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Blending and Characterization of Gold Nanoparticles with Omega-3 Oils Induces Antidiabetic, and Antioxidant Activities in-vivo

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THE aim of the study is to explore the antidiabetic and antioxidant activities of gold nanoparticles (AuNPs) mixed with either flaxseed oil (FLO) or fish oil (FIO). A pyrolysis method was used to prepare AuNPs in oleic acid. The characteristics of the sample were determined by using the transmission electron microscope (TEM) and UV spectroscopy. Male albino rats were divided into two main groups. Group one (n=10 rats) was fed on the basic diet (serves as healthy control). The other group was fed on the high fructose diet (HFD) for four weeks and then was divided into five subgroups. 1st subgroup was fed on HFD only, 2nd and 4th subgroups were fed on HFD mixed with omega-3 oils while 3rd and 5th fed on HFD+ omega-3 oils mixed with AuNPs. Results revealed that there was a significant decrease in glucose level, serum insulin, homeostasis model assessment of insulin resistance (HOMA-IR) index and lipid peroxidation in oils nanoparticles treated groups compared to the HFD group and a significant increase in the homeostasis model assessment of β-cell function (HOMA-β) and Glutathione (GSH). It can be concluded that AuNPs mixed omega-3 oils have more anti-diabetic, hepatic protection and antioxidant properties than omega-3 oils alone.

Keywords: Diabetes, Liver, Omega-3 oils, Gold nanoparticles.

Introduction

Gold nanoparticles (AuNPs) were applied for the treatment of different diseases because of their special optical, chemical, and biological effects [1]. Gold compounds may act as anti-inflammatory agents because they could inhibit NF-kappa B expression and consequent inflammatory reactions [2]. In addition they showed anti-oxidative effect [3] and did protect liver against high-fat diet by lowering liver enzymes [4]. However AuNPs do not have negative effect on the human body, and have a good safe guarantee in the human edible field [5].

Flaxseed oil (FLO) is the richest source of omega-3 fatty acids especially α-Linolenic acid (ALA). Fish oil (FIO) is also a rich source of omega- 3 fatty acids, especially Eicosapentaenoic acid (EPA) and Docosahexaenoic acid (DHA) [6]. FLO is obtained from the Linumusitatissimum seeds [7]. A dietary omega-3 fatty acid (EPA, ALA, and DHA) could have protected experimental animals against cardiovascular diseases (CVD), heart attack "as discussed by Yang et al [8]". Moreover, increasing omega-3 fatty acids' supplementation could have raised membrane fluidity, red blood cell membrane contents of α-and γ-Tocopherol [9].

The elevated blood glucose level was a characteristic of diabetes mellitus (DM). DM is considered as a developed, standard, dangerous,

expensive, potentially preventable general medical problem [10]. It is a common metabolic disorder of the three energy's supplements: fats, carbohydrates, proteins [11]. Shaw et al [12] defined diabetes mellitus as a group of metabolic disorders characterized by impairment of glucose metabolism regulation result from a defect in insulin secretion, diminished insulin responsiveness, or both. "As demonstrated by Gupta et al [13]" that the high fructose diet (HFD, 50-60% fructose), alloxan, streptozotocin commonly could stimulate diabetes in experimental animals. Fructose could cause insulin resistance of liver, pancreatic damage, metabolic degeneration in rats fed with HFD [14]. The current paper aimed to study the effects of adding flaxseed and fish oils (as rich sources of omega-3 fatty acids) combined with AuNPs on rats receiving high fructose diet.

Experimental Section

Instruments

TEM (JEOL-JEM 1200) was used to measure the AuNPs images. The TEM was working at a voltage equal to 90 kV. For the TEM estimations, a drop of the sample containing AuNPs was put on a copper grating enveloped with indefinite carbon. After enabling the film to stand for two minutes, the extra solution was expelled using a drying paper, and the grid was permitted to dry before the examination.

Materials and Chemical Reagents

FLO, FIO were obtained from Everline Company, 6-October City, Giza, Egypt. Fructose was purchased from El-Gomhouria Company for chemicals and drugs, Cairo, Egypt. Tetra chloroauric ($HAuCl_4$) and oleic acid were purchased from Sigma-Aldrich (Cairo, Egypt).

Preparation of AuNPs

About 10 ml of 5×10^{-3} mol / dm3 of HAuCl4 added drop by drop to 100 ml of oleic acid at 200 °C. Heating was continued for an additional 5 minutes after which, the solution removed from the heater and stirred for 15 minutes until, the color turned into orange-brown ("as discussed elsewhere [15-17]"and modified by Al-Sherbini et al [18].

Animals

60 healthy male albino rats "Sprague Dawley strain" were weighing (150 ± 10g); they were purchased from the animal colony, Helwan Farm, Vaccine and Immunity Organization, Helwan Governorate, Egypt. The rats were housed below

the basic conditions (12:12 h light: dark cycle and 22 ± 2 °C temperature) and supplied with the freshwater and the standard diet.

Experimental design

The experimental rats were kept individually in wire cages under standard and hygienic conditions. Food and water were supplied ad-libitum and checked every day. Rats consumed a standard diet for 7-days for adaptation after that; they were divided into two main groups. The first group (G1) (n = 10 rats) was fed on the healthy basic diet only for eight weeks as a negative control group [19]. The second group (n = 50 animals) was fed HFD (fructose 50%) for four weeks [20]. After determining glucose values, rats were divided into five subgroups (n = 10 rats/subgroup). The first subgroup (G2) was fed on HFD (50%) until the end of the experimental period as a positive control group. The 2nd subgroup (G3) and the 4th subgroups (G5) were treated with 10% omega-3 oils (FLO or FIO). The 3rd (G4) and the 5th (G6) subgroups were fortified with 10% omega-3 oils (FLO or FIO) mixed with 17 ppm AuNPs [18]. The animals were sacrificed and blood samples were collected after eight weeks. Liver of every animal was carefully removed and washed in saline (0.9%). A part of the liver was put in 10% formalin for histopathological tests and the other part was used for lipid oxidation (MDA), GSH determination. Blood was centrifuged at 4000 rpm (round per minute) for 10 min at 37 °C to separate serum and was put in a plastic vial at – 20 °C for future analysis.

Methods

Serum analyses

Fasting serum glucose was determined in all experimental animals [21]. Level of fasting insulin was evaluated in serum using the rat insulin Enzyme-linked immunoassay (ELISA) kit [22]; Homeostasis model assessment of insulin resistance (HOMA-IR) index; homeostasis model assessment of β-cell function (HOMA-β) were calculated as follow "as discussed elsewhere [23, 24]":

HOMA-IR = [fasting serum insulin (μIU / l) × fasting serum glucose (mmol / l)] / 22.5
HOMA-β = [20 × fasting serum insulin (μIU/ l)] / [fasting serum glucose (mmol / l) – 3.5].

Serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were determined by the method of Young et al [25]. Alkaline phosphatase (ALP) level estimated "as explained Tietz [26]".

Determination of oxidative and antioxidant Biomarkers in Liver tissue

Malondialdehyde (MDA) determination

Lipid oxidation as MDA reacts with thiobarbituric acid (TBA) in acidic medium and giving a pink-colored TBA-complex that could be measured by the calorimetric method [27].

Glutathione (GSH) determination

The assay of GSH was performed using the spectrophotometric method [28], which depends on the reduction of 5, 5'-dithiobis 2-nitrobenzoic acid with GSH to produce a yellow color whose absorption is measured at 405 nm.

Determination of Relative Liver Weight:

After eight weeks, the animals sacrificed. The liver carefully dissected out and weighed in grams (absolute weight). The relative liver weight (RW) of each animal then calculated according to Aniagu et al [29]:

"Relative Organ Weight = [Absolute organ weight (g) / Final body weight of rat (g)] × 100"

Histopathological examination

Tissues were prepared for the laboratory of dehydrated lower to higher alcohol degrees, filtered in Xylene and fixed in paraffin, paraffin blocks were ready and serially sectioned to four microns thick sections, then dyed with H& E for routine histopathological study [30].

Statistical analysis

The results were presented as mean ± SE. One way analysis of variance (ANOVA) and post hoc least significant difference analyses (LSD) were performed using the statistical package for social science (SPSS) version16 to compare between groups. The value of P < 0.05 was considered statistically significant [31].

Results and Discussion

It is well known that the optical absorption spectra of AuNPs originates from the surface Plasmon resonance (SPR) and the nanoparticles of 30-50 nm showed a peak in the 520-530 nm region [32]. Fig. 1 a, b showed the UV spectra and TEM image of the prepared AuNPs in oleic acid. Fig. 1a illustrated that the absorption spectrum for the solution of AuNPs is at 520 nm due to the SPR of AuNPs. The observed spectra at λ=970 nm might be due that most of the nanoparticles are spherical but some of them in heterogeneous in shape. While Fig. 1b presented the size and shape of AuNPs, they were measured by the TEM imaging. The particles were shown mostly spherical or triangle shapes,

it showed that the average size of the prepared AuNPs ranged from 30-50 nm.

Effect of omega-3 oils (FLO / FIO) and AuNPs on diabetes parameters

The results in Fig. 2 a, b, c showed that blood glucose, insulin and HOMA-IR of positive control group (G2; HFD 50%) were significantly higher than that of the healthy control group (G1).Whereas HOMA-β was significant less than that of the healthy control group (Fig. 2d). This could be explained with development of insulin resistance because of chronic utilization of HFD or the dropdown of β-cell function, because of the continuous burden put on β-cells to neutralize insulin resistance [33]. These findings were similar to results of "Mahfouz and colleagues [34]" which showed that the high flow of fructose to the liver disrupts metabolism and absorption of glucose and results in metabolic commotions that influence the induction of insulin resistance, a trademark of type-2 diabetes. When the insulin receptor range or its affinity decreased, imperfections at the level of molecules like glucose transfer (GLUT) and enzymes concerned into glucose metabolism occurred, the imperfection of insulin linking might take place which subsequently lead to insulin resistance [35].

The fasting glucose, serum insulin and HOMA-IR value of the omega-3 oils treated groups (G3, G5) are a highly significant (P < 0.0001) lower than that of HFD group (Fig. 2 a, b, c)while HOMA-β of these groups (G3, G5) is higher than that of G2 (HFD-group) (Fig. 2d). This confirmed "results of El-Khayat et al [36]; Chen et al [37]" who reported that abnormal pancreatic changes caused by diabetes were prevented using omega-3 supplementation. This might be as a result of either FLO supplementation which could up-regulate PPAR-γ gene expression in PBMC of diabetic patients with CHD "as discussed by Hashemzadeh et al [38]", or FIO could improve insulin sensitivity through elevation of plasma member fluidity and stimulation of existing β-cells of the islets of Langerhans for insulin synthesis [39]. Another suggestion that FIO might protect the pancreatic β-cells against free radicals damage produced in DM [40].

The groups treated with AuNPs and the omega-3 oils (FLO / FIO) had the lowest glucose level, insulin, HOMA-IR and also had the highest HOMA-β among the all groups fed HFD (G2: G6) (Fig. 2 a, b, c, d) but conversely, the glucose level was still far from the healthy level. The current findings "lined with Daisy

& Saipriya [41] reports" concluding that the use of AuNPs reduced glucose concentrations and induced appropriate body weight changes for the treatment of diabetic rats induced with streptozotocin; this may be because the AuNPs might increase fat glucose transporter-4 (GLUT4) [4, 42]. As, the GLUT4 glucose transporter is a major mediator of the removal of glucose from circulation and a key regulator of homeostasis of whole-body glucose [43].

Effect of omega-3 oils (FLO / FIO) and AuNPs on serum liver enzymes

As shown in Fig. 3 a, b, c the mean ALT, AST and ALP of HFD group were193, 396 and 438 U/L respectively compared with 66, 222, 281 U/L in control group (G1). These results confirmed the previous explanations that the injury of the hepatic cells changes their transport function and permeability of the membrane causing the leakage of those enzymes from the cells into the blood [44]. These data were also in coherence with other findings "reported by Ren et al [45]".

The FLO treated group (G3) had lower ALT, ALP (P < 0.01) (Fig.3 a, c) than that of G2, but no significant impact on serum AST level compared with G2 (Fig. 3 b). Whereas the group fed FIO (G5) had highly lower ALT and ALP (P < 0.0001) (Fig.3 a, c), moderately lower AST (P < 0.001) (Fig.3 b) than that of the HFD group. FIO groups that have high levels of EPA& DHA might protect the liver from steatosis, prevent NAFLD (Non-alcoholic fatty liver disease) and stimulate high levels of anti-inflammatory eicosanoids production [46]. Reduced activity of ALT, AST, ALP liver enzymes in the HFD with FIO group is also related to the hepatoprotective effect of FIO [47].

The AuNPs treated groups had highly significant lower liver enzymes (ALT, AST, ALP) (P < 0.0001) (Fig.3 a, b, c) than that of the group fed HFD only (G2). Moreover, there is no significant difference between ALP of G6 and that of G1 (Fig.3 c). These might be due to AuNPs could elevate anti-inflammatory cytokine interleukin-10 (IL-10) level [48]. Additionally, these results were in line with "Chen et al [4] who noted that AuNPs could reduce liver enzymes because AuNPs could protect liver against high-fat diet.

The hepatic oxidative status

MDA is the most important aldehyde compound producing from the oxidation of fatty acids content of LDL [49].

The results in Fig.4 a, b showed the elevation of hepatic oxidant activities (MDA) and the reduction of GSH levels (P < 0.0001) in HFD groups caused by increasing the fructose catabolism. These results conformed to results of "Hu et al [50]" who observed that chronic fructose consumption was resulting in a significant elevation in hepatic peroxidation of lipid (MDA) and a significant reduction in antioxidant enzyme activities (GSH) of the liver in high fructose-fed mice.

The mean MDA of FLO and FIO groups were 62 and 58 nmol / g respectively compared to 68 nmol / g of HFD group. These may be because of the free radical scavenger activity of omega-3 fatty acids (such as ALA, EPA, and DHA) [51]. "Han et al [52] demonstrated that" FLO could improve the antioxidant system, by raising GSH activity and reducing MDA level. This may be because FLO could stimulate the activity of glutathione reductase enzyme, which could counteract MDA as FLO contains phenolic acid and tocopherol; they are antioxidant components [53, 54].

FIO group's MDA and GSH were58 nmol /g and 2 µg / g respectively compared with 62 nmol / g and 1.6 µg / g of FLO group (Fig.4 a, b). These indicated that MDA of FIO was less than that of the FLO group, but FIO group has higher GSH than that of the FLO group. These were because the antioxidant protection of FIO supplements lowering the level of oxidative stress in both young and old rats [55]. These findings agreed with "Ciftcia et al [56] who investigated" the protective impact of FIO on cisplatin toxicity in the rat liver.

The AuNPs fortified groups had the highest GSH and the lowest hepatic MDA among the HFD groups (Fig.4 b, a). "Barathmanikanth et al [57]" disclosed the effective role of AuNPs as an antioxidative agent for lipid peroxidation and reactive oxygen species in diabetic mice. AuNPs are extremely capable of scavenging free radicals and have antitumor and cytoprotective actions [58] because GSH growing and reduction of MDA levels can protect non-tumor cells [59].

Histopathology and relative weight of the liver

The HFD group had the highest RW and was significantly different from the control group (see Fig. 5). Additionally, steatosis of hepatocytes, hepatic focal necrosis related to infiltration of inflammatory cells, fibroplasia in the portal triad and Kupffer cells activation observed in the control group HFD (Fig. 6b).
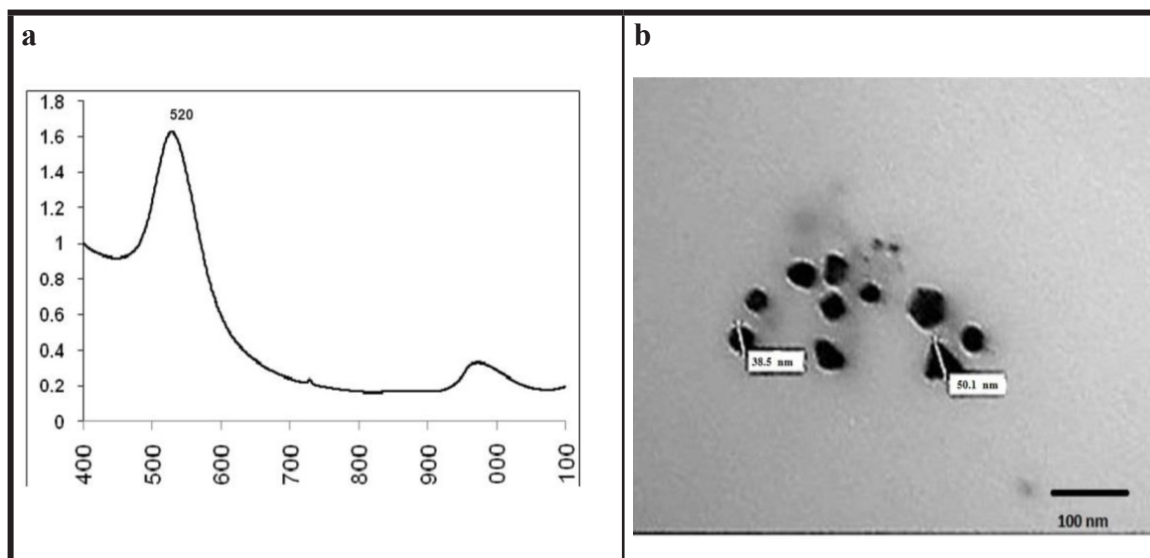
Fig.1. a) Absorption spectra of gold nanoparticles prepared in oleic acid, b) TEM image of gold nanoparticles.
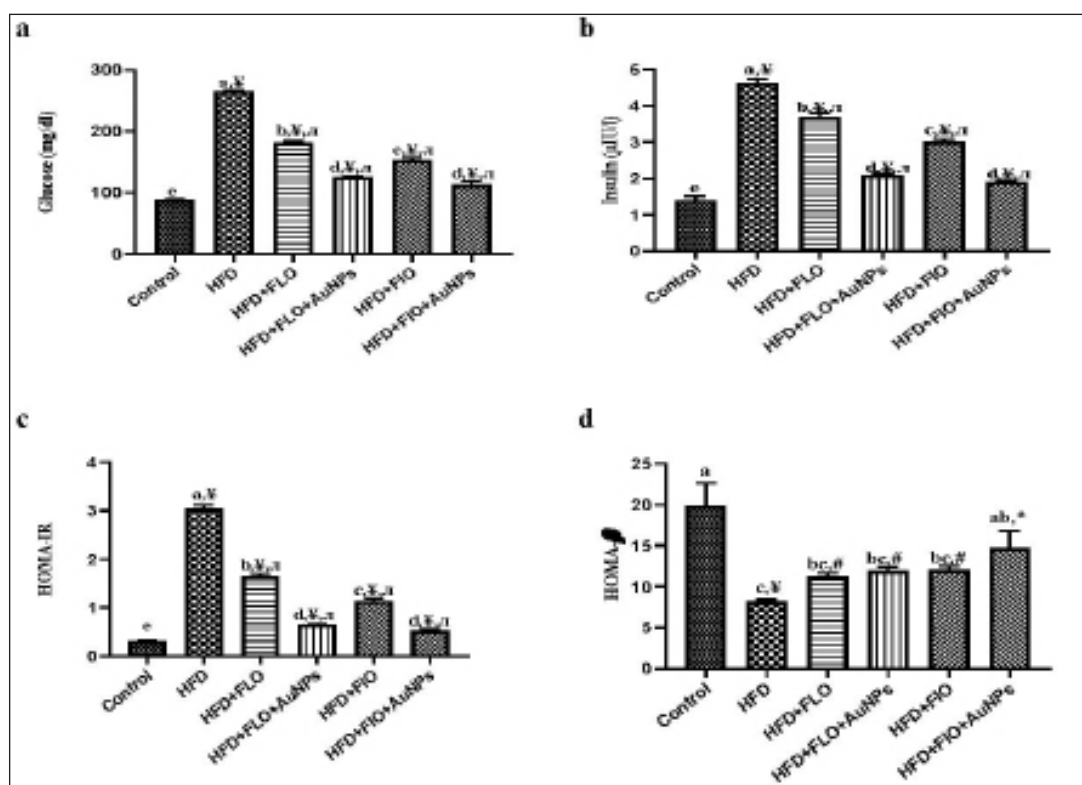

Fig. 2. Effect of omega-3 oils (FLO/ FIO) and AuNPs on a) serum fasting glucose (mg/ dl), b) serum fasting insulin (µIU/ ml), c) HOMA-β, d) HOMA-IR value in different experimental groups.

(a-e) Represents the mean value ± S.E. (n=10 rats / group), Means that do not share a letter are significantly different using One-way ANOVA. ($P < 0.05$)

(¥,#) Represents significant difference between control group and treated group using student's unpaired t-test, ¥ ($P < 0.0001$), # ($P < 0.01$).

(л,*) Represents significant difference between HFD group and treated group using student's unpaired t-test, л ($P < 0.0001$), * ($P < 0.05$).
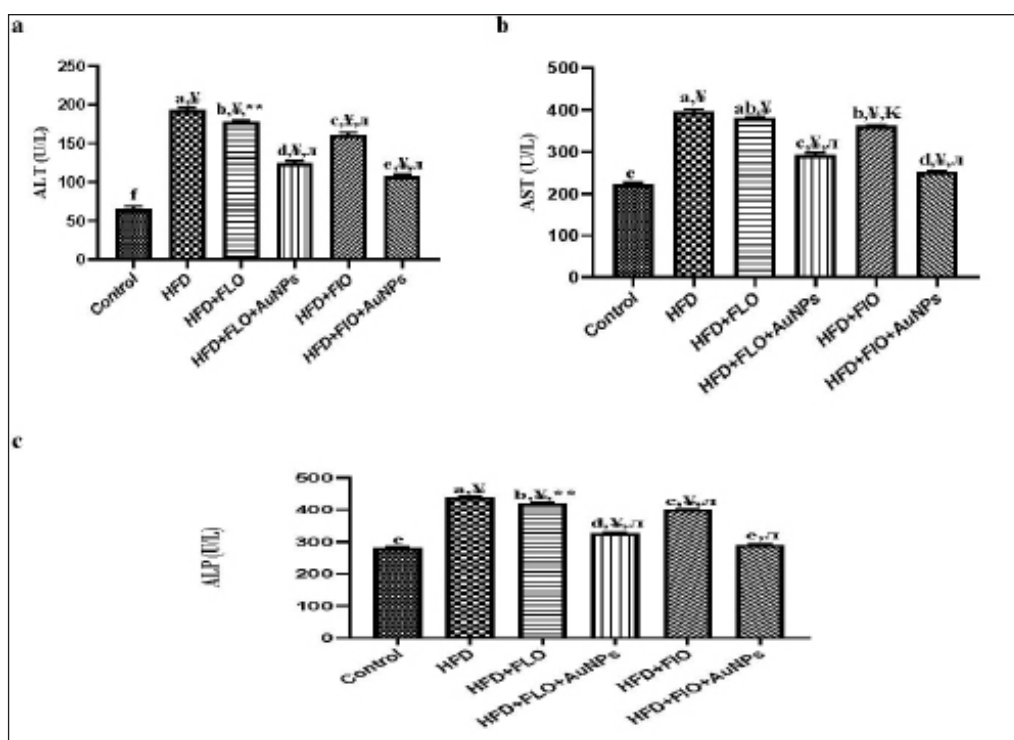
Fig. 3. Effect of omega-3 oils (FLO/ FIO) and AuNPs on a) ALT, b) AST, c) ALP in different experimental groups.
(a-e) Represents the mean value ± S.E. (n=10 rats / group), Means that do not share a letter are significantly different using One-way ANOVA. (P < 0.05) .
(¥, €) Represents significant difference between control group and treated group using student's unpaired t-test, ¥ (P < 0.0001),€ (P < 0.05).
(л,Ҝ, **, *) Represents significant difference between HFD group and treated group using student's un-paired t-test, л (P < 0.0001), Ҝ (P < 0.001), ** (P < 0.01) and * (P < 0.05).
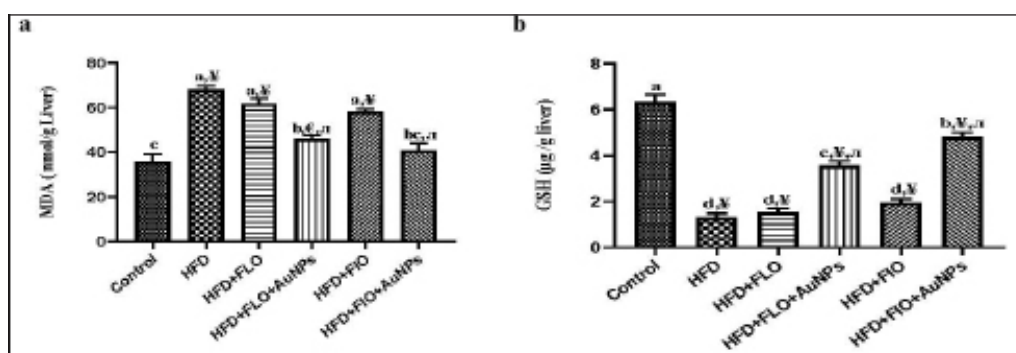

Fig. 4. Effect of omega-3 oils (FLO/ FIO) and AuNPs on oxidative liver state markers: a) MDA, b) GSH value in different experimental groups.
(a-d) Represents the mean value ± S.E. (n=10 rats / group), Means that do not share a letter are significantly different using One-way ANOVA. (P < 0.05)
(¥, €) Represents significant difference between control group and treated group using student's unpaired t-test, ¥ (P < 0.0001), € (P< 0.05).
(л, *) Represents significant difference between yeast group and treated group using student's unpaired t-test, л (P < 0.0001), * (P < 0.05).

This might be a result of large quantities of fructose is turned into triose-phosphates and fructose-1-phosphate. The basic aldolase-B and fructokinase enzymes concerned aren't controlled by hepatocyte energy status. Therefore, the majority of ingested fructose is metabolized to triose-phosphates within the liver, where these intermediates are oxidized and converted to lactate, glucose, or triglyceride (TG). Hepatic de novo lipogenesis made TG share in increased accumulation of hepatic lipids and plasma TG levels [60]. These results were lined with findings from "Svendsen et al [61]" who reported that the livers of Sprague-Dawley rats displayed morphological signs of steatosis with fat accumulation in hepatocytes two weeks after initiation of HFD. Then after five weeks, hepatocyte swelling was observed as a morphological character of early hepatic steatosis but after three months extremely elevation of the relative weight of liver in rats fed on HFD compared to those fed on a standard diet. Additionally, these results have been in harmony with results of "Leibowitz et al [62] who indicated that HFD caused clear changes of micro-vesicular steatosis but not noticed in the livers of his control group. Feeding rats a fructose-rich diet for six weeks induced metabolic syndrome with fatty liver [63].

The groups that fed HFD with omega-3 oils (FLO / FIO) showed a decrease in the RW of the liver compared with that of the positive control group (G2) as shown in Fig. 5. Liver of rats from G3 (HFD 50%+ FLO 10%) indicated steatosis of hepatocytes (Fig. 6c). Cytoplasmic vacuolation of hepatocytes and Kupffer cells activation (Fig. 6d) were observed in the liver of rats from group 5 (HFD 50%+ FIO 10%) suggesting omega-3 fatty acids have protective effects inhibiting eicosanoids and forming resolvins and protectins that were new biologically active lipid mediators with insulin-sensitizing, antisteatotic and adiponectin inducer effects [64]. These data were in coherence with "Han et al [65]" who concluded that FLO significantly diminished RW liver raised by high cholesterol and high-fat diet by reducing lipid drop in the liver of FLO mice. Omega-3 protects the liver or reduces liver injury caused by HFD through

improving homeostasis [66] and modifying derivatives of lipids formed by inflammation [67]. Omega-3 also activated Kupffer cells, by limiting liver cell damage which was caused by infection or increase clearance of pathogens and dead erythrocytes [68].

From Fig.5, it had been clear that G4 and G6 fortified with AuNPs made a significant decrease in the relative weight of liver (P < 0.0001) in comparison with the untreated group fed HFD (G2). Liver of rats from G4 (HFD 50% + FLO 10% + AuNPs) showed no histopathological changes (Fig.6 e). Meanwhile, the liver of rats from G6 (HFD 50% + FIO 10% +AuNPs) showed cytoplasmic vacuolation of hepatocytes and sinusoidal leukocytosis (Fig.6 f). Previous studies to understand the cytotoxicity of AuNPs showed that AuNPs did not show any toxicity compared to gold ions [69]. "Parveen et al [70] reported that" the administration of AuNPs didn't show any toxicity in activity day-to-day of female and male rats. Furthermore, the continuous intra-articular AuNPs administration doesn't have toxic effects on the internal organs (lungs, kidneys, spleen, and Liver) [71].

Conclusion

The present study showed that mixing between omega-3 oils (FLO / FIO) and AuNPs increase their antioxidant, anti-diabetic, hepatoprotective properties and is a better hypoglycemic agent for the treatment of DM and its associated complications. Its supplementation did not show any toxicity in the male rats' daily activity.

Conflict of interest

The authors declare that they have no conflict of interest regarding the publication of this paper

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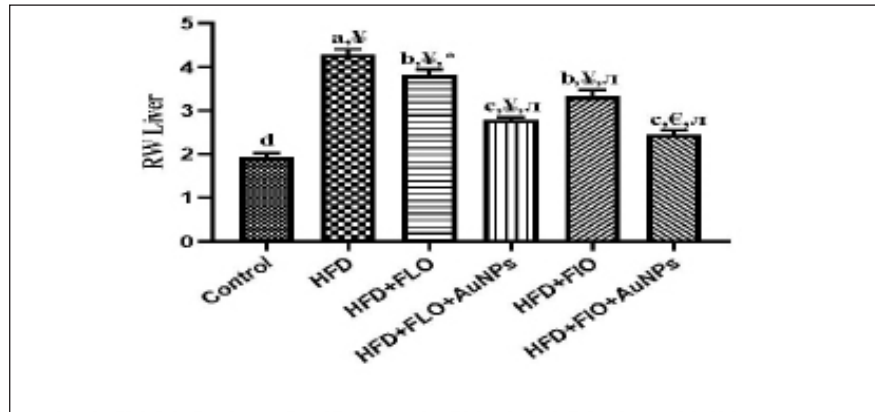
Fig. 5. Effect of omega-3 oils (FLO/ FIO) and AuNPs on relative weight of liver (RW Liver) in different experimental groups.

(a-e) Represents the mean value ± S.E. (n=10 rats / group), Means that do not share a letter are significantly different using One-way ANOVA. (P < 0.05)

(¥, €) Represents significant difference between control group and treated group using student's unpaired t-test, ¥ (P < 0.0001),€ (P < 0.05)

(л,Қ, **, *) Represents significant difference between HFD group and treated group using student's un-paired t-test, л (P < 0.0001), Қ (P < 0.001), ** (P < 0.01), * (P < 0.05).
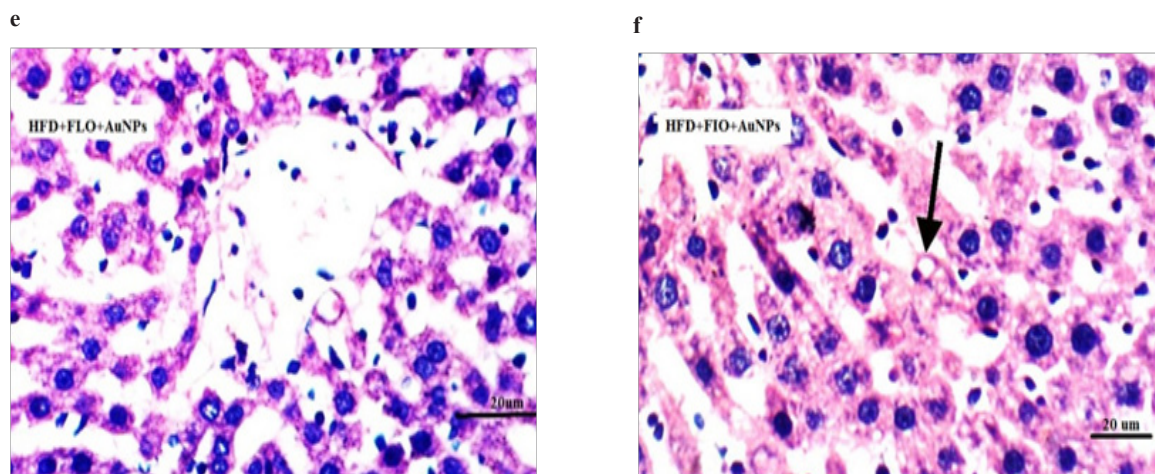
a

b

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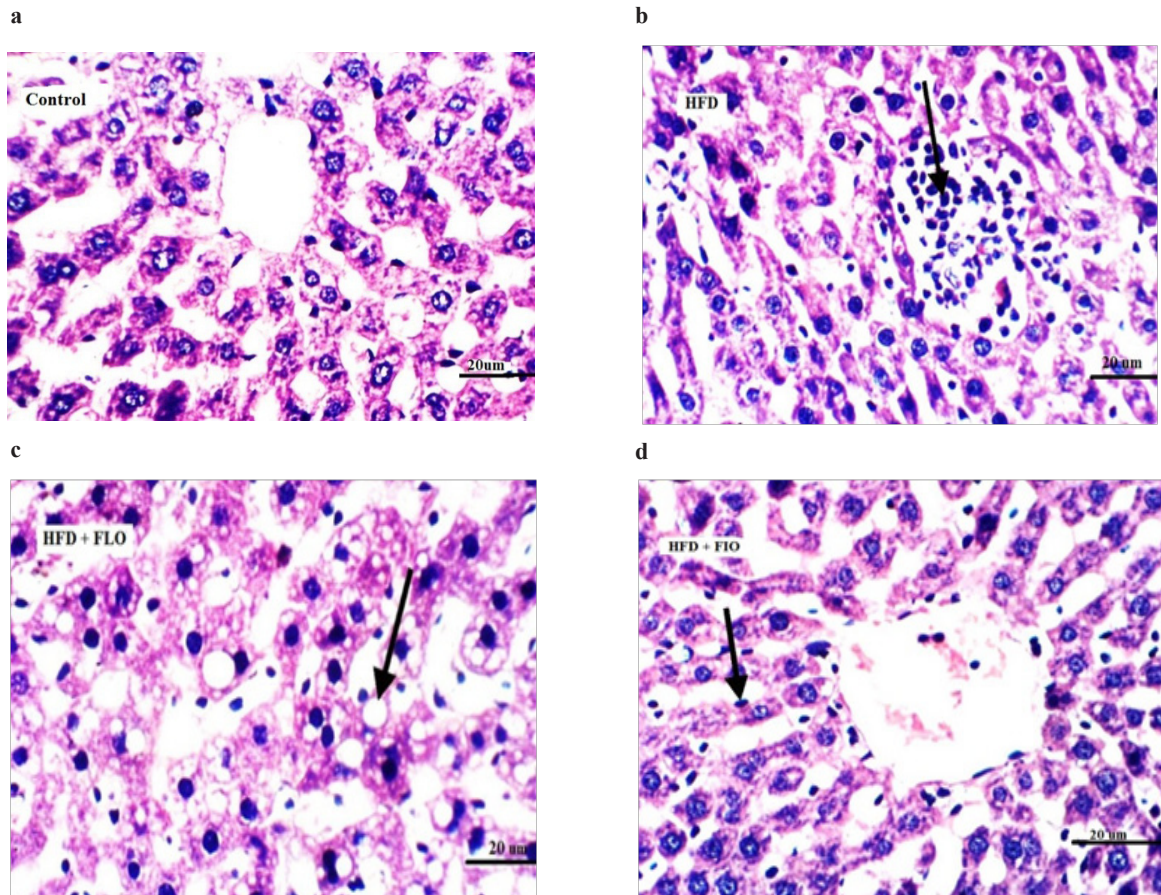
Fig. 6. TS of liver of a) normal control rats shows the normal architecture of hepatic lobule, b) HFD control rats' shows steatosis of hepatocytes, focal hepatic necrosis associated with inflammatory cells infiltration and fibroplasia in the portal triad and Kupffer cells activation, c) Rats fed HFD 50%+ FLO 10% shows steatosis of hepatocytes, d) Rats fed HFD 50%+ FIO 10% showed Cytoplasmic vacuolation of hepatocytes and Kupffer cells activation, e) Rats fed HFD 50%+ FLO 10% + AuNPs showed no histopathological changes, f) Rats fed HFD 50%+ FIO 10% + AuNPs showed cytoplasmic vacuolation of hepatocytes and sinusoidal leucocytosis . (H&E, scale bar 20.00μm, magnification ×400).

Abbreviations

AI: Antherogenic index	EPA: Eicosapentaenoic acid	NAFLD: Non-alcoholic fatty liver disease
ALA: α-linolenic acid	FIO: Fish oil	PBMC: Peripheral blood mononuclear cells
ALP: Alkaline phosphatase	FLO: Flaxseed oil	PPAR-γ: Peroxisome proliferator activated receptor gamma
ALT: Alanine aminotransferase	GLUT4:Glucose transporter-4	
AST: Aspartate aminotransferase	GSH: Glutathione	ROS: Reactive oxygen species
AuNPs: Gold nanoparticles	H&E: Hematoxylin and Eosin	rpm: round per minute
CHD: coronary heart disease		RW: Relative weight
CVD: Cardiovascular diseases	HFD: High fructose diet	SPR: Surface Plasmon Resonance
DHA: Docosahexaenoic acid	HOMA-IR: Homeostasis model assessment of insulin resistance	TEM: Transmission electron microscopy
DM: Diabetes Mellitus	HOMA-β: Homeostasis model assessment of β-cell function	
ELISA: insulin Enzyme linked immunoassay	MDA: Malondialdehyde	

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مزج وتوصيف الجسيمات الذهب النانويه مع زيوت اوميغا-3 يحفز الأنشطة المضادة للسكري ومضادات الأكسدة في الجسم الحي

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تهدف الدراسة إلى استكشاف الأنشطة المضادة لمرض السكرى ومضادات الأكسدة لجسيمات الذهب النانوية الممزوجة إما بزيت بذور الكتان أو زيت السمك. وقد استخدمت طريقة الانحلال الحراري لتحضير جسيمات الذهب النانوية في حمض الأوليك. و تم تحديد خصائص العينة باستخدام المجهر الإلكتروني و التحليل الطيفي للأشعة فوق البنفسجية.

تنقسم الفئران البيضاء الذكور إلى مجموعتين رئيسيتين:
المجموعة الأولى (العدد = ١٠ جرذان): والتى تم تغذيتها على النظام الغذائي الصحى.
المجموعة الثانية: التى تم تغذيتها بنظام غذائي عالي الفركتوز (HFD) لمدة أربعة أسابيع ، ثم تم تقسيمها إلى خمس مجموعات فرعية. تم تغذية المجموعة الفرعية الأولى على النظام الغذائي عالي الفركتوز فقط ، وتم تغذية المجموعات الفرعية الثانية والرابعة على النظام الغذائي عالي الفركتوز الممزوج بزيوت أوميغا ٣ ، بينما تمت تغذية المجموعتين الثالثة والخامسة على النظام الغذائي عالي الفركتوز و زيوت أوميغا ٣ المختلطة مع جسيمات الذهب النانوية. و قد أظهرت النتائج أن هناك انخفاضًا كبيرًا في مستوى الجلوكوز والأنسولين في الدم وتقييم نموذج التوازن من مقاومه الانسولين (HOMA-IR) ومؤشر اكسدة الدهون في مجموعات جزيئات الزيوت النانوية مقارنة مع المجموعة الفرعية الأولى و زيادة ملحوظة في تقييم نموذج التوازن خلية البيتا (HOMA-β) والجلوتاثيون (GSH). و يمكن أن نخلص من ذلك أن زيوت أوميغا ٣ المختلطة مع جسيمات الذهب النانوية تتمتع بحماية مضادة للسكري والكبد وخصائص مضادة للأكسدة أكثر من زيوت أوميغا ٣ وحدها.