Nutritional Interaction Effect of Zinc and Coffee on Serum Lipid Profile and Copper in Rats

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Abstract

Background: Coffee is one of the most popular beverages in the world. The present work was designed to study the effect of dietary coffee and zinc on serum lipid profile, zinc and copper in serum and liver.

Material and Methods: Forty two adult male albino rats were divided into six groups and fed on different diets ad libitum for 7 weeks as follows: Group (1) fed on standard basal diet, group (2) fed on standard diet supplemented with zinc chloride (ZnCl₂) at dose of (20 mg/kg diet), groups (3 and 4) fed on standard diet supplemented with boiled coffee at two doses (15 g and 30 g/kg diet) respectively, groups (5 and 6) fed on the previous two diets respectively but in combination with ZnCl₂ (20 mg/kg diet). At the end of the experiment, some biochemical analyses were measured in serum and liver.

Results: The results showed significant elevation of serum total lipids (TL), total cholesterol (TC), triacylglycerols (TG), low density lipoprotein-cholesterol (LDL-c), while a significant decrease of high density lipoprotein-cholesterol (HDL-c) in rats fed diet supplemented with low or high dose of coffee. When rats fed zinc with low or high dose of coffee, improvement in lipid parameters were observed. Also serum lipid peroxides as malondialdehyde (MDA) showed significant increase while blood glutathione (GSH) showed significant decrease in rats fed zinc, coffee (low or high dose) or zinc plus coffee (low or high dose).

Diet supplemented with zinc caused significant increase in serum and liver zinc, on the other hand, a significant decrease in serum and liver copper were recorded. Moreover, the high dose of coffee led to significant decrease in liver zinc. On feeding zinc and coffee together resulted in significant elevation of serum and liver zinc levels, but serum and liver copper decreased significantly.

Conclusion: So zinc may be able to overcome the adverse effects of coffee also administration of zinc plus coffee could decrease lipid peroxidation.

Keywords: Zinc, Chloride, Coffee, Lipid Peroxidation, Zinc, copper

Introduction

Coffee (the second most traded commodity after petroleum) is one of the most popular beverages in the world. Seventy five percent of soft drinks consumed regularly are coffee (Rojo Camargo, and Fatah, 1999). Coffee is a commonly consumed beverage with potential health benefits. There are three preparations of coffee that are commonly consumed and thus worthy of examinations; boiled unfiltered coffee, filtered coffee, and decaffeinated coffee. Also coffee has over a thousand chemicals, many are formed during the roasting process (Bonita et al., 2007).

The green coffee composition is carbohydrates, lipids, pentacyclic diterpenes (methyl cafestol, cafestol, kahweol), α, β, γ tocopherols, and protein content. Coffee contains several species of xanthines such as caffeine, theobromine and theophylline, also contains polyphenols. The potential bioactivities are caffeine, the deterpenes (cafestol and kahweol) found in the oil and the polyphenols (Parras et al., 2007). Several clinical and epidemiological studies have suggested that coffee consumption is associated with significant increases in total and low-density lipoprotein cholesterol levels.
lipoprotein cholesterol levels. Other studies, however, suggested that it is not the caffeine in coffee that is responsible for its hypercholesterolaemic effect (Nawrot et al., 2003). Two diterpenoid alcohols, cafestol and kahweol, found at significant levels in boiled coffee have been identified as hypercholesterolaemic components. Although these components are largely trapped by the use of a filter paper in coffee preparation, there is some evidence that consumption of filtered coffee is associated with small increases in serum cholesterol levels (Thelle 1995).

Bonita et al. (2007) hypothesize that the bioactive ingredients in coffee relating to heart disease are the polyphenols which in the body may be acting as protective antioxidants but also could have other beneficial mechanisms. However, the most well-known ingredient in coffee is the alkaloid caffeine. This compound has a variety of pharmacological effects with respect to mood, cognitive performance, and motor activity.

Caffeine has been reported as a scavenger of hydroxyl radical at millimolar concentrations in the study of electron spin resonance (ESR) spin trapping (Shi et al., 1991).

Although the mechanism of anticarcinogenic effect of caffeine is not clear, much of clinical interest underlies its potential role as an antioxidant in the control of oxidative damage. Uric acid that is structurally similar to caffeine is an important scavenger of both hydroxyl and peroxyl radicals (Lee, 2000).

Zinc is an essential trace element and plays an important role in the growth and development of animals and humans. Also, zinc is critical in maintaining membrane structure and function. Consequently, reduced Zn levels as a result of chronic caffeine consumption during the rapid growth period of developing organs might permanently affect their growth, which could lead to an impairment in organs function. (Yazdani et al., 2002).

Several studies have demonstrated that zinc supplementation can reduce oxidative damage, although zinc is not redox active compound. It probably does not act directly as an antioxidant (Alissa et al., 2004).

Zinc has also been observed in several studies to have an antiatherosclerotic effect and has been inversely associated in epidemiological studies with cardiovascular disease, but its mechanism of action is unclear (Lee et al., 2005).

There are pronounced nutritional and physiological interactions between Cu and Zn. For example, Cu absorption is increased in dietary Zn deficiency, and a high level of dietary Zn inhibits Cu absorption. The elements Zn and Cu may have a pronounced effect on cellular free radical production and metabolism. It is also possible that extra dietary Zn has an effect on the metabolism of hydrogen peroxide. The selenoprotein glutathione peroxidase (GSH-Px) is shown to be lower in the liver of Cu-deficient rats (Rossowska et al., 1995).

This study was conducted to investigate the antioxidant capacity of coffee and zinc and the effect of administration of one dose of zinc with coffee (at two levels) on lipid profiles and to evaluate serum and liver zinc and cupper after the administration of zinc and coffee.

Material & Methods

Material

Zn as ZnCl₂ was obtained from El Gomhoria company for chemicals and drugs. El Ameriya Cairo. Egypt. Coffee powder (Brazilian) moderate roasting which obtain from local markets.

Experimental Animals

Forty two male Sprague Dawley albino rats weighing (100-150 g) were purchased from the Egyptian organization for Biological products and vaccines (Helwaan Farm) and were fed on a standard laboratory diet for one week to acclimatize, food and water were offered ad libitum. Animals were divided randomly into six groups, each of 7 rats.

Preparation of Coffee

15 or 30 g coffee powder were prepared and supplemented to each 1 kg of standard diet. Two samples of coffee were prepared by dissolving 15 gm coffee/100
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ml water (low coffee) and 30 g coffee/100 ml water (high coffee), boiling for 10 minutes and added to standard diets after cooling to 50°C (Beynen et al., 1996).

**Experimental Design**

Rats were fed standard laboratory diet according to AIN (1977) and treated as following.

- **Group (1)** fed standard diet and served as control.
- **Group (2)** fed standard diet + ZnCl$_2$ (20 mg/kg diet) (Garg et al., 2008).
- **Group (3)** fed standard diet + Coffee (15 g/kg diet). (Beynen et al., 1996).
- **Group (4)** fed standard diet + Coffee (30 g/kg diet).
- **Group (5)** fed standard diet + ZnCl$_2$ (20 mg/kg diet) + Coffee (15 g/kg diet).
- **Group (6)** fed standard diet + ZnCl$_2$ (20 mg/kg diet) + Coffee (30 g/kg diet).

At the end of the experiment (7 weeks), the animals were fasted overnight then anaesthetized with ether, blood samples were collected from the portal vein and centrifuged for serum separation and kept at -20°C till used for biochemical analyses. Liver, kidney, spleen and heart were removed and washed in saline solution (0.9%), dried on filter paper and weighed.

Part of liver was preserved at -20°C for zinc and copper analyses.

**Biological Evaluation**

Body weight of each rat was measured, absolute and relative organs weight, food consumption and feed efficiency ratio were calculated.

**Biochemical Analyses**

Serum total lipids (TL) were analyzed by colorimetric method as published by Knight et al. (1972), serum triacylglycerols (TG) were determined by the method described by Fossati and Prencipe (1982). Serum total cholesterol (TC) was estimated by enzymatic colorimetric method described by Roeschlau et al. (1974).

Serum high density lipoprotein-cholesterol (HDL-C) was determined according to the method described by Richmond (1973). Serum low density lipoprotein-cholesterol (LDL-C) was estimated as described by Assmann et al. (1984).

Serum lipid peroxides expressed as malondialdehyde (MDA) was measured as described by Draper and Hadley (1990). Glutathione content (GSH) was measured as described by Beutler et al. (1963).

Samples tissues were digested in a digesting mixture. (concentrated sulphuric acid and perchloric acid 1:1) until it becomes clear and dilute with deionized water, filter and estimate by atomic absorption.

**statistical Analysis**

The data were statistically analyzed using one-way analysis of variance (ANOVA) according to Bailey (1995). Values of P < 0.05 were considered statistically significant.

**Results**

Table (1) shows no significant differences in body weight gain in all groups except for group of rats which fed on low coffee diet which shows significant decrease (P < 0.05) as compared to control.

There were significant increase (P < 0.05) of feed efficiency ratio for rats which fed on zinc diet, while significant decrease was observed in rats which fed on low or high coffee diet as compared to normal control.

Rats administered zinc plus low or high coffee diet showed significant increase in food intake with significant decrease in feed efficiency ratio as compared to group administered zinc only.

Also as shown in table (1), there were no significant differences in cardio somatic, nephro somatic and spleeno somatic index in all groups while rats which fed zinc diet, showed significant decrease in hepato somatic index as compared to normal control group , on the other hand rats fed on zinc plus low coffee diet showed significant decrease in hepato somatic index as compared to rats which fed zinc or low coffee diet

Table (2) revealed that rats fed on zinc diet showed significant decrease in serum total cholesterol (TC) and low
density lipoprotein-cholesterol (LDL-C), while high density lipoprotein cholesterol (HDL-C) showed a significant increase as compared to control. Low and high doses of coffee caused a significant elevation in serum total lipids (TL), total cholesterol (TC), triacylglycerols (TG), LDL-C while significant decrease was recorded in serum HDL-C as compared to control group.

When rats administered zinc plus low or high dose of coffee, there were significant increase in TL, TC, LDL-C as compared to zinc supplemented group; while HDL-C showed significant decrease as compared to group of zinc. Also, when rats fed on zinc with low or high coffee diet, an improvement in all lipid parameters were observed as compared to rats fed on low or high coffee diet.

As shown in table (3), there were significant decrease of serum malondialdehyde (MDA) with significant increase of blood glutathione (GSH) levels in all groups when compared to normal rats. There were significant decrease in serum MDA but significant increase in blood GSH content in rats fed on zinc with low or high coffee diet as compared to groups fed on low or high coffee diet only or rats received zinc only.

Table (4) revealed that diet that supplemented with zinc caused significant increase in serum and liver zinc, on the other hand significant decrease in serum and liver copper were detected in comparison to the control group.

Moreover, receiving high coffee dose led to significant increase of serum zinc and significant decrease in liver zinc and serum copper. The administration of zinc with low or high coffee dose led to significant decrease of serum and liver zinc with significant increase of liver copper in comparison to rats received zinc but not restored it to the control values. While on comparing the groups which fed on zinc with low or high coffee diet with the groups fed on low or high coffee diet only, significant increase in serum and liver zinc was detected, while a significant decrease in serum and liver copper were observed.
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### Table (2): Serum Lipid Parameters of Rats Fed on Diets Contain Zinc and/or Coffee.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>TL (mg/dl)</th>
<th>TC (mg/dl)</th>
<th>TG (mg/dl)</th>
<th>HDL-c (mg/dl)</th>
<th>LDL-c (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>609.86 ± 0.639</td>
<td>221.77 ± 0.779</td>
<td>107.51 ± 0.400</td>
<td>50.06 ± 0.265</td>
<td>150.20 ± 0.764</td>
</tr>
<tr>
<td>Zinc</td>
<td>608.73 ± 0.329</td>
<td>218.99 ± 0.488</td>
<td>109.64 ± 0.520</td>
<td>56.5 ± 0.207</td>
<td>140.56 ± 0.401</td>
</tr>
<tr>
<td>Low coffee</td>
<td>687.1 ± 0.287</td>
<td>249.86 ± 0.498</td>
<td>137.99 ± 0.757</td>
<td>41.7 ± 0.204</td>
<td>180.69 ± 0.637</td>
</tr>
<tr>
<td>High coffee</td>
<td>779.61 ± 0.496</td>
<td>279.79 ± 0.599</td>
<td>179.56 ± 2.79</td>
<td>36.11 ± 0.157</td>
<td>207.76 ± 0.683</td>
</tr>
<tr>
<td>Zinc + Low coffee</td>
<td>618.94 ± 0.441abc</td>
<td>223.91 ± 0.544bc</td>
<td>106.10 ± 0.547c</td>
<td>50.33 ± 0.36bc</td>
<td>144.54 ± 0.617abc</td>
</tr>
<tr>
<td>Zinc + High coffee</td>
<td>659.94 ± 0.329abd</td>
<td>243.06 ± 1.76ad</td>
<td>112.07 ± 0.482ab</td>
<td>45.60 ± 1.14ab</td>
<td>160.90 ± 1.690abs</td>
</tr>
</tbody>
</table>

Each value represents the mean ± SE.

a: Significant differences from control at (P < 0.05).
b: Significant differences from zinc supplemented group at (P < 0.05).
c: Significant differences from low coffee supplemented group at (P < 0.05).
d: Significant differences from high coffee supplemented group at (P < 0.05).

### Table (3): Serum Malondialdehyde (MDA) and Blood Glutathione (GSH) of Rats Fed on Diets contain Zinc and/or Coffee

<table>
<thead>
<tr>
<th>Parameters</th>
<th>MDA (µmol/L)</th>
<th>GSH (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>4.971 ± 0.053</td>
<td>15.94 ± 0.062</td>
</tr>
<tr>
<td>Zinc</td>
<td>4.48 ± 0.068a</td>
<td>18.99 ± 0.142a</td>
</tr>
<tr>
<td>Low coffee</td>
<td>4.69 ± 0.077a</td>
<td>16.58 ± 0.248a</td>
</tr>
<tr>
<td>High coffee</td>
<td>4.67 ± 0.067a</td>
<td>17.45 ± 0.058a</td>
</tr>
<tr>
<td>Zinc + Low coffee</td>
<td>4.02 ± 0.100abc</td>
<td>22.78 ± 0.130abc</td>
</tr>
<tr>
<td>Zinc + High coffee</td>
<td>3.91 ± 0.082abcd</td>
<td>24.92 ± 0.071abcd</td>
</tr>
</tbody>
</table>

Each value represents the mean ± SE.

a: Significant differences from control at (P < 0.05).
b: Significant differences from zinc supplemented group at (P < 0.05).
c: Significant differences from low coffee supplemented group at (P < 0.05).
d: Significant differences from high coffee supplemented group at (P < 0.05).
Table (4): Serum and Liver Zinc and Copper in Rats Fed on Diets Contain Zinc and/or Coffee.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Serum zinc (µg/dl)</th>
<th>Liver zinc (µg/g tissue)</th>
<th>Serum copper (µg/dl)</th>
<th>Liver copper (µg/g tissue)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>76.98 ± 0.071</td>
<td>49.09 ± 0.106</td>
<td>98.61 ± 0.141</td>
<td>6.97 ± 0.127</td>
</tr>
<tr>
<td>Zinc</td>
<td>92.99 ± 0.124 a</td>
<td>63.98 ± 0.108 a</td>
<td>59.86 ± 0.079 a</td>
<td>5.07 ± 0.089 a</td>
</tr>
<tr>
<td>Low coffee</td>
<td>77.01 ± 0.180</td>
<td>48.88 ± 0.093</td>
<td>95.61 ± 1.94 a</td>
<td>6.94 ± 0.066</td>
</tr>
<tr>
<td>High coffee</td>
<td>78.30 ± 0.173 a</td>
<td>46.84 ± 0.126 a</td>
<td>96.06 ± 0.11 a</td>
<td>6.97 ± 0.068</td>
</tr>
<tr>
<td>Zinc + Low coffee</td>
<td>88.51 ± 0.190 abc</td>
<td>60.50 ± 0.183 abc</td>
<td>68.74 ± 0.132 abc</td>
<td>5.89 ± 0.082 abc</td>
</tr>
<tr>
<td>Zinc + High coffee</td>
<td>85.74 ± 0.153 abd</td>
<td>58.93 ± 0.102 abd</td>
<td>57.79 ± 0.205 ad</td>
<td>5.70 ± 0.131 abd</td>
</tr>
</tbody>
</table>

Each value represents the mean ± SE.

a: Significant differences from control at (P < 0.05).
b: Significant differences from zinc supplemented group at (P < 0.05).
c: Significant differences from low coffee supplemented group at (P < 0.05).
d: Significant differences from high coffee supplemented group at (P < 0.05).

Discussion

In the present study administration of zinc reduced the body weight in rats fed on low coffee diet. Organo somatic index showed no significant effect in all groups, except for hepato somatic index which showed significant decrease in rats fed on zinc plus low coffee diet as compared to rats which fed on zinc or low coffee diet, also hepato somatic index showed significant decrease in rats fed on zinc diet as compared to normal rats.

However administration of zinc plus low or high coffee dose resulted in significant increase in food intake, but significant decrease in feed efficiency ratio.

Yazdani et al. (2002) found that there were no significant differences in body weight among groups fed caffeine or zinc plus caffeine.

Similarly, Bonita et al. (2007) established that there was no significant effect of any dose of coffee, caffeinated or decaffeinated, on weight gain or food consumption.

El-Hendy et al. (2001) found that there was significant increase in liver and spleen weights. However, kidney and heart weights were not affected in rats fed adequate or low levels of zinc.

The present study demonstrated that low and high doses of coffee caused significant elevation in serum total lipids (TL), TC, TG and LDL-C but a significant decrease of HDL-C.

Several studies have suggested that coffee consumption is associated with significant increase in total and low density lipoprotein-cholesterol levels. Other studies suggest that it is not the caffeine in coffee that is responsible for its hypercholesterolemic effect, but two diterpenoid alcohols (cafestol and kahweol) found at significant levels in boiled coffee has been identified as hypercholesterolemic components (Nawrot et al., 2003).

Bonita et al. (2007) reported that only heavy consumption (> 6 cups/day) of boiled unfiltered coffee is
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harmful to the heart as a result of the does related plasma cholesterol and LDL increase due to the diterpene oils.

When zinc administered to rats fed low or high coffee diet an improvement in all lipid parameters were observed.

Hyper cholesterolemic rabbits supplemented with zinc had trend to lower plasma cholesterol and LDL increase due to the diterpene oils. When zinc administered to rats fed low or high coffee diet an improvement in all lipid parameters were observed. Jenner et al., 2007).

Wood et al. (1991) found that zinc treatment produced decrease in TG concentration from (211.44 to 181.77 mg/dl).

The study of Partida-Hernandez et al. (2006) in diabetic patients demonstrated that administration of zinc resulted to reduction of TC and increase of HDL-Cholesterol which indicate that its administration improved the individual’s metabolic condition.

Female rats fed coffee oil for 4 weeks showed significantly higher plasma cholesterol and triacylglycerols levels (Al-Kanhal et al., 1999).

Administration of zinc only, low or high coffee dose could decrease lipid peroxidation by reducing MDA level and increasing blood glutathione as compared to normal control. Zinc in combination with low or high coffee dose could inhibit lipid peroxidation more than the administration of each of them alone.

Plasma, liver and kidney Malondialdehyde (MDA) levels were lower in zinc supplemented group than control, whereas the GSH concentration of the same tissues had an inverse trend (Ozturk et al., 2003 and Duzguner and Kaya, 2007).

The trace element zinc has antioxidant effects, protecting -SH groups and also known to restrict the production of free radicals such as superoxide and malondialdehyde (Powell, 2000). In accordance with our results Parras et al. (2007) found inhibition of lipid peroxidation in the presence of coffee beverages from 12 different points of origins. All the coffees studied, regardless of origin were very effective scavengers of lipoperoxyl radicals.

Bonita et al. (2007) hypothesize that the polyphenols metabolites from coffee were bound to the LDL , VLDL and protected it from oxidation.

The coffee components, kahweol and cafestol increased GSH levels apparently through the induction of the rate limiting enzyme GSH synthesis which may be a key factor in the chemo preventive potential of coffee components (Huber et al., 2002).

The present study shows that zinc alone or in combination with low or high coffee concentrations led to increased serum and liver zinc and decreased serum and liver copper. High dose of coffee resulted in significant increase of serum zinc, while liver zinc and serum copper were significantly decreased.

Irato and Albergoni (2005) observed that rats fed with Zn-supplemented diet (6 g/kg solid diet) for 30 days had increased zinc contents in liver, kidney and intestine and reduced the copper content similar to controls, corresponding to the synthesis of metallothioneins (MTs). Painemalgorale and Bebe (1996) indicated that moderately high zinc diet (60 mg/kg diet) decreased plasma copper concentration and this was accompanied by comparable changes in plasma ceruloplasmin level.

Rossowska et al. (1994) found that zinc supplemented diet (0.6 g Zn Cl2/kg diet) or zinc plus caffeine (2 mg/kg body wt of dams/day) increased liver zinc and decreased liver copper levels than control. In summary, the results from the present investigation showed that coffee and zinc are highly effective antioxidant. When zinc administered to rats in combination with coffee an improvement of all lipid parameters and an inhibition of lipid peroxidation were observed.

References


تأتي التداخل الغذائي للزنك والقهوة على مستوى الدهون والنحاس في مصل الفئران

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تأتي القهوة من أكثر المشروبات الشعبية شيوعاً في العالم. يهدف هذا البحث إلى دراسة تأثير القهوة والزنك على مستوى الدهون في مصل الفئران وكذلك على مستوى الدهون والنظاس في مصل وكبد الفئران.

أجريت هذه الدراسة على ذكور الفئران البديعة والبالغ عددهم 42 تم تقسيمهم إلى 6 مجموعات وتتم تغذيتهم على الوجبات لمدة 7 أسابيع كالتالي:

المجموعة الأولى: تغذية على الوجبة القائمة والمجموعة الثانية: تغذية على الوجبة القياسية المدعمة بزنك كلوروري (20 مجم/كلغ)، و المجموعة الثالثة: مع رابعه تم تغذيتهم على الوجبة القياسية المضاف إليها القهوة السابقة غليانها بجرعتين (15 جم/كلغم/كجم غذاء، 30 جم/كجم غذاء) على التوالي.

أما المجموعة الخامسة والسادسة فقد تم تغذيتهم على نفس الوجبات السابقة على التوالي ولكن مضاد الفيما كلوروري الزنك (20 مجم/كلغم غذاء). وفي نهاية التجربة تم تقييم بعض التحاليل البيولوجية في مصل وكبد الفئران.

أظهرت النتائج وجود ارتفاع معنوي في مستوى الليبيدات الكلي والكولسترول والجلوكيريدات الثلاثية وكولسترول الليبروتينات منخفضة الكثافة وكذلك حدث انخفاض معنوي في مستوى كولسترول الليبروتينات مرتفعة الكثافة في حالة التغذية بجرعتين القهوة فقط للفئران.

وقد حدث تحسن ملحوظ في هيئة الليبيدات عند إعطاء الزنك مع القهوة (الجرعة المنخفضة أو المرتفعة).

كما ارتفع مستوى بيروكسيديت الليبيدات المعبر عنها بالمالونداي الدهيد بالسربين بينما ارتفعت مستويات الجلوتاتيون بالذات انخفاضاً معنويًّا في حالة إعطاء الفئران الزنك والقهوة كل على حدة و الزنك مع القهوة سواء بالتركيز المنخفض أو المرتفع.

كما ارتفع مستوى الزنك انخفاض مستوى النحاس في مصل وكبد الفئران. باعطاء الزنك علاوة على ذلك ركبت الجلعة العالية من القهوة إلى انخفاض معنوي في مستوى الزنك في الكبد.

ويرجى ذلك إلى القدرة على تقليل انسداد الزنك عند مقارنة المجموعات التي تغذت على الوجبة تحتوي على الزنك مع القهوة لوحظ ارتفاع ملحوظ في مستوى الزنك مع انخفاض ملحوظ في مستوى النحاس في كل من مصل وكبد الفئران.

وذلك يُرجى الملاحظ أنه يكون للزنك القدرة على التغلب على التأثير العكسي للقهوة على مستوى الزنك والنحاس وكذلك على صورة الدهون بمصل الفئران كما أن إعطاء الزنك مع القهوة أدى إلى انخفاض أقصى الليبيدات.