into absent, (-), mild (+), moderate (++), and severe (+++).

Lesions	Control	Mo. (1)	Mo. (2)	Mo. (3)
Hepatic necrosis	-	+	+++	+++
Cytoplasmic vacuolation of hepatocytes	+	++	++	+++
Interstitial cells infiltration	-	+++	++	+++
Hyperactivity of kupffer cells	-	++	++	+++
dilatation and inflammation of the blood vessels	-	+++	+++	+++
Perivascular inflammation	-	+	++	++

Absent, (-), mild (+), moderate (++), and severe (+++).

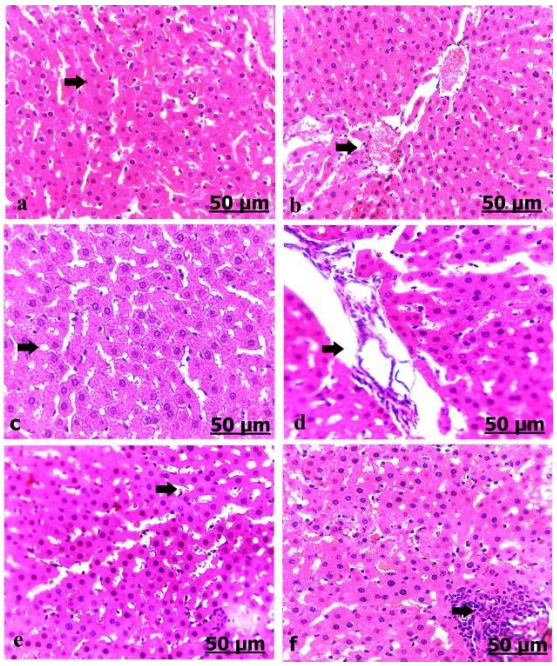

Fig. 2 (a-f). Liver of Mo. (1) showed necrosis and cytoplasmic vacuolation of the hepatocytes (a), congestion and inflammation in the portal area (b). Liver of Mo. (2) showed cytoplasmic vacuolation of the hepatocytes (c), dilatation in the blood vessels comprising portal area, in addition to inflammatory cells infiltration (d). Liver of Mo. (3) showed severe signs of vacuolar degeneration and hyperactivity of kupffer cells (e), and focal aggregation of inflammatory cells (f). (H& E., bar= 50 µm)

Discussion

Globally, the consumption of coffee has significantly grown as regarded to be beneficial to health and it tastes good. By chemical analysis, our results have conducted that coffee enjoyed higher percentage from oleic acid which plays crucial modulatory and therapeutic approaches against diseases and infection (Sales-Campos et al., 2013). However, sweetened coffee has dangerously affected the health status of the consumer. It has been hypothesized that liver was intended for caffeine or other coffee constituents. Definitely and fortunately, strong increases in the mean level of AST, ALT, and ALP were detected in our results. Such results support adverse correlation between coffee administration and liver enzymes (Nakanishi et al., 2000). In detail, various investigator researchers suggested that caffeinated coffee take role in hepatic upset, where liver is the main target of caffeine regardless of whether it is beneficial or useless (Casiglia et al., 1993). As well, a disruption in the liver function was stemmed from containment coffee with acrylamide (ACR) which is considered another major compound fundamentally affects liver induced hepatocellular disruption resulted in the enzymatic leakage into the blood circulation (Hamdy et al., 2017). Newer studies correlated

consumption of coffee exerts liver health hazards such as liver cirrhosis (Klatsky et al., 2006), parenchymatous carcinoma (Bravi et al., 2013), and nonalcoholic hepatic toxicity (Chen et al., 2014). The suitability of liver damage is biochemically compromised with substainsly alterations in enzymes activities of transaminases.

The miserable effect of instant coffee on total testosterone hormone is eventually attributed to caffeine detrimentally impact testes weight, sperm count and motility; consequently, significantly diminished fertility (Bassey et al., 2011). Caffeine could be able to alter testicular microarchitecture directly focus on Leydig cells conflicted in testosterone production (Bae et al., 2016). Parallel to Oluwole et al. (2016) who found that prolonged exposure to caffeine could weaken male reproductive function and modulate the cyto-architecture of testes.

With respect to carcinogenic influence, Bøhn et al. (2014) was implying a nearby connection between coffee consumption and the veritable cancer risk. This connection deduced when the level of carcinoembryonic antigen (CEA) was proceeding increased at the highest concentration of coffee. The carcinogenicity is due to the analysis as a result of the presence of acrylamide as mostly attributed to its metabolism by liver (Hamdy et al., 2017).

Concerning to histopathologically, heavy coffee ingestion was associated with extent of liver fibrosis and cirrhosis (Leung et al., 2011). Also, it was demonstrated that liver is specific targeted for caffeine, thus obviously resulted in hepatotoxicity and loss of integrity (Manne and Saab, 2015). Moreover, Choi et al. (2018) noticed hepatic damage and necrosis when mice daily administrated coffee extracts at 300 mg/kg.b.wt for 10 days. Consistently with histological results detected in liver was discretely characterized by cytoplasmic vacuolation and centrilobular necrosis, besides lymphocytic infiltration (Asha et al., 2008).

By the chemical composition of coffee, our results demonstrated higher concentration in fatty acids and palmitic acid (PA), which illegally exceeded limits. Palmitic acid (PA) occupied the main representative in saturated fatty acid. Impact of PA-enriched coffee was resulted in inflammatory conditions with an injury in the different tissues even liver (Marra and Svegliati-Baroni, 2018). Linoleic acid (LA) is a polyunsaturated fatty acid; consequent of LA in coffee exerted hepatoxicity and destructive damage of the tissues (Bilal et al., 2015).

Conclusion

The aforementioned results proved that variable concentrations of cappuccino advanced even in the dose and/or time are contributed in public health risks; thus asserted the possibility that liver is targeted locality for coffee metabolism. Wherefore, coffee should be prudently consumed particularly in in the drinkers possess chronic diseases.

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Conflict of interest statement

The authors declare that there is no potential conflict of interest.

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