

Spirulina as Anti-Obesity and Hepato-Renal Protective Agent in MSG - Exposed Female Rats

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ABSTRACT

High intake of monosodium glutamate (MSG) – containing foods was reported as a major health problem in Egypt. Thus, dietary interventions aim to prevent the deleterious effects of this additive were badly needed. This study was carried out to investigate the effect of concurrent consumption of MSG and *Spirulina* (*S. platensis*) on body weight as well as hepato-renal functions and histopathology in female rats. Twenty adult female albino rats were divided into four equal groups, including the control group, while groups 2 to 4 were administered 6 mg MSG/kg body weight daily, and kept untreated (group 2), or concurrently fed pelleted balanced diet supplemented with 0.5 and 1 % of *Spirulina* powder for 6 weeks (groups 3 and 4, respectively). At the end, body weight gain, feed intake and feed efficiency ratio were calculated. Liver and kidney functions were determined in sera, while oxidative stress markers were determined in liver and kidney tissue homogenates. Moreover, specimens from liver and kidney of sacrificed rats were histopathologically examined. On the other hand, phenolic profile of *Spirulina* powder was identified and quantified by HPLC. Findings indicated that MSG consumption resulted in overweight, liver and kidney dysfunction along with oxidative stress, which was further confirmed by histological staining. Due to the high content of phenolic compounds rather than other antioxidant agents, *Spirulina* powder exerted anti-obesity properties and alleviated the toxic hepato-renal effects caused by MSG consumption. So, dietary supplementation of MSG –containing food products with *Spirulina* powder is recommended to prevent its accompanied health abnormalities.

Keywords: Body weight; hepato-renal insufficiency; monosodium glutamate; oxidative stress; *Spirulina platensis*.

INTRODUCTION

Monosodium glutamate (MSG) is a highly used flavor enhancer in commercial foods worldwide (Niaz et al., 2018). It is the sodium salt of the nonessential amino acid, L-glutamic acid, which is found naturally in many foods, including mushrooms, algae, soy, certain types of cheese such as Roquefort and Parmesan as well as some vegetables, the most important being tomatoes and broccoli. Packaged and processed foods (frozen foods, potato chips, salty snacks, sauces, sausages, candies and others) may contain MSG (Lölicher, 2000

and Yamaguchi and Ninomiya, 2000). It gives a special taste, known as umami, brothy or savory (Silva et al., 2017).

Although MSG was approved by Food and Drug Administration (FDA), pre-clinical studies revealed that repeated and over MSG ingestion was associated with many health problems, including cancer-induced obesity, diabetes, and asthma. Toxicities including hepatotoxicity, renal toxicity, reproductive toxicity and genotoxicity, as well as neurotoxic effects were indicated to accompany MSG intake (Bera et al., 2017 and Kazmi et al., 2017). MSG –induced oxidative stress is the main cause that lies behind its harmful effects.

Dietary interventions, particularly those that are plant -based, have been proposed as preventive agents against low-grade inflammation and oxidative stress (Arabzadegan et al., 2020). *Spirulina platensis* (*S. platensis*), known as blue-green algae, is a filamentous cyanobacterium often used as a single cell protein. Nutritionally, these microalgae were designated as a highly nutritious food by the World Health Organization (WHO). In food fortification and manufacturing, *S. platensis* can be considered one of the best alternative treatments, since it is a good source of essential amino acids, fatty acids, minerals and vitamins. It also provides antioxidant pigments, the most important being phycocyanin, chlorophyll, and beta-carotene. According to the National Institutes of Health (NIH), *S. platensis* can be used as a treatment for the nervous system abnormalities and metabolism disorders, including diabetes and dyslipidemia. It has also anti-viral, anti-bacterial, anti-oxidant, anti-inflammatory, anti-anemic, and anti-cancer properties. Therefore, *Spirulina* is called as a "superfood" and a "miracle from the sea" (Jung et al., 2019 and Bitam and Aissaoui, 2020).

High exposure to MSG represents a major health problem in Egypt, as it is found in a large number of food products; especially those directed to kids, within levels exceed the European limit of 10 g/kg (1%) of product. Even if found within the allowable limits in some food products, the over and repeated consumption of these products per day without paying attention to the total amount of MSG ingested exposes the person to critical health challenges in the next years (Abdel

DOI: 10.21608/asejaiqjsae.2021.181204

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Received May 15, 2021, Accepted, June 27, 2021.

Moneim et al., 2018). This study was carried out to investigate the weight control and hepato-renal protective effects of *Spirulina* in MSG-exposed female rats.

MATERIAL AND METHODS

Materials:

Plant material

Spirulina (S. platensis) powder was obtained from Free Trade Egypt Company, Behira Governorate, Egypt.

Animals

A total of 20 normal female albino rats (Sprague-Dawley strain) weighing 140 ± 5 g were obtained from the animal colony, Helwan Farm, Vaccine and Immunity Organization, Cairo Governorate, Egypt.

Diet, chemicals and kits

Pelleted balanced diet was purchased to feed rats from Agricultural Development Company, 6-October City, Giza Governorate, Egypt.

Pure MSG (white colored crystals) was purchased from Sigma supplier in Cairo City, Cairo Governorate, Egypt. All other required chemicals were obtained from El-Gomhoreya Company for trading drugs, chemicals and medical appliances, Tanta City, Al-Gharbia Governorate, Egypt.

Kits used for biochemical determinations were obtained from Gama Trade Company for chemicals, Cairo City, Cairo Governorate, Egypt.

Methods:

Animals & study design

Animals were housed in well-aerated cages under hygienic conditions in a room maintained at suitable humidity, 22 – 25 °C and a 12 h light-dark cycle, and fed pelleted balanced diet for one week for adaptation. The diet was already consisting of sunflower oil (15%), concentrate mixture 45% (10%), yellow corn (49%), soybean meal 44% (11%), wheat bran (10%), molasses (3%), common salt (0.5%), ground limestone (0.2%), dicalcium phosphate (0.1%), lysine (0.2%), dl-methionine (0.7%) and mineral-vitamin premix (0.3%).

By the end of adaptation period, rats were weighed and divided into four groups of 5 rats each. The first group was kept as a negative control group, while groups from 2 to 4 were administered 6 mg MSG/kg body weight daily by a stomach tube according to Ibrahim et al. (2011) to induce hepatotoxicity. The used dose also mediated the doses used by Okon et al. (2020) to induce nephrotoxicity in rats. At the same time, group 2 was kept untreated (positive control), while the third and the fourth groups were fed pelleted balanced diet supplemented with 0.5 and 1% of *Spirulina* powder

(SP), respectively. Rats were received MSG and *Spirulina* powder all over the experiment (protective study). The experiment lasted for 6 weeks. Meanwhile, feed and water were provided *ad-libitum* and body weight was recorded once a week. Generally, marked changes in body weight of MSG-administered groups versus the negative control one were noticed at the 4th week. By change of the body weight of each rat, its MSG dosage was also changed.

Blood and tissue sampling

By the end of the experiment, animals were weighed and fasted overnight before sacrificing. Blood samples were collected from the aorta of each rat into dry clean centrifuge tubes. Sera were carefully separated by centrifugation of blood samples at 3000 rpm (round per min) for 10 min at room temperature, then transferred into dry clean eppendorf tubes and kept frozen at -20 °C till analyzed. Moreover, livers and kidneys were removed by careful dissection, washed in ice-cold NaCl (0.9 g/100 mL) and dried using filter paper. After that, a specimen from each liver as well as the right kidney were stored at -80 °C until homogenate preparation, while other specimen from each liver as well as the left kidney were immersed in buffered neutral formalin solution (10 %) for latter histopathological examination.

Preparation of liver and kidney tissue homogenates

In order to prepare liver tissue homogenate, one gram of liver tissue was homogenized in ice-cold solution of potassium chloride (1.15 g/100 mL) in 50 mmol L⁻¹ potassium phosphate buffer solution (pH 7.4). For preparation of kidney tissue homogenate, 500 mg of each kidney tissue was homogenized in 5 mL phosphate buffer (0.1 M, pH 7.4). Homogenization was performed using Sonicator, 4710 Ultrasonics Homogenizer (Cole-Parmer Instrument Co., USA). The homogenates were centrifuged at 4000 rpm for 5 min at 4 °C. The supernatants were collected and stored at -80 °C for latter biochemical analysis.

Calculation of body weight gain, total feed intake and feed efficiency ratio

Body weight gain (BWG) was calculated by subtracting the initial weight of each rat from its final weight. Daily feed intake was calculated by subtracting the remainder food for each animal from that allocated to it every day. At the same time, the wasted food was weighed and subtracted. Total feed intake (TFI) was calculated through multiplying daily feed intake by 42. Feed efficiency ratio (FER) was then calculated through dividing BWG by TFI.

Assessment of antioxidant/oxidant biomarkers in liver and kidney tissue homogenates

In liver and kidney tissue homogenates, lipid peroxidation expressed as malondialdehyde (MDA) was determined following the method suggested by Ohkawa et al. (1979), while total antioxidant capacity (TAC) was determined according to Koracevic et al. (2001).

Determination of liver function –related markers in sera

In sera, the activities of liver enzymes including aminotransferases' (AST and ALT) and alkaline phosphatase (ALP) were determined according to Reitman and Frankel (1957) and Kind and King (1954), respectively. Moreover, total protein (T.P.) and albumin were determined according to the methods described by Gornall et al. (1949) and Doumas et al. (1971), respectively. In addition, the difference between serum total protein and albumin contents was calculated as serum globulin content. Albumin/globulin ratio was then calculated through dividing albumin value by that of globulin.

Determination of kidney function –related markers in sera

Urea and creatinine concentrations were determined in sera according to the methods described by Patton and Crouch (1977) and Houot (1985), respectively.

Histopathological examination

After sacrificing of rats, small pieces of each liver and left kidney were immediately fixed in neutral buffered formalin solution (10 %) for 24 h. The tissue samples were dehydrated in ascending grades of ethyl alcohol, cleared by xylene and embedded in paraffin. Sections of 5 μ thickness were mounted and stained with Haematoxylin and Eosin stain (Bancroft and Gamble, 2008). All specimens were microscopically examined for the histopathological assessment.

Phenolic compounds determination

Phenolic compounds found in *Spirulina* powder were identified and quantified by high-performance

liquid chromatography (HPLC) according to the method described by Goupy et al. (1999).

Statistical analysis

Statistical analysis was carried out using the program of Statistical Package for the Social Sciences (SPSS), PC statistical software (Version 20; Untitled – SPSS Data Editor). The results were expressed as mean \pm standard deviation (mean \pm SD). Data were analyzed using one-way classification, analysis of variance (ANOVA) test. The differences between means were tested for significance using Duncan test at $p<0.05$ (Sendcor and Cochran, 1979).

RESULTS

Body weight gain, total feed intake and feed efficiency ratio

At the beginning, there were no significant differences in the body weight of all experimental groups, while at the end, a significant difference between the negative control and untreated MSG – administered groups was noticed. Hence, body weight gain (BWG) of untreated MSG-administered group was found to be significantly higher than that of negative control group. Supplementation of diet offered to MSG –administered groups with *Spirulina* powder (0.5 and 1%) resulted in a significant decrease, however, neither the low concentration nor the high one could return BWG to its normal value (Table 1).

Total feed intake (TFI) was also increased significantly as a result of MSG administration. *Spirulina* –fed groups showed lower values of TFI than that of untreated MSG-administered group, however with no significance ($P\geq 0.05$). Like BWG, the mean value of feed efficiency ratio (FER) was increased significantly due to MSG administration, while the two concentrations of *Spirulina* powder decreased it significantly, with a significant rise compared to negative control group in the same time (Table 1).

Table 1. Effect of experimental diets supplemented with two concentrations of *Spirulina* powder on body weight gain, total feed intake and feed efficiency ratio in MSG -administered versus control rats

Groups \ Parameters	Negative control	MSG -administered	MSG -administered + 0.5% SP	MSG -administered + 1% SP
Initial weight (g)	138.35 \pm 17.62 ^a	139.12 \pm 17.72 ^a	144.20 \pm 19.12 ^a	142.00 \pm 18.56 ^a
Final weight (g)	173.60 \pm 22.30 ^a	227.48 \pm 29.76 ^b	208.90 \pm 26.33 ^{ab}	206.50 \pm 25.50 ^{ab}
BWG (g)	35.25 \pm 4.45 ^a	88.34 \pm 11.36 ^c	64.70 \pm 8.05 ^b	64.50 \pm 8.09 ^b
TFI (g)	655.10 \pm 83.37 ^a	846.85 \pm 106.92 ^b	740.72 \pm 93.13 ^{ab}	730.85 \pm 92.01 ^{ab}
FER	0.05 \pm 0.01 ^a	0.10 \pm 0.01 ^c	0.09 \pm 0.01 ^b	0.09 \pm 0.01 ^b

• Results are expressed as mean \pm SD.

• Values that have different letters in each row differ significantly ($P<0.05$), while the difference among those with similar letters completely or partially is not significant.

• MSG= Monosodium glutamate, SP= *Spirulina* powder, BWG= Body weight gain, TFI= Total feed intake, FER= Feed efficiency ratio.

Oxidative stress –related markers

As shown in table 2, lipid peroxidation marker values in liver and kidney tissue homogenates (L. MDA and K. MDA, respectively) of untreated MSG – administered group were significantly higher than their levels in negative control group. In contrast, total antioxidant capacity in the same tissues (L. TAC and K. TAC) were reduced significantly.

Consumption of diets supplemented with 0.5 and 1% of *Spirulina* powder resulted in significant increase in total antioxidant capacity in both liver and kidneys, which in turn resulted in a significant reduction in lipid peroxidation marker (MDA) in both tissues. The high concentration of *Spirulina* powder (1%) was better than the low one in enhancing antioxidant defense system in the studied tissues, and hence alleviated lipid peroxidation rate.

Liver and kidney function –related markers

Effect of experimental diets supplemented with two concentrations of *Spirulina* powder on liver and kidney function –related markers in sera of MSG-administered versus control rats was illustrated in table (3). It could be noticed that the activities of liver enzymes, including transaminases and alkaline phosphatase (AST, ALT and ALP, respectively), were significantly higher in sera of untreated MSG-administered group than their levels in sera of negative control group. *Spirulina* –fed groups recorded significant decrease in AST and ALP activities compared to untreated MSG-administered group, while ALT activity was reduced insignificantly ($P \geq 0.05$).

On the other hand, the mean values of total protein, albumin and globulin were decreased significantly in sera of untreated MSG-administered group compared to negative control group. Supplementation of diet introduced to MSG –administered groups with *Spirulina* powder (0.5 and 1%) resulted in a significant increase in

Table 2. Effect of experimental diets supplemented with two concentrations of *Spirulina* powder on oxidative stress –related markers in liver and kidney tissue homogenates of MSG-administered versus control rats

Groups \ Parameters	Negative control	MSG - administered	MSG - administered + 0.5% SP	MSG - administered + 1% SP
L. MDA (nmol/g)	3.57±0.45 ^a	13.37±1.73 ^d	9.40±1.20 ^c	5.87±0.80 ^b
L. TAC (mM/L)	0.69±0.09 ^d	0.05±0.01 ^a	0.19±0.02 ^b	0.47±0.06 ^c
K. MDA (nmol/g)	9.87±1.17 ^a	23.20±2.24 ^d	17.23±1.63 ^c	12.20±1.50 ^b
K. TAC (mM/L)	0.83±0.10 ^d	0.12±0.02 ^a	0.30±0.04 ^b	0.59±0.07 ^c

• Results are expressed as mean ± SD.

• Values that have different letters in each row differ significantly ($P < 0.05$), while the difference among those with similar letters completely or partially is not significant.

• MSG= Monosodium glutamate, SP= *Spirulina* powder, L. MDA= Liver malondialdehyde, L. TAC= Liver total antioxidant capacity, K. MDA= Kidney malondialdehyde, K. TAC= Kidney total antioxidant capacity.

Table 3. Effect of experimental diets supplemented with two concentrations of *Spirulina* powder on liver and kidney function –related markers in sera of MSG-administered versus control rats

Groups \ Parameters	Negative control	MSG - administered	MSG - administered + 0.5% SP	MSG - administered + 1% SP
AST (U/L)	73.25±8.99 ^a	189.45±23.67 ^c	147.82±18.99 ^b	142.63±15.63 ^b
ALT (U/L)	26.30±2.10 ^a	64.77±7.44 ^b	60.74±7.90 ^b	56.45±6.31 ^b
ALP (U/L)	55.36±6.38 ^a	207.62±25.35 ^c	134.16±16.31 ^b	112.97±14.36 ^b
T. P. (g/100mL)	6.65±0.82 ^c	3.76±0.46 ^a	5.30±0.59 ^b	4.99±0.63 ^b
Albumin(g/100mL)	4.26±0.52 ^c	1.85±0.20 ^a	2.36±0.32 ^b	2.50±0.31 ^b
Globulin(g/100mL)	2.39±0.34 ^b	1.91±0.29 ^a	2.94±0.35 ^c	2.49±0.34 ^b
Albumin/globulin ratio	1.79±0.25 ^b	0.97±0.12 ^a	0.80±0.11 ^a	1.00±0.13 ^a
Urea (mg/100mL)	36.79±4.43 ^a	62.13±7.27 ^c	43.71±5.07 ^{ab}	46.13±5.58 ^b
Creatinine(mg/100mL)	0.39±0.05 ^a	0.77±0.10 ^c	0.54±0.06 ^b	0.56±0.07 ^b

• Results are expressed as mean ± SD.

• Values that have different letters in each row differ significantly ($P < 0.05$), while the difference among those with similar letters completely or partially is not significant.

• MSG= Monosodium glutamate, SP= *Spirulina* powder, AST= Aspartate aminotransferase, ALT= Alanine aminotransferase, ALP= Alkaline phosphatase, T. P.= Total protein.

total protein and albumin, however, neither the low concentration nor the high one could normalize them. As for serum globulin, both concentrations of *Spirulina* powder could increase it significantly compared to untreated MSG-administered group. The high concentration of *Spirulina* powder (1%) was so efficient that could normalize serum globulin level, while the low concentration (0.5%) induced a significant increase compared to control group. Accordingly, albumin/globulin ratio in sera of untreated MSG-administered group was significantly lower than in negative control group, and accompanying feeding SP supplemented diets caused no significant changes.

MSG administration with no accompanying treatments induced renal dysfunction manifested through increasing the mean levels of urea and creatinine significantly. Both *Spirulina* -fed groups recorded significant decrease in urea and creatinine compared to untreated MSG-administered group with no significant differences between them. Moreover, the mean value of serum urea in SP (0.5%) -treated group was close to that recorded by negative control group.

Histopathological findings

Liver histopathology

Examination of haematoxylin and eosin stained hepatic sections of rats from negative control group showed hepatocytes normally arranged in radial plates around central veins (CV) with normal sinusoids (s) and portal areas (PA). Hepatic sections of rats from untreated MSG -administered group showed marked changes including micro-(green arrows) to macro (black arrows) -vesicular steatosis in hepatocytes with prominent mononuclear cells aggregation (yellow arrow). In hepatic sections of rats from 0.5% SP -fed group, dilated central veins (CV) and sinusoids (s) with some mononuclear cells around central veins (yellow arrow) were noticed. Examination of liver sections of rats from 1% SP -fed group showed noticeable improvement of liver architecture, as only mildly congested portal veins (red arrow) with very few mononuclear cells infiltrating the portal area (yellow arrow) were found (Figure 1).

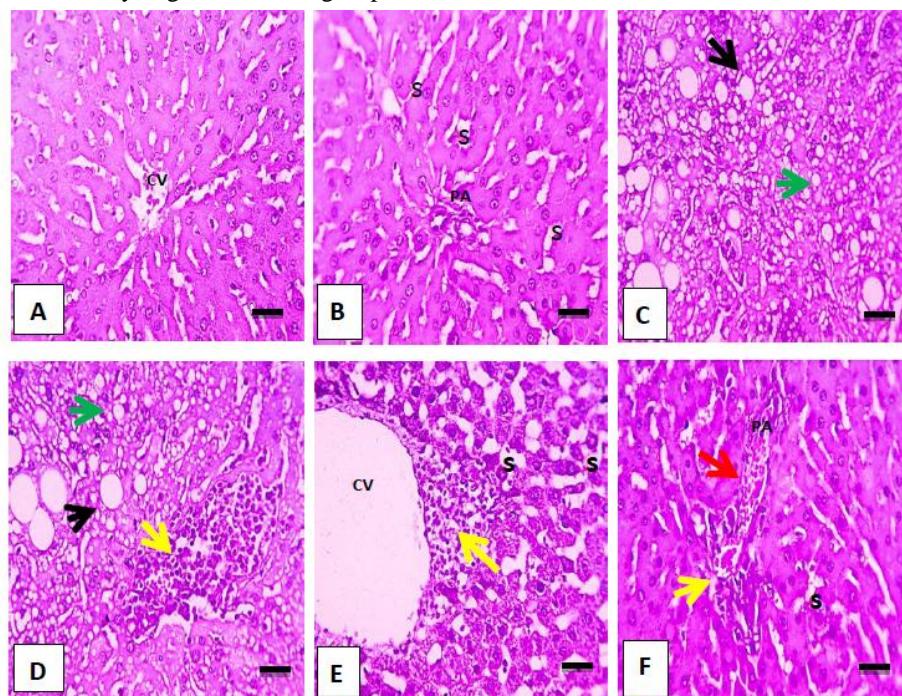


Figure 1 (A-F). Microscopic pictures of H&E stained hepatic sections showing hepatocytes normally arranged in radial plates around central veins (CV) (fig. 1A) with normal sinusoids (s) and portal areas (PA) (fig. 1B) in rats from negative control group. Hepatic sections of rats from untreated MSG -administered group showed marked changes including micro-(green arrows) to macro (black arrows) -vesicular steatosis in hepatocytes with prominent mononuclear cells aggregation (yellow arrow) (figs. 1C and 1D). Hepatic sections of rats from 0.5% SP -fed group showed dilated central veins (CV) and sinusoids (s) with some mononuclear cells around central veins (yellow arrow) (fig. 1E). Hepatic sections of rats from 1% SP -fed group showed mildly congested portal veins (red arrow) with very few mononuclear cells infiltrating the portal area (yellow arrow) (fig. 1F) (Low magnification X: 100 bar 100, high magnification X: 400 bar 50).

Kidney histopathology

Microscopic examination of haematoxylin and eosin stained renal sections of rats from negative control group showed normal glomeruli and tubules with minimal interstitial tissue. In contrast, marked abnormal changes were noticed in renal sections of rats from untreated MSG –administered group. They include tubular dilation (black arrow), vacuolar degeneration in renal epithelium (green arrow), congested blood vessels veins (red arrows) with prominent interstitial (dashed yellow arrows) and perivascular mononuclear cells

infiltration (yellow arrow). Renal sections of rats from 0.5% SP –fed group showed tubular dilation (black arrow) with few perivascular mononuclear cells infiltration (yellow arrow) and very mild interstitial collagen deposition (blue arrow). In renal sections of rats from 1% SP –fed group, however, tubular dilation (black arrow) with cast formation (arrowhead), congested blood vessels (red arrow) with few perivascular mononuclear cells infiltration (yellow arrow) and mild interstitial collagen deposition (blue arrow) were found (Figure 2).

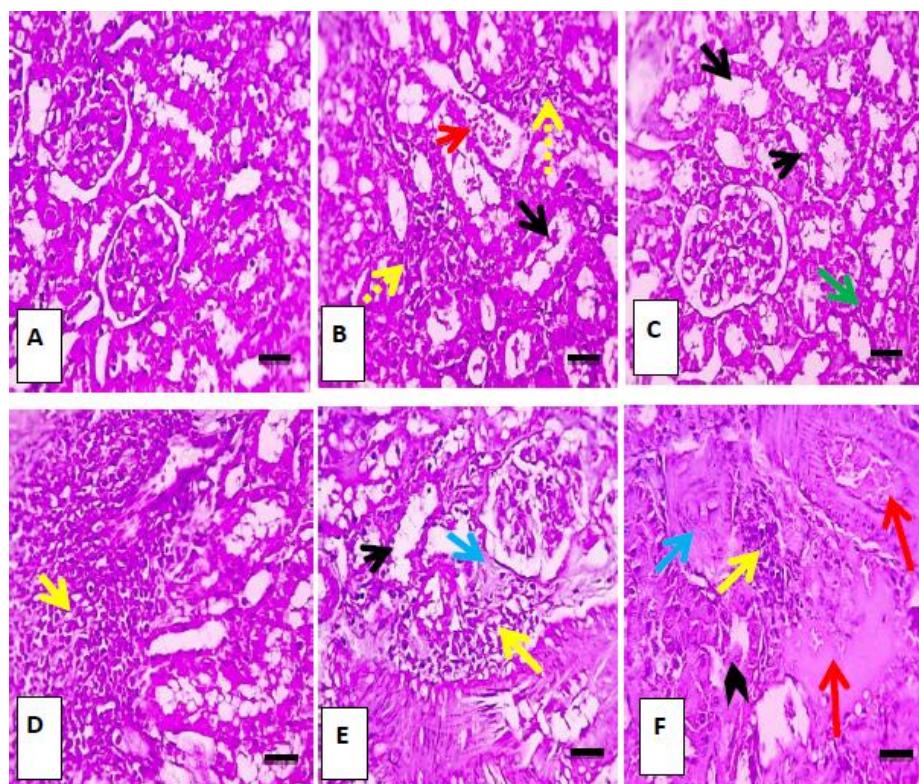


Figure 2 (A-F). Microscopic pictures of H&E-stained renal sections showing normal glomeruli and tubules with minimal interstitial tissue in rats from negative control group (fig. 2A). Renal sections of rats from untreated MSG –administered group showed marked changes including: tubular dilation (black arrow), vacuolar degeneration in renal epithelium (green arrow), congested blood vessels veins (red arrows) with prominent interstitial (dashed yellow arrows) and perivascular mononuclear cells infiltration (yellow arrow) (figs. 2B, 2C and 2D). Renal sections of rats from 0.5% SP –fed group showed tubular dilation (black arrow) with few perivascular mononuclear cells infiltration (yellow arrow) and very mild interstitial collagen deposition (blue arrow) (fig. 2E). Renal sections of rats from 1% SP –fed group showed tubular dilation (black arrow) with cast formation (arrowhead), congested blood vessels (red arrow) with few perivascular mononuclear cells infiltration (yellow arrow) and mild interstitial collagen deposition (blue arrow) (fig. 2F) (Low magnification X: 100 bar 100, high magnification X: 400 bar 50).

Phenolic profile of *Spirulina* powder

HPLC analysis indicated the presence of 14 phenolic compounds in *Spirulina* powder, being the major is pyrogallol (58693×10^{-6} %). Chlorogenic acid was the second compound found abundantly (8.733×10^{-3} %) in SP, followed by catechin (3.616×10^{-3} %) and ellagic acid (3.294×10^{-3} %). The other 10 compounds were found in small concentrations ranged between 7.93×10^{-4} and 2.49×10^{-4} %. In descending order, they included salicylic acid, P-OH-benzoic acid, caffeine, gallic acid, catechol, ferulic acid, 4-aminobenzoic acid, vanillic acid, caffec and coumarin (Table 4).

Table 4. HPLC analysis of phenolic profile in *Spirulina* powder

Items	Phenolic compounds in SP (%)
Pyrogallol	58693×10^{-6}
Gallic acid	4.92×10^{-4}
Catechol	4.50×10^{-4}
4-Aminobenzoic acid	3.74×10^{-4}
Catechin	3.616×10^{-3}
Chlorogenic acid	8.733×10^{-3}
P-OH-benzoic acid	6.98×10^{-4}
Caffeic acid	2.53×10^{-4}
Vanillic acid	3.12×10^{-4}
Caffeine	6.26×10^{-4}
Ferulic acid	4.09×10^{-4}
Ellagic acid	3.294×10^{-3}
Salicylic acid	7.93×10^{-4}
Coumarin	2.49×10^{-4}

DISCUSSION

The present results revealed that MSG administration induced significant increase in body weight gain, total feed intake, and hence feed efficiency ratio. These results were in agreement with several previous studies (Gomathi et al., 2008; Akataobi, 2020 and Hossain et al., 2020). Studies have shown that MSG administration can induce over weight through various mechanisms. One of them, is that it can induce hyperphagia and elevate the energy intake (Bergen et al., 1998). Hyperphagia itself was found to be associated with MSG exposure due to improving palatability of food as it is a taste enhancer, reduction of brain and plasma serotonin (Gomathi et al., 2008), and interruption in the hypothalamic signaling process of leptin which causes the exposed animal to eat more food while being hypoactive (He et al., 2011 and Roman-Ramos et al., 2011).

Recently, Moradi et al. (2019) revealed that *Spirulina* supplementation significantly induces weight loss, especially in obese individuals. This study supported the anti-obesity effect of *Spirulina* noticed in the present study. The proposed anti-obesity mechanisms of action of *Spirulina* are macrophage infiltration reduction into visceral fat, combating hepatic fat accumulation, reduction in oxidative stress, improvement in insulin sensitivity and satiety (DiNicolantonio et al., 2020). *Spirulina* was also found to render fat digestion and absorption in the small intestine (Han et al., 2006).

As revealed by the present findings, MSG enhanced oxidative stress in liver and kidney tissue homogenates, evidenced by increased level of malondialdehyde, the lipid peroxidation marker, as well as decreased level of total antioxidant capacity. These results were in agreement with many previous studies (Yaqub et al., 2008 and Ibrahim et al., 2011). El Agouza et al. (2010) reported that administration of MSG induced oxidative stress leading to an increase in the intracellular concentration of Ca^{2+} ; the increased Ca^{2+} levels could theoretically act either to enhance lipid peroxidation or to stimulate degeneration of phospholipids. Yaqub et al. (2008) concluded that MSG exposure increases the production of free radicals, which in turn react with polyunsaturated fatty acids in cell membranes leading to lipid peroxides production and impairment of mitochondrial and plasma membranes.

According to these toxic effects of MSG on liver and kidney tissues in the current study, their morphological structure and functions were found to be impaired, which can be noticed through: 1) marked changes in hepatocytes including micro- to macrovesicular steatosis with prominent mononuclear cells aggregation, associated with marked changes in kidney tissues including tubular dilation, vacuolar degeneration in renal epithelium, congested blood vessels veins with prominent interstitial and perivascular mononuclear cells infiltration, 2) increased activities of liver enzymes, including AST, ALT and ALP, in serum, 3) lowered concentrations of serum proteins, and 4) increased concentrations of protein metabolites, including urea and creatinine, in serum. These results were in line with many previous studies (Yaqub et al., 2008; Ibrahim et al., 2011 and Hossain et al., 2020). The elevated activities of transaminases in serum could be due to increased free radical production caused by MSG which reacts with the liver cell membrane and damage the cellular structure, resulting in enzyme leakage (Tawfik and Badr, 2012). As for ALP, it is present in the intra and extra – biliary duct walls, thus its elevation may express a damage of the biliary cells (Suzuki et al.,

2006), and is often used, generally, as an indicator of liver adaptation to toxic agents. Regarding the proteins, their lowered levels in serum of untreated MSG – administered group confirmed the marked liver dysfunction induced by MSG, as liver is the organ responsible for the synthesis and transport of proteins to the bloodstream (Naganna, 1989). On the other hand, the significant increase in creatinine and urea contents of the serum, following the administration of MSG, is usually a result of kidney dysfunction, which in turn can be attributed to the oxidative stress on the renal tissue, resulting in impairment of kidneys ability to excrete waste materials from the body (Vinodini et al., 2010).

As noticed in the present study, dietary supplementation with *Spirulina* induced hepato-renal preventive effects, as it strengthened antioxidant defense system, decreased lipid peroxidation, and alleviated the histopathological and biochemical abnormalities associated with liver and kidney functions in MSG –administered rats. Similar results were reported in deltamethrin-intoxicated rats (Abdel-Daim et al., 2013). In general, most of *Spirulina*'s health benefits are associated with its antioxidant pigments including carotenoids (especially β -carotene and zeaxanthin), chlorophyll and the unique blue pigment phycocyanin (Asghari et al., 2016).

According to Subhashini et al. (2004), phycocyanin is not only the predominant compound in the antioxidant capacity of the *Spirulina*, but also is a vital difference between it and other green foods like chlorella, wheat grass and barley. It was reported to be able to scavenge free radicals, including alkoxyl, peroxy and hydroxyl radicals. It also reduced nitrite production, lowered inducible nitric oxide synthase (iNOS) expression, and inhibited liver microsomal lipid peroxidation.

Spirulina powder also was found to be very rich (1080 units/ 1 g) in the superoxide dismutase, an important free radical scavenging enzyme, which was recommended to be used as a treatment of various diseases related to oxidative stress (Asghari et al., 2016). Moreover, dried *Spirulina* contains nonenzymatic antioxidants such as vitamins E and C (about 5 and 10.1 mg/ 100 g, respectively) according to The United States Department of Agriculture (USDA) Food Composition Databases.

Regarding to the phenol profile of *Spirulina* powder, four compounds were found in high concentrations, the major being pyrogallol, followed by chlorogenic acid, catechin and ellagic acid. Pyrogallol-type phenolic compounds have generally been shown to possess markedly high activity to scavenge free radicals (Biskup et al., 2013). Chlorogenic acid, as a strong antioxidant, was also

reported to overcome oxidative stress-induced hepatorenal toxicity in a number of experimental models as a pure compound (Ding et al., 2021) or a phytochemical constituent of some fruits (El-hawary et al., 2019). It was also reported to have anti-obesity effects (He et al., 2021). Catechin and ellagic acid also induced anti-obesity and antioxidant effects in many previous studies. For example, findings of Yan et al. (2013) in obese rats suggested that green tea catechins exert their anti-obesity mechanism in part by modulating peroxisome proliferator activated-receptor signaling pathways. Catechin pre-treatment in mice abrogated tamoxifen-induced hepato-renal toxicity, as it decreased lipid peroxidation levels, H₂O₂ generation and protein carbonyl contents, while normalized non-enzymatic antioxidants, and restored the activities of antioxidant enzymes when compared with tamoxifen-treated group (Parvez et al., 2006). On the other hand, ellagic acid was proved to have a significant anti-obesity effect (Wang et al., 2019), as well as a protective effect against lead – induced hepatotoxicity in a dose dependent manner in rats (Ananya et al., 2020).

Conclusion

Promising effects of *Spirulina* powder in the prevention of obesity and hepato-renal insufficiency are recorded in MSG –exposed female rats. Isolation and re-evaluation of the active principles of *Spirulina* are required.

Acknowledgements

The author thanks Nutrition and Food Science Dept., Faculty of Home Economics, Al-Azhar University for supporting this research.

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الملخص العربي

الاسبيرولينا كعامل مضاد للسمنة وواقي للكبد والكلى فى إناث الجرذان المعرضة لأحادي جلوتامات الصوديوم

سامح أحمد الحشاش

دلائل الأكسدة في أنسجة الكبد والكلى. علاوة على ذلك تم فحص أنسجة الكبد والكلى هستوباثولوجيا وتحديد وتقدير التركيب الفينولي لمسحوق الاسبيرولينا بواسطة جهاز الكروماتوجراف السائل فائق الأداء. أسفرت النتائج عن اقتران استهلاك مادة أحادي جلوتامات الصوديوم بحدوث زيادة في الوزن واختلال وظائف الكبد والكلى وتلف تأكسدي بهما وهو ما أكدته الفحص الهستوباثولوجي. أما مسحوق الاسبيرولينا فأظهر خصائص مضادة للسمنة وقلل من التأثيرات السامة التي أحدثتها مادة أحادي جلوتامات الصوديوم في الكبد والكلى وهذا يرجع إلى محتواه العالي من المركبات الفينولية، فضلاً عن عوامل أخرى عديدة مضادة للأكسدة. لأجل ذلك، يوصى بتدعيم المنتجات الغذائية المحتوية على مادة MSG بمسحوق الاسبيرولينا للوقاية من التحديات الصحية المصاحبة لها.

الكلمات المفتاحية: وزن الجسم، عدم كفاءة الكبد والكلى، أحادي جلوتامات الصوديوم، التلف التأكسدي، طحلب الاسبيرولينا.

يعد المأكولات العالية من الأطعمة المحتوية على أحادي جلوتامات الصوديوم مشكلة صحية كبيرة في مصر؛ وهكذا كانت الحاجة ماسة إلى التدخلات الغذائية التي تستهدف الوقاية من التأثيرات الضارة لهذه المادة المضافة. أجريت هذه الدراسة لبحث تأثير استهلاك أحادي جلوتامات الصوديوم متزامناً مع طحلب الاسبيرولينا على وزن الجسم بالإضافة إلى وظائف وتركيب أنسجة كل من الكبد والكلى في إناث الجرذان، حيث تم تقسيمهن (إجمالي ٢٠ جرذ بالغ) إلى أربع مجموعات متساوية العدد تشمل المجموعة الضابطة، بينما تم تجريب المجموعات من الثانية إلى الرابعة بمادة أحادي جلوتامات الصوديوم (٦ ملجم/ كجم من وزن الجسم/ اليوم) وترك المجموعة الثانية دون معالجة مصاحبة، في حين غذيت المجموعات الثالثة والرابعة على علبة قياسية مدعمة بمسحوق طحلب الاسبيرولينا بجرعاتي ١٪ ، ٥٪ على التوالي، وذلك لمدة ستة أسابيع. وفي النهاية، تم حساب الزيادة المكتسبة في الوزن والمأكولات الغذائية ومعدل كفاءة الغذاء، وتقدير وظائف الكبد والكلى في السيرم، بينما تم تقدير