STUDY THE EFFECT OF CAFFEINE ON BODY WEIGHT GAIN AND LIPID PROFILE IN ADULT MALE RATS TREATED WITH HYDROGEN PEROXIDE.

Faraj H.Joni AL-Bidhani , .Adel M. Hassen Alzobidy

.Abdul Razzak N. Khudair

Department of physiology and pharmacology,Veterinary Medicine Collage, University of Basrah, Basrah, Iraq.

(Received 17 December 2017 ,Accepted 2 January 2018)

Keywords:- Caffeine ,H_2O_2 lipid profile.

Corresponding author E.mail: alzobidy_dr.adel@yahoo.com

ABSTRACT

The present study was aimed to determine the ameliorative effect of caffeine on Body weight and lipid profile in male rats treated with hydrogen peroxide (H_2O_2) .

Seventy Two adult male rats were used in this study . The study included two experiments ,in each experiments 36 males were randomly assigned two six equal groups of six animals in each group .The animals in both experiments were treated with the same substances and doses for each group as follows .Group one (control) animals were drenched normal saline ,Group two animals were treated (5.63 mg/kg. Bw)H_2O_2 daily by oral gavage also group three , animals were treated with low dose caffeine (150 mg /kg Bw) daily .Group four , animals were treated with high dose caffeine (250 mg /kg Bw) daily . Group five . animals were treated (5.63 mg/kg. Bw)H_2O_2 dose after 1 h animals were given low dose of caffeine (150 mg/kg Bw) .Group Six animal were treated with H_2O_2 dose(5.63 mg/kg Bw) each rat after one hour was given high dose of caffeine (250 mg /Kg Bw) . The first experiments lasted for one month and second experiments lasted for two months . At the end of the two experiments, animals of all group were sacrificed under chloroform anesthesia .Blood samples were collected from

the heart directly by cardiac puncture and the serum was separated to measure the lipid profile .

The result revealed a significant decreased in body weight gain in H_2O_2in first and second experiments compared with control group . While a significant improvements were recorded in body weight gain in all treated groups compared with H_2O_2 group but still significantly lower compared with those of control group. A significant increase in TC ,TG and LDL-c were recorded in H_2O_2 group in both experiments compared with control group on the other hand no significant difference was recorded in HDL-c level in H_2O_2group in first experiments while a significant decreased was recorded in second experiments compared with control group . Finally a significant degrees of improvement were observed in lipid profile in all treated group compared with H_2O_2 .

INTRODUCTION

The word caffeine originated from the German word "Kaffee" and the French word "café" both directly translating to mean "coffee" , caffeine has been a part of our global history for thousands of years.Each country has its own story and source of caffeine .One of the most caffeine findings was in Ethiopia, the folk stories passed between generations says that a farmer, who had recently moved his goats to a new pasture found them to be restless and ancy, and noted that they were grazing on small berries, these berries were later dried and called "coffee beans",[1],The caffeine molecule is classified as an alkaloid, meaning that it is nitrogen- based compound that is extracted from plants. The chemical formula for coffee is C8 H10N4O2, is found in coffee [2].Caffeine from coffee and other beverages is absorbed by the small intestine within 45 minutes of ingestion and distributed throughout all bodily tissue, peak blood concentration is reached within 1-2 hours, caffeine can also absorbed rectally, caffeine reaches peak values in the blood between 15 and 120 minutes dependent on individual physiology and vehicle (liquid, capsule, gum, etc.), [3]. Hydrogen peroxide is produced naturally in the atmosphere when ultraviolet (UV) rays of the sun react with oxygen when there is existence of moisture in the air ozone (chemical formula O_3) is basically comprised of one free oxygen (O) and two additional oxygen atoms. As ozone is exposed to water, the additional oxygen atoms separates quite effort Lesley the water (chemical

formula H_2O), unites with the additional oxygen atom turns into hydrogen peroxide (H_2O_2), [4].H2O2 applications in the industrial about 60% of the world production , H_2O_2 is used for pulp and paper in bleaching also in the manufacture of sodium per carbonate and sodium per carbonate which are used in certain waste-water treatment processes to remove organic impurities, this achieved by oxidation processes such as fentosre action, [5]. H_2O_2 is dangerous when swallowed lead to internal bleeding and burn to throat, nausea, vomiting, obstruction of the respiratory tract, confusion, coma, convulsion, cyanosis and cardio-respiratory arrest, [6]H_2O_2 is very un stable compound that breaks down readily to form molecular oxygen and water, the boiling point of H_2O_2 is much higher than the boiling point of water, H_2O_2 never boiling because the heat up explodes and decomposes into water and oxygen, critical temperature is 457-degree Celsius [7].

MATERIALS AND METHODS

Seventy two adults male rats were used ,their body weight arranged between (٢٥0±25)g, the animals were housed in individual cages measuring (50x50 cm), in animal house of Veterinary Medicine College-University of Basra. All animals were exposed to the same environmental including climate feeding and adaptive on place for two weeks before treatment. The study included two experiments in which the animals were divided equally between the experiments with 36 animals per experiments. The animals of each experiments were divided randomly into six equal groups on of six animals per group and the animals of each group in both experiments treated with the same substances and doses as follow :

1-control group, in which rats were given normal saline orally by oral gavages.

2-Group two, in which rats were given H_2O_2doses (5.63 mg/kg BW) orally by gavages.

3-Group three , in which rats were given low doses caffeine(150 mg /Kg/BW)by oral gavages daily.

4-Group four, in which rats were given high doses caffeine(250 mg /Kg/BW) by oral gavages daily

5-Group five, male rats given H_2O_2doses (5.63 mg/kg Bw) and after one hour given low dose caffeine(150 mg /Kg/BW) by oral gavage.

6-Group six, male rats given H_2O_2doses (5.63 mg/kg Bw) and after one hour given high dose caffeine(250 mg /Kg/BW) by oral gavage

The experiments continue for one and two month respectively .At the end of each experiments the following parameters were measures :-

Measurement of Body Weights Gains

The weight of each animal was recorded in the zero and thirty day in first experiments and in zero and sixty day in second experiments, by using electronic balance .

Collection of blood samples.

Blood sample (٥ ml) were collected by from heart puncher , after the rats anaesthetized with chloroform . Three ml of blood collected from each animal were stored in tube without anticoagulant and allowed to clot at room temperature . Then the blood samples were centrifuged at(5000 rpm) for 30 minutes and serum sample were stored in polyethylene tubes at (– 20c) until used for estimation of lipid profile .

Measurements of lipid profile :

The lipid profile tests were done in the laboratory by using chemistry auto analyzer made in Germany by human star company serial no.20628 ,the machine has 54 wells which numbered from 1 to 54 , The serum samples deposited in each specific wells . The reagent was put in a special container beside the wells. The serum biochemical parameters estimated by this instrument were lipid profile TC,TG , LDL-c and HDL-c.

RESULTS

The result showed a significant decreased ($p \leq 0.05$) in final body weight and body weight gain in the animal group treated with H_2O_2compared with control group in both first and

second experiments , Table (1and 2) .How ever no significant differences were recorded in final body weight in all treated groups compared with control with exception of the group treated

with H_2O_2 + 250 mg /kg bw caffeine where final body weight was significantly lower ($p \leq 0.05$) than control group after one month of treatment . A significant increased in the body weight gain were recorded in all treated group compared with H_2O_2 treated group after one month of treatment .Similar result were obtained for final weight and weight gain after two month of treatment. Table (3and 4) revealed a significant increased ($p \leq 0.05$) in serum TC,TG and LDL –c of H_2O_2 treated group compared with control in both first and second experiments while no significant difference recorded in HDL –c level between H2O2 treated group and control group after one month of treatment ,however a significant decreased ($p \leq 0.05$) in HDL-c was recorded in H_2O_2 treated group after two month of treatment compared with control group . Moreover significant degrees of improvement were recorded in all other treated groups compared with H_2O_2 treated group in both one and two month treated groups

Table (1): The effects of caffeine and H2O2 on final B.W and weight gain after one

Treatment	Initial B.W. (g)	Final B.W (g)	Weight gain (g)
Control	263.33 ± 5.43 a	294.5 ± 7.11 a	31.16± 3.31 a
H2O2	264.31 ± 5.46 a	273.83±6.14 b	9.5±1.32 b
Caffeine 150 mg/kg	267.16 ± 7.13 a	293.52±8.35 a	26.33±2.05 ac
Caffeine 250 mg/kg	259.16 ± 7.77 ab	283.16±6.45 a	24.83±2.43 cd
Caffeine 150 mg/kg + H2O2	261.66 ± 8.71 a	283.16±9.41 a	20.60±2.07 d
Caffeine 250 mg/kg + H2O2	257.53 ± 7.82 a	263.33±8.32 b	5.66±2.53 e

month of treatments (Mean ± SD) n=6

The different small letters refer to significant differences at ($p \leq 0.05$) among treatment.

Table (2): The effects of caffeine and H2O2 on final B.W and weight gain after two month of treatments (Mean ± SD) n=6

Group	TC mg/dL	TG mg/dL	HDL mg/dL	LDL mg/dL
Control	54.94±3.97 a	٣٧.19±3.22 a	39.65±4.19 ab	14.38±1.10 c
H2O2	73.10±7.83 b	70.13±2.49 b	36.21±3.33 ab	21.49±3.35 b
Caffeine 150 mg/kg	51.34±3.98 a	38.70±3.31 a	38.20±5.11 ab	14.13±3.27 c
Caffeine 250 mg/kg	52.81±6.42 a	39.30±4.20 a	41.01±3.71 b	31.70±1.71 a
Caffeine 150 mg/kg + H2O2	60.71±4.77 c	47.15±4.77 c	39.44±3.0 b	18.96±2.94 b
Caffeine 250 mg/kg + H2O2	59.10±4.66 c	41.45±3.91 c	42.97±4.90 b	17.60±2.14 bc

The different small letters refer to significant differences at (p≤0.05) among treatment.

Table (3): Effects of caffeine and H2O2 on serum lipid profile: after one month of treatments . (Mean ± SD) n=6

Treatment	Initial B.W. (g)	Final B.W (g)	Weight gain (g)
Control	٢٥٩.66 ± 8.93 a	315.01 ± 8.93 a	55.33± 3.59 a
H2O2	261.83 ± 8.11 a	240.03±6.39 b	- 21.83±3.49 b
Caffeine 150 mg/kg	262.50 ± 7.49 a	290.83±8.25 c	28.33±3.41 c
Caffeine 250 mg/kg	258.33± 5.28 a	278.41±4.39 d	19.66±2.33 d
Caffeine 150 mg/kg + H2O2	262.31 ± 7.41 a	276.66±8.99 d	14.33±1.85 e
Caffeine 250 mg/kg + H2O2	256.32± 6.52 a	269.33±8.91 d	12.66± 1.81 e

The different small letters refer to significant differences at (p≤0.05) among treatment.

Table (4): Effects of caffeine and H2O2 on serum lipid profile: after two month of treatments. (Mean ± SD) n=6

Group	TC mg/dL	TG mg/dL	HDL mg/dL	LDL mg/dL
Control	53.78±4.11 c	٣8.54±4.44 c	36.38±2.34 a	22.18±2.11 c
H2O2	84.18±6.77 a	75.37±5.74 a	27.95±2.67 c	29.53±3.11 a
Caffeine 150 mg/kg	49.75±3.95 c	41.48±3.99 d	35.09±4.21 b	22.99±2.93 c
Caffeine 250 mg/kg	48.24±5.59 c	42.63±4.81 d	32.89±3.44 b	21.34±2.37 c
Caffeine 150 mg/kg + H2O2	59.69 ±4.12 b	48.37±3.41 c	33.03±2.97 b	30.94±3.95 a
Caffeine 250 mg/kg + H2O2	61.67± 3.95 b	53.75±3.66 b	35.94± 2.77 b	27.85±2.99 b

The different small letters refer to significant differences at ($p \leq 0.05$) among treatment.

DISCUSSION

The results observed in the present study showed that the administrations of H2O2 orally caused decreased in body weight gain in one and two month of treatment table(1and 2) .These result confirmed with study by (8) who showed that H2O2 decreased the body weight due to improved insulin resistance,(9)they noted that H2O2 caused body weight loss by that the toxin interfere with the cells ability to absorb the oxygen which cause low oxygen concentration in body fluid .(10)showed that the H2O2 caused significant decrease of body weight by reducing the connective tissue formation .

Administration of two doses caffeine 150 and 250 mg/kg.bw orally for one and two months caused slight increase in body weight gain , the present study agreed with study done by (11) who showed that consumption of caffeine for 14 days caused suppressive effect on weight gain and visceral fat accumulation in mice .The (12) and (13) observed a statistically significant increase as association between caffeine and leptin concentration , which is a protein hormone that is produced by fat cells , it gives instruction to the

hypothalamus and can serve as measure of total fat stores , leptin is produced and released by fat cells in proportion to the level of fat storage. study by (14) showed an increased leptin levels may have effects on tissue and organs that remain sensitive to high leptin. Also the result disagreed with (15) noted that ingestion of caffeine caused significant decrease in body weight due to caffeine has low glycemic index .

The results in the present study in table (3and 4)showed that significant increase of TC ,TG , LDL and decrease a significant the level of HDL in group H2O2 treated animals .Results are similar to study done by(16)they showed that H2O2 significantly lead to increase on TC ,TG and LDL levels in animals treated with H2O2 .(17)showed that the levels of TC , TG , HDL and LDL were significantly increased in H2O2 treated group for two months than control group due to the effect of H2O2 on suppressing lipid peroxidation .

Treated animal with H2O2 caused significant increase of lipid peroxide(18)

The same table showed that the levels of TC ,TG and LDL increased in caffeine treated animals.These results are similar to study done by(١٩) they demonstrate that the serum cholesterol level increate with caffeine treated group in experimental animals , also they showed that caffeine increase the level of HDL in animals treated , study done by (2٠) showed that the caffeine significantly increase the levels of TC , HDL and TG and no significant difference in the serum LDL in treated group .Caffeine increase the serum TC in treated animals, (2١) those results agreed with the results of current study .

The increase of TC, TG , LDL concentration due to suppressed bile acid synthesis (2٢).The result in present study differs from study done by (٢٣)who showed that caffeine with treated group significantly increase HDL .The present study un coordinated with study done by(٢٤) who showed that lower level of TC in caffeine treated group and no significant difference in the level of serum HDL and TG concentration compared with control group .

دراسة تاثيرالكاقئين على الزيادة الوزنية وصورة الدهون في لذكور الجرذان البالغة المعاملة ببيروكسيد الهيدروجين .

فرج حسن البيضاني ،عادل موسى الزبيدي ،عبد الرزاق نعيم خضير

فرع الفسلجه،كلية الطب البيطري،جامعة البصره ،البصره ،العراق.

الخلاصة

هدفت هذه الدراسة لتحديد التاثير المحسن لاعطاءالكافائين على الزيادة الوزنية و صورة الدهون في ذكور الجرذان البالغة المعاملة ببيروكسيد الهايدروجين.

اجريت الدراسة على اثنتان وسبعون من ذكور الجرذان البالغة التي وزعت عشوائيا الى ست مجموعات متساوية (احتوت كل محموعة على ستة ذكور) واجريت التجربة لمدة شهر ولمدة شهرين .

جرعت المجموعة الاولى المحلول الفسلجي الطبيعي بواسطة الانبوب الفموي واعتبرت مجموعة سيطرة بينما جرعت المجموعة الثانية مامقداره (٥.٦٣ ملغم /كغم من وزن الجسم) بيروكسيد الهيدروجين بواسطة الانبوب الفموي وعوملت المجموعة الثالثة بجرعة منخفظة من الكافائين (١٥٠ ملغم /كغم) من وزن الجسم يوميا وعوملت المجموعة الرابعة بجرعة مرتفعة من الكافائين(٢٥٠ ملغم /كغم) من وزن الجسم يوميا وعوملت المجموعة الخامسة (٥.٦٣ ملغم /كغم من وزن الجسم) بيروكسيد الهايدروجين وبعد ساعة اعطيت جرعة منخفضة من الكافائين(١٥٠ ملغم /كغم) من وزن الجسم بينما عوملت المجموعة السادسة بجرعة (٥.٦٣ ملغم /كغم من وزن الجسم) بيروكسيد الهايدروجين وبعد ساعة اعطيت جرعة عالية من الكافائين(٢٥٠ ملغم /كغم) من وزن الجسم . استمرت التجربة الاولى لمدة شهر و التجربة الثانية لمدة شهرين .

بعد ذلك تمت التضحية بجميع الحيوانات وجمعت نماذج الدم بواسطة وخز القلب وعزل مصل الدم وذلك لقياس الصفات الكيموحيوية والتي شملن صورة الدهون في مصل الدم كالبروتين الدهني عالي الكثافة وواطيء الكثافة والكوليسترول الكلي والكليسريدات الثلاثية اضافة الى الزيادة الوزنية .

اظهرت النتائج ان هنالك انخفاض معنوي في الزيادة الوزنية والبروتين الدهني عالي الكثافة في مجموعة الحيوانات المعاملة ببيروكسيد الهيدروجين بينما كانت هناك زيادة معنوية في كل من الكوليسترول الكلي والكليسريدات الثلاثية والبروتين الدهني واطيء الكثافة مقارنة بمجاميع التي اعطيت بالكافائين ومجموعة السيطرة .

REFERENCES

1- **Escohotado,;** (1999). A Brief history of Drugs: from the stone age to the stoned Age. park street press. ISB No. 89281-826-3.

2- **Balentine,. D. A;** (1998),. Tea the plant and its manufacture, chemistry and consumption of the Beverage in caffeine, ed. G Spiller.

3- **Fredholm B. B.,** (1999). Actions of caffeine in the brain with special reference to factors that contribute to its wide spread use. Pharmacol Rev.

4- **Edwards,.,(** 2009). switching of H2O2 Hydrogenation in the direct synthesis process. Science 323 (5917):1037-1041.

5- **Pera –titus,** (2004). Degredation of chlorphenols by means of advanced oxidation processes: ageneva review. Applied catalysis B. Environmental. 47(4): 219-256.

6- **Sean pritchelt, M. P.;(** 2007). Accidental ingestion of 35% hydrogen peroxide can J Gastroenterol . 21(10): 665-667.

7- **Gerhart Z., Ullmann'sw**.(1985).Encyclopedia of Industrial chemistry. 5th ed., vol. AI: Deerfield Beach , FL: VCH publisher, to present p. VA13 (89) 445.

8- **Jun wang and Ho kyungRyu** ,(2015) .The effect of momordicacharantia on obesity and lipodprofiles of mice fed a high – fat diet , Nutr Res pract ,a(5):489-495.

9- **Carralhol F,(**2001).Hydrogen peroxide production in mouse tissue after acute damphetamine administration influence of mono amine oxidase inhibition , Arch Toxicol,75(8):465-9.

10- **NiethammerP,** ,(2009). Atissue-scale gradient of hydrogen peroxide media tesrapidwonnd detection in zebra fish ,Nature 459:996-999.{pubMed/NCBI}.

11- **Jun wang and Ho kyungRyu** ,(2015) .The effect of momordicacharantia on obesity and lipodprofiles of mice fed a high – fat diet , Nutr Res pract ,a(5):489-495.

12- **Friendman J.M. and Halaas J.L**(1998). Leptin and the regulation of of body weight in mammals .Nature 395-p.763-70.

13- **Lagiou P,** ,(1991). Hormonal, life style , and dietary factors in relation to leptin among elderly men – Ann Nutr Metab.43:23-29.{pub Med}.

14- **Martin S,** ,(2008).Leptin resistance :a possible inter face of inflammation and metabolism in obesity . related cardio – vascular disease . J Am coll cardiol,52 :1201-1210.

15- **khan** (2017) . The effect of caffeine on the body weight of Blab/Mice,Vol.67,Issne.2,p:287-291,3charts,1graph.

16- **Sheridan A.M,** ,(1996).Lipid peroxidation contributes to hydrogen peroxide induced cytotoxicity in renel epithelial cells ,kidney lnt , Jan ; 49(1):88-93.

17- **Parikh N.H** (2014).Indigenonus plant medicines for health care. Treatment of Diabetes mellitus and hyper-lipidemia.chinJNat Med.12:335-3444.{pubmed}.

18- **Yoon J.H,** (2010). Reaction of ferritin with H2O2 induced lipid peroxidation ,BMB Reb.43(3):219-24.

19- **Uto-konoH,** ,(2010).Coffee consumption enhances high –2002 Effect density lipoprotein –mediated cholesterol efflux in macro phages , circ 5;106(4):779-87.

20- **Nehlig A.**,(2016).Effect of coffee , caffeine on brain health and disease : what should I tell my patient? PractNeurol, 16(2): 89-59.

21- **Olfat Nassar M** ,(2013). Different Doses of Arabic caffee improve serum lipid profile , uric acid and liver enzymes of Experimental Rats, Food and Public Health ,3(4):228-233.

22- **ZangarA,** (2013).The effect of acute café latte ingestion on fasting serum lipid levels in healthy individuals .J clinLipidol ;7:165-168.

23- **Jansen D.F**,(1 995).Coffee consumption ,alcohol use ,and cigarette smoking as determinants of serum total and HDL cholesterol in two Ser cohorts of the seven countries study .ArteriosclerThrombvase Biol.15:1793-1797.

24- **Kazuoko BAYASHI-HATTORI**, (2005) . Effect of caffeine on the body fat and lipid metabolism of Rats fed on High –fat diet . Biosci .Bioteclinol .Biochem-69(11):2219-2223.