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#### MOLECULAR BIOLOGICAL STUDIES ON THE EFFECT OF GREEN TEA ON OBESE RATS

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#### ABSTRACT

The current study was conducted on 70 male albino rats for elucidation the role of different green tea either local (E) or imported (C) and capsulated like green tea (CAP) as well as two common active principle Epigallocatechin gallate (EPI) and polyphenols (POL) on high fat ration (HFR) fed rats. The results showed that EPI and POL were showing a significant improvement in lipid profile and anti-oxidant status of rats. Meanwhile, EPI and CAP groups revealed the highest improvement in kidney function, while the Egyptian, Chinese green tea leaves and POL groups showed a significant enhancement in hepato-billiary system. All the studied groups revealed a significant improvement in cardiac function. Moreover, MEST relative expression increased in HFR group and showed a significant down-regulation in all green tea groups but EPI revealed the lowest relative expression in MEST which suggested its effect on improving obesity in rats.

Key words: MEST, Epigallocatechin gallate, polyphenols

#### **INTRODUCTION**

Obesity is considered as a serious health problem that increased in both developed and developing countries all over the world (**Cunha et al., 2013**). The imbalance between food consumption and the energy loss was considered an important factor implicated obesity problems with the presence of environmental and genetic factors (Apovian and Mechanick, 2013).

The implications of obesity and its role in jeopardizing human race drew a lot of attentions as it is considered the sixth most important risk factor for several diseases problems affecting human beings (Badran and Laher, 2011).

Several techniques are used for preventing obesity through surgical intervention to dietary restriction with the administration of some pharmaceuticals drugs (Sae-tan et al., 2011). However, the use of both surgical intervention and pharmaceutical compounds would result in severe health hazards that might reach to death in several occasions. Therefore, the need to find natural sources to reduce body weight and preventing obesity is a major aim of several researchers due to low cost and high safety margin which could match different variety of obese persons taste (Poulose et al., 2005).

Different medicinal plants were used extensively for preventing obesity and encouraging weight loss (**Rayalam et al.**, **2008**). One of the most popular medicinal plants is green tea that derived from *Camellia sinensis* which is characterized by the presence of polyphenols that is used extensively in several research studies (**Yang and Hong**, **2013**). The presence of asepigallocatechin (EGC), epigallocatechin-gallate (EGCG), and epicatechin (EC) in green tea are also revealing a high efficiency in lowering body weight in several animal models through inhibition of lipogenesis and stimulation of oxidation of fatty acids (Wolfram et al., 2006). Green tea is expansively used in several research works as antioxidant, neuroprotective, anti-arthritic, antiangiogenic, antibacterial, antiviral and antiinflammatory agent (Henning et al., 2014).

Mesoderm specific transcript (MEST) is considered as a biomarker for obesity due to its expression in postnatal phase in fat mass expansion (Nikonova et al., 2008). The gene expression of MEST was up-regulated in white adipose tissue (Koza et al., 2009). Several dietary intervention was used to decreased obesity in animal and human, however the studying of MEST gene expression after green tea supplementation was require further investigation

Therefore, this study aimed to evaluate the use of different sources of green tea in local markets, imported markets and capsules-like green tea extract in preventing obesity in obese rats as well as to held a comparison between them and two important active principles in green tea ,polyphenols and epigallocatechingallate, in separate groups to evaluate their potency in preventing obesity. In addition, the use of Mesoderm Specific Transcript as a biomarker for obesity in the high fat fed diets of rats.

#### **MATERIAL AND METHODS**

#### Chemicals:

Liquid soft-gel capsules of green tea were supplied from Applied nutrition<sup>TM</sup>, Los Anglos, USA . Green tea imported packets were supplied from Fujianxiamentiantianxiang tea products lines<sup>TM</sup>, China . Local imported green tea packets were purchased from Ahmed green tea<sup>TM</sup>, Egypt. Epigallocatechin gallate (EGCG) was bought from Sigma Aldrich, USA under catalog number of (E4143-50MG). Polyphenon 60 powder extract from green tea was bought from (Sigma Aldrich, USA) under product number of (P1204-25G).

#### Animals and experimental procedures:

A total number of 70 male albino rats were bought from laboratory animals' research center, faculty of veterinary medicine, Benha University where there weight ranged from 200 to 250 grams. The rats were housed at separate wire mesh cages, exposed to good ventilation, humidity and to 12-hour light-dark cycle. They were given daily a constant supply of clean source of water and standard pellet diet. The rats subjected to the current study were housed at Nile Center for Experimental Research, Mansoura, Egypt for 2 weeks to acclimate the new environmental conditions. The rats were divided into seven equal groups where all the rats were received a high fat diet for induction of obesity except Control group (CI). The normal diet for rats and the high fat rations were prepared in table (1) according to NRC, 1995

The first group of this study received a normal diet and named as (C1). The second group called high fat ration (HFR). The third and fourth groups were received commercial green tea diet of Chinese green tea powder (HFR-C) and Egyptian green tea powder (HFR-E), respectively with high fat diet. The fifth group was received a high fat diet with Epigallocatechin gallate (HFR-EPI), while the sixth group received polyphenol (HFR-POL) with high fat diet. Finally, the seventh group were fed on high fat diet and orally supplemented with green tea capsules (HFR-CAP). All groups except (C1) were received a high fat ration for 8 weeks, then a normal diet was given for them for another 8 weeks. In HFR-C and HFR-E groups, the required dose was estimated according to the concentration of Epigallocatechin gallate in the powder form of tea where each 1 g contained 46.4% of Epigallocatechin gallate (Lu et al., 2012). Therefore, one gram from each green tea powder was soaked in 100 ml warm water and 2 ml from each type was given orally every day for 8 weeks by stomach tube. In HFR-EPI and **HFR-POL** groups, 464 mg of both epigallocatechin gallate and polyphenol were added to 100 ml of distilled water, and then 2 ml of the mixture was given daily for 8 weeks. In the last group, HFR-CAP, a daily dose of 0.75 ml contained 0.5% of epigallocatechin gallate were administrated orally for 8 weeks.

After completing the experimental protocol, the rats in all seven groups were weighted and anesthetized with thiopental sodium according to the procedure of **Singh and Boyd**, (1966). 5 to 10 ml of blood was drawn from the heart through cardiac puncture for serum separation that had been stored at -20°c till the analytical methods were performed and the sedated rats were sacrificed by head dislocation for the completion of dissection procedures. The rats were dissected under complete sterile condition for collecting of liver sample.

#### ANALYTICAL METHODS

#### Serum lipid profile

The collected serum samples of all groups were used to determine serum lipid profile. Serum total cholesterol (Allain et al., 1974), while serum triacylglycerol (Fossati and Prencipe, 1982). The determination of serum HDL-cholesterol (Lopez – Virella, 1977). The calculation of both LDL-cholesteol and VLDL was calculated according to the equation adapted by Friedewald et al. (1972).

The concentration of serum total lipid was estimated by the work of **Tietz**, (1961).

## Antioxidant defense system and oxidative stress markers:

The determination of antioxidant system in the collected liver samples was crucial for this study. Liver samples were homogenized according to the method of Fernandez et al., (1985). The homogenized liver samples were used to determine of Glutathione-S- transferase (GST) activity (Habig et al., 1974). The use of both the method of Beutler et al., (1963) and Aebi, (1984) in the determination of both reduced glutathione concentration (GSH) and catalase activities, respectively in liver of rats. The technique of **Drapper and Hadley**, (1990) used for the determination was of Malondialdehvde (MDA) concentration in liver and serum samples of rats. Finally, total antioxidant activity (TAC) was also determined in serum of rats (Koracevic et al., 2001).

#### Kidney function test

The determination of serum urea, creatinine and uric acid was done according to the protocol of Kaplan, (1969), Heinegård & Tiderström (1973) and Fossati et al., (1980), respectively.

#### Liver function test

bilirubin Serum concentration was measured spectrophotometrically (Walter and Gerarde, 1970), while serum glutamate oxaloacetate transaminases (sGOT) and glutamate pyruvate transaminases (sGPT) activities ( Reitman and Frankel 1957). Serum total protein and albumin (Gonall et al. (1949) and Doumas 1971), respectively. The protocol of EL-Aaser and EL-Merzabani (1975) was used to determine the activity of serum alkaline phosphatase (ALP).

#### **Cardiac function test**

The use of both creatine phosphokinase (CKMB) and lactate dehydrogenase (LDH) for the determination of cardiac function test (**Hess et al., (1964)** and **Koh & Choi 1987),** respectively.

## Mesoderm Specific Transcript gene expression (MEST)

RNA was extracted from liver by using Trizol reagent (Invitrogen, Life Technologies, NY, USA) according to the instruction manual. RNA integrity and concentration was checked using Nano drop spectrophotometer (Implen, USA). RNA template was used to synthesize cDNA using (RevertAid First Strand cDNA Synthesis, Thermoscientific) with MMLV reverse transcriptase enzyme and then the amplification of **c**DNA with LuminarisHiGreenqPCR Master Mix. Thermoscientific using the following PCR cycling conditions: Initial denaturation at 94°c for 9 minutes that followed by 35 cycles of denaturation at 94 °c for 40 seconds, annealing step at 58 °c for 30 seconds and a final extension step at 72 °c for 20 seconds. The primer used in the current study was listed in Table (2).

#### Statistical analyses:

All the measured biochemical parameters were analyzed using one way ANOVA at a significant level of 0.05. The statistical analysis was performed with SPSS V.17 and the data was represented by the mean  $\pm$  standard error of mean. The gene expression analysis of mesoderm specific transcript gene was analyzed with 2<sup>- $\Delta\Delta$ ct</sup> method according to the method of **Pfaffl, 2004** in comparison with rat  $\Box$  actin

#### RESULTS

### Final body weight of rats after different green tea supplementation:

The Chinese green tea leaves as well as HFR-EPI and HFR-POL significantly reduced body weight in comparison with HFR group (Table 3)

#### Serum lipid profile:

In Table (4), HFR-EPI and HFR-POL produced a sig decrease in blood cholesterol, serum triglycerides and serum total lipids significantly in comparison with C1and HFR. On the other side, HFR-E showed the highest concentration of HDL that significantly increased in comparison with C1 and HFR

Antioxidant defence system and oxidative stress marker in obesity induced rats. All the treated groups with either tea leaves or green tea extract showed a significant enhancement of GST, GSH, CAT and TAC. However, HFR-EPI and HFR-POL revealed the highest improvement of anti-oxidant defence system. Moreover, all studied groups showed a decline of lipid peroxidation level in obese rats. (Table 5)

## Kidney and liver function test in obesity induced rats:

Considering kidney function test, HFR-EPI and HFR-CAP decreased serum creatinine and urea significantly in comparison with HFR. The Egyptian green tea leaves and HFR-EPI group were showing a significant decreased in serum bilirubin, while HER-E and HFR-POLgroups were significantly increased serum albumin. Neither of all studied group improved liver transaminases. In the same respect, HFR-C and HFR-POL significantly depressed the activity of alkaline phosphatase (Table 6)

#### Cardiac function tests:

The results of this study indicated that all green tea leaves and its active principles significantly decline CKMB and LDH except HFR-E. (Table 7).

#### **MEST gene expression:**

The MEST relative expression revealed the presence of a significant decline in MEST in all studied groups; however HFR-EPI revealed a significant decrease among all studied groups

Food constituent	Normal diet/one kg	High fat ration /one Kg
Crushed Yellow Corn	0.40 Kg	0.35 Kg
Wheat Flour	0.25 Kg	0.20 Kg
Soya bean meal	0.08 Kg	0.08 Kg
Milk powder	0.07 Kg	0.07 Kg
Wheat bran	0.08 Kg	0.08 Kg
Crushed Hay	0.10 Kg	0.05 Kg
Mineral and vitamins premix*	2.50 g	2.50 g
Salt	8.75 g	8.75 g
Lime stone	8.75 g	8.75 g
Animal butter	-	0.15 Kg

Table 1: The formulated diet for induction of obesity in rats

\*Each **1 kg** of diet contains: vit A= 10000IU, Vit D<sub>3</sub>= 1800 IU, Vit E= 8.3 mg, Vit K= 1.6 mg, Vit B<sub>1</sub>=0.8 mg, Vit B<sub>2</sub>= 4.1, Vit B<sub>6</sub>= 1.25 mg, Vit B<sub>12</sub>= 0.008mg, Niacin= 25 mg, Biotin= 0.04 mg, folate= 0.8 mg, pantothenate= 8.3 mg, Mn= 49.8 mg, Fe= 24.9 mg, Cu= 3.2 mg, I= 0.8 mg, Se= 0.08 mg, Co= 0.08 mg, Zn= 41 mg

**Table 2:** The primer sequence of Mesoderm specific transcript and Rat  $\beta$  actin which kept as a house keeping gene for gene expression analysis:

Name	sequence	Amplicon size (bp)	Accesion number	
Mesoderm specific transcript	F: AGAATCGTTCTGGCCGTCTC R: CCCGTCATTGTTGCGAATCC	251 bp	NM_001009617	
Rat β actin (house keeping gene)	F/TCCTCCTGAGCGCAAGTACTCT R/GCTCAGTAACAGTCCGCCTAGAA	116 bp	V01217	

Group	Final body weight
C1	$199 \pm 7.55^{acdfg}$
HFR	$413\pm17.31^{bdeg}$
HFR-C	$320 \pm 24.76^{\text{aef}}$
HFR-E	$371 \pm 24.13^{beg}$
HFR-EPI	$265 \pm 42.62^{acf}$
HFR-POL	$302 \pm 15.03^{\rm acef}$
HFR-CAP	$384\pm32.03^{bdeg}$

 Table 3 : Final body weight of rats after different green tea supplementation.

Data expressed as mean  $\pm$  SEM. a significant vs HFR-CAP, b significant vs HFR-EPI, c significant vs HFR-E, d significant vs HFR-C, e significant vs C1, f significant vs HFR, g significant vs HFR-POL. Values are mean  $\pm$  standard error of mean where P<0.05.

Group	Cholesterol	Triacylglycerol	HDL	LDL	VLDL	Total lipids
C1	$64.96{\pm}4.01^{bf}$	46.32±2.7 acdf	31.67±3.73 <sup>abcg</sup>	$20.27{\pm}2.04$ <sup>adf</sup>	$10.71 \pm 0.33$ f	$706\pm60^{\text{ acdf}}$
HFR	77.53±5.67 bdeg	98.41±2.37 bdeg	31.58±3.15 abcg	26.22±1.54 abcdeg	27.99±3.82 abcdeg	1422±36.5 abcdeg
HFR-C	59.97±3.09 acf	71.16±5.69 <sup>ef</sup>	33.06±4.47 <sup>abcg</sup>	13.18±1.75 aefg	15.81±1.06 <sup>f</sup>	916±15.19 cef
HFR-E	71.86±1.73 <sup>bdg</sup>	81.92±11.38 beg	42.88±2.96 abdefg	16.43±1.74 <sup>af</sup>	16.15±2.29 <sup>f</sup>	1180±118.41 abdefg
HFR-EPI	51.34±0.44 acef	53.66±0.45 acf	19.23±1.3 <sup>cdef</sup>	16.70±3.51 afg	10.73±0.09 <sup>f</sup>	887±19.38 <sup>cf</sup>
HFR-POL	57.83±3.5 <sup>acf</sup>	55.63±2.26 acf	20.82±1.23 <sup>cdf</sup>	19.18±1.51 adf	15.55±1.99 <sup>f</sup>	826±23.01 <sup>cf</sup>
HFR-CAP	73.23±1.37 <sup>bdg</sup>	84.09±7.42 beg	20.2±2.15 <sup>cdef</sup>	33.98±1.87 <sup>bcdefg</sup>	14.23±1.33 <sup>f</sup>	919±34.93 cef

Table 4: Serum lipid profile in obesity induced rats.

Data expressed as mean  $\pm$  SEM. a significant vs HFR-CAP, b significant vs HFR-EPI, c significant vs HFR-E, d significant vs HFR-CAP, e significant vs HFR-POL. Values are mean  $\pm$  standard error of mean where P<0.05.

 Table 5: Antioxidant defense system and oxidative stress marker in obesity induced rats.

Group	GST	GSH	Catalase	MDA	TAC
C1	$10.83 \pm 0.39$ abcdfg	$24.16 \pm 1.76^{acf}$	2.62±0.16 abcdfg	98.58±3.48 <sup>cdfg</sup>	1.52±0.1 <sup>f</sup>
HFR	2.3±0.27 <sup>abcdeg</sup>	4.99±0.15 bdeg	0.17±0.02 bcdeg	268±11.83 <sup>abcdeg</sup>	0.87±0.06 <sup>abdeg</sup>
HFR-C	7±0.24 <sup>ef</sup>	17.24±3.16 <sup>f</sup>	1.94±0.3 <sup>abefg</sup>	35.65±6.22 aef	1.6±0.14 <sup>cf</sup>
HFR-E	$5.31 \pm 0.85^{aef}$	9.09±2.79 beg	1.63±0.22 aefg	50.50±23.11 aef	1.11±0.07 <sup>d</sup>
HFR-EPI	4.90±0.73 <sup>aef</sup>	23.36±2.5 <sup>acf</sup>	1.18±0.11 <sup>adef</sup>	$73.68 \pm 10.09$ f	1.36±0.06 <sup>f</sup>
HFR-POL	5.92±0.32 <sup>ef</sup>	24.98±5.66 acf	1.02±0.23 acdef	56.05±12.9 <sup>ef</sup>	1.43±0.15 <sup>f</sup>
HFR-CAP	7.69±1.15 <sup>bcef</sup>	12.44±2.25 beg	0.49±0.02 bcdeg	89.55±3.5 <sup>cdf</sup>	1.36±0.26 <sup>f</sup>

Data expressed as mean ± SEM. a significant vs HFR-CAP, b significant vs HFR-EPI, c significant vs HFR-E, d significant vs HFR, c significant vs HFR, g significant vs HFR-POL. P<0.05.

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Group	Urea	Uric acid	Creatinine	Bilirubin	Albumin	Total protein	SGPT	SGOT	ALP
C1	15.86±1.31 <sup>f</sup>	1.12±0.75 acfg	$0.19{\pm}0.04^{\text{ f}}$	$0.05{\pm}0.01$ bef	3.16±0.05 afg	7.49±0.17 af	$90.25{\pm}1.74$ <sup>abfg</sup>	178±2.07 bd	388±115.64 acdg
HFR	23.43±1.62 abceg	3.86±0.49 bdeg	0.46±0.05 abde	0.41±0.07 bce	2.35±0.38 cdeg	9.72±0.62 abcdeg	44.49±0.54 bcde	148±11.62 ab	548±92.74 <sup>dg</sup>
HFR-C	18.59±2.15 ab	$2.34{\pm}0.27$ f	$0.23{\pm}0.05^{\text{ f}}$	0.27±0.03 bc	3.04±0.24 cfg	$6.49 \pm 0.42^{\text{ f}}$	84.54±23.47 <sup>abfg</sup>	122±17.69 abce	123±75.14 acef
HFR-E	$14.58 \pm 2.02^{\text{ f}}$	3.07±0.36 °	0.30±0.12	$1.57 \pm 0.27$ abdefg	3.77±0.23 abdfg	7.12±0.52 <sup>f</sup>	88.63±7.65 <sup>abfg</sup>	193±16.46 bd	651±50.74 bdeg
HFR-EPI	11.08±2.26 df	2.38±0.25 <sup>f</sup>	$0.23{\pm}0.05^{\text{ f}}$	$0.86{\pm}0.2$ acdefg	2.89±0.06 cg	7.14±0.32 <sup>f</sup>	168±0.66 acdefg	401±0.66 acdefg	296±81.71 ac
HFR-POL	14.90±1.95 <sup>f</sup>	2.46±0.14 ef	0.29±0.07	0.31±0.07 bc	4.51±0.19 abcdef	7±0.43 <sup>f</sup>	50.72±5.69 bcde	148±11.73 ab	105±14.22 acef
HFR-CAP	11.76±2.36 df	2.97±0.42 °	$0.26{\pm}0.05^{\text{ f}}$	0.32±0.03 bc	2.38±0.21 ceg	5.92±0.25 <sup>ef</sup>	43.20±2.01 bcde	201±26.23 bdfg	660±132.47 bdeg

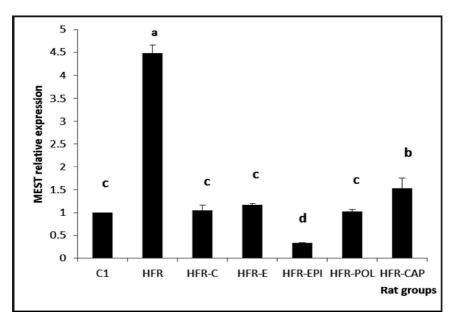
Table 6 :Kidney and liver function test in obesity induced rats.

Data expressed as mean  $\pm$  SEM. a significant vs HFR-CAP, b significant vs HFR-EPI, c significant vs HFR-E, d significant vs HFR-C, e significant vs C1, f significant vs HFR, g significant vs HFR-POL. P<0.05.

Table 7: Cardiac function tests.

Group	СКМВ	LDH
C1	74±1.79 abcfg	1218±252.12 <sup>adg</sup>
HFR	381±12.1 abcdeg	1775±193.9 <sup>abdg</sup>
HFR-C	113±7.39 acfg	443±105.55 <sup>cef</sup>
HFR-E	167±14.95 <sup>adefg</sup>	1470±347.04 abdg
HFR-EPI	170±29.25 <sup>aefg</sup>	781±26.41 <sup>cf</sup>
HFR-POL	264±14.98 bcdef	564±71 <sup>cef</sup>
HFR-CAP	266±33.25 bcdef	625±10.09 cef

Data expressed as mean ± SEM. a significant vs HFR-CAP, b significant vs HFR-EPI, c significant vs HFR-E, d significant vs HFR-C, e significant vs C1, f significant vs HFR, g significant vs HFR-POL. P<0.05.



#### DISCUSSION

Obesity was induced by ingestion of large amount of fat in diet with the lack of physical exercise that would eventually caused liver impairment and cardiovascular diseases which might end with death (Murase et al., 2002).

In several occasions, green tea was thought to be efficiently reduced body weight for rats but the type and the dose of the given green tea showed a great variation in their capability in reducing weight. The obtained data revealed that only Chinese green tea polyphenols leaves. EGCG and were effectively capable of significantly decreased final weight of rats which was mainly attributed to the highest contents of catchins in green tea that caused an inhibition of the absorption of cholesterol and plasma levels, impair fatty acid synthesis and the sympathoadrenal system, having antioxidant function by reducing the LDH and decreasing the expression of adhesion molecule (Hernandez Figueroa et al., 2004).

In the current study, an improvement in lipid profile was observed after the administration of EGCG and polyphenol especially which appeared by the reduction in serum cholesterol, triglycerides and total lipid. On the same side, an improvement in the levels of HDL was occurred after administration of HFR-E. This result was supported by the current study performed by Raederstorff et al., 2003; Bakr and Header, (2014) who found that green tea extract was responsible for the decrease absorption of triacylglycerol cholesterol and with the increase in fat excretion. Moreover. Hussein et al., (2011) added that both catechins and

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polyphenols possessed a hypolipidemic effect through decreasing LDL with a constant increase in HDL.

The treated groups of rats treated with either green tea leaves or its extract achieved a significant increase in GST, GSH, CAT and TAC and significant suppression in lipid peroxidation. In the same respect the use of both HFR-EPI and HFR-POL showed the highest significant improvement in antioxidant defense system. The potency of EGCG was suggested due to the binding of thiol group of glutathione with its molecular structure that was further oxidized resulting in an increase in its potency (Sang et al., 2005). On the other side, polyphenols were capable in increase the stage II anti-oxidant such as glutathione- S- transferase and glutathione peroxidase in rodent liver that illustrated the increase of anti-oxidant enzyme system (Lee et al., 1995). Green tea leaves and its extract significant showing a decrease in lipid peroxidation that was indicated by the decrease of MDA concentration which was also an observed result detected by Coimbra et al., (2006) and Haidari et al., (2013).

An improvement of renal function was observed after supplementation of both HFR-EPI and HFR-CAP as well as the Chinese imported green tea leaves which was also observed by **Sano et al., (1995)** who attributed the improvement of renal function due to the enhancement of anti-oxidant status of renal tissues by the presence of EGCG in green tea. Moreover, **Choi et al.,(2004)** suggested that the content of catchin (Polyphnols and EGCG) in green tea was as an anti-inflammatory agent that regain kidney function after high fat diet supplementation. The Egyptian green tea leaves and EGCG were showing a significant decrease in the concentration of serum bilirubin suggesting that the levels of EGCG in Egyptian leaves was sufficient for the integrity of hepatobilliary system of obese rats only but the extended protection for hepatocelluar function of liver is limited which was suggested due to the dosage and duration of the experiment (Jin et al., 2008). Moreover, the supplementation with both Egyptian green tea and polyphenols would resulted in a significant improvement of serum albumin when compared with HFR group groups which was supported by the work of Gad and Zaghloul, (2013). Finally, a depression in the activity of ALP after green tea supplementation in HFR-E and HFR-POL groups which was suggested due to the ability of green tea in maintaining the integrity of heptaobilliary system through decreasing the incidence of bile stones formation (Zhang et al., 2006). In addition, polyphenols were further used in blocking surgical induced hepatic fibrosis through bile duct ligation surgery (Zhong et al., 2003).

The supplementation of green tea leaves and their extract would result in a significant decline of CKMB and LDH which suggested the protective effect for cardiac function in rats. However, the Egyptian green tea leaves showing a significant improvement in CKMB only and the Chinese green tea leaves only reduced the activity of LDH, which reinforced the idea of the potency of EGCG and polyphenols in enhancing the cardiac function of rats which was suggested due to the ability of catechins in impeding cardiac dysfunction through the improvement of brachial artery (Widlansky et al., 2007) and bronchial artery (Schroeter at al., 2006) blood flow. In general, the improvement of lipid profile and the stimulation of antioxidant status showed an important role in maintain cardiovascular function of rats supplemented with green tea (Shenouda and Vita, 2007).

MEST gene was up-regulated in high fat fed diet group which was started to downregulated significantly after supplementation with green tea. However, the rats group supplemented with EGCG was showing a significant decrease in MEST relative expression in comparison with other studied group. It was investigated by Voigt et al., (2015) that MEST was up-regulated after the ingestion of high fat diet and several dietary intervention was used for reduction of MEST gene expression which made the importance of MEST as a biomarker for obesity. From this point, it was considered that EGCG was an excellent active principle of green tea leaves which was indicated through the significant down-regulation of MEST gene expression

#### CONCLUSION

It can be concluded from the current study that chinese, EPI and POL caused a significant reduction in body weight. Moreover, EPI and POL can be used to improve anti-oxidant status and lipid profile of rats, while EPI and CAP would be a suitable candidate for improving kidney function. Local and imported green tea leaves as well as POL showed an important role in maintaining in hepato-billiary system obese rats. Furthermore, all studied groups fed on green tea revealed an excellent agent for improving cardiac function. Finally, **MEST** gene expression was thought to be an important marker for obesity which was revealed the lowest relative expression after supplementation with EPI.

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#### الملخص العربي

دراسات جزيئية بيولوجية عن تأثير الشاي الأخضر على الفئران السمينة

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خضع للدراسة التى بين أيدينا ٧٠ ذكرا من الفئران المهق للوصول لتفسير دور الشاى الأخضر المحلى أو المستورد والمنتج فى شكل أقراص، إضافة إلى تأثيرين شائعين أساسيين ونشطين للإبيجاللوكاتيشين الجال والبوليفينولات على الفئران التى تم تغذيتها بمعدلات دهون مرتفعة.

وقد أظهرت النتائج أن الإبيجاللوكاتيشين الجال والبوليفينولات كانا ذو تأثير كبير وملحوظ في تحسن مستويات الدهون وحالة مضادات الأكسدة في الفئران.

فى الوقت نفسه، أظهرت مجموعات الإبيجاللوكاتيشين الجال وأقراص الشاى الأخضر أعلى معدل تحسن لوظائف الكلى، بينما أظهرت مجموعات الشاى الأخضر المصرى والشاى الأخضر الصينى والبوليفينولات تحسنا ملحوظا فى لوظائف جهاز الكبد والمسالك البولية.

واظهرت كافة المجموعات التى تمت دراستها تحسنا ملحوظا فى وظائف القلب.

إضافة إلى ذلك، فقد سجل جين البروتين زيادة ملحوظة فى مجموعة الدهون مرتفعة النسبة وأظهر إنخفاضا كبيرا فى مجموعات الشاى الأخضر، إلا أن الإبيجاللوكاتيشين قد أظهر أقل نسبة لوجود جين البروتين مما يشير إلى فعالية تأثيره على تحسن نسبة السمنة فى الفنران.

الكلمات الدلالية: جين البروتين، الإبيجاللوكاتيشين الجال، البوليفينولات