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Amelioration effects of Saffron on body weight and oxidative stress in male rats with induced Diabetes Mellitus

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Abstract-**The study aim to investigate the mitigating effect of Saffron on body weight and oxidative stress in male rats exposed to streptozotocine to induced Diabetes Mellitus. 40 male rats aged 8-10 weeks included in this study, it divided to 4 group.10 male rats don't receive any type of treatment and only injected an equal volume of vehicle. Saffron group: 10 male rats receive daily orally dose of Saffron extract 500mg/kg/bw daily for 28 days. Streptozotocin group :10 male rats received only one single I/P injection of streptozotocin 65mg/kg /bw. Streptozotocin + Saffron group: 10 male rats receive one single I/P injection of streptozotocin 65mg/kg/ BW, followed by daily orally dose of Saffron extract 500mg/kg /bw for 28 days. The results showed a significant improvement ($p \leq 0.05$) in body weight and oxidative stress values in the saffron + streptozotocine group compare with streptozotocine alone group. In conclusion, Saffron has an enhanced effect on the body weight and oxidative stress in diabetes mellitus induced rats.**
KEYWORDS:Saffron (Crocus sativus), streptozotocine, diabetes mellitus

INTRODUCTION

Millions of individuals worldwide suffer from diabetes mellitus(DM), a chronic metabolic condition caused by either insulin resistance or insufficiency. It is therefore divided into Types 1 and 2, particular forms of diabetes brought on by different factors, and Gestational Diabetes (diabetes related to pregnancy). Insulin injections are used to treat type 1 diabetes, which is caused by an extreme insulin deficit as a result of the autoimmune destruction of β-cells. Insulin resistance (lack of insulin function) and impaired insulin secretion combine to cause type 2 diabetes, the most prevalent kind (1).

Numerous people worldwide are living with undiagnosed diabetes and prediabetes, according to other studies (2). Diabetes causes several health complications include cardiovascular diseases (3) pancreatic, liver and a variety of cancers (4,5). Furthermore, it is also known to cause organ damage following microvasculature obstruction and this includes diabetic nephropathy, retinopathy, peripheral neuropathy and peripheral artery disease (6,7).

Patients with DM are treated with a variety of medications to reduce their plasma glucose levels. In many parts of the world, the use of herbal medicine to treat a variety of illnesses has a long history (8). Other studies have showed the many benefits of these medicinal plants and the substances they extract. There is proof that type 2 diabetes mellitus was considered a serious "Xiao-Ke disease" in ancient Chinese texts, and Chinese people have been using herbal remedies to cure illnesses for about 1500 years (9) Many studies have been conducted recently in an effort to determine the potential benefits of herbal medicine in managing diabetic complications and enhancing patient outcomes (9, 10). Saffron (Crocus sativus) is one of the spices whose anti-diabetic properties have been assessed in a number of research. (11).

One of the priciest spices, saffron is often farmed as a perennial crop. Saffron is used as a spice because of its scarlet stigmas. Saffron has antidepressant, anti-inflammatory, and anticancer effects. Additionally, it is used to treat a number of illnesses, including diabetes, Alzheimer's disease, and certain types of cancer (12). It is said that saffron is an old spice. In addition to its therapeutic properties, saffron is used as a food spice and for coloring. Many nations, including Iran, Greece , India, Afghanistan, Italy , and Spain, grow saffron. 90% of the world's saffron is produced in Iran. (13).

Numerous substances, such as vitamins, minerals, protein and carbs, flavonoids, and anthocyanins, are found in saffron. Safranal, crocin , crocetin and picrocrocin are the main saffron constituents. Saffron contains two carotenoid compounds: crocetin and crocin . Crocin, which gives saffron its red hue, makes up between 6% and 16% of its total dry matter, depending on the production technique (14). In vivo, the antihypertensive effects of crocin and safranal have been demonstrated. Additionally, crocin has demonstrated lipid-lowering and anti-diabetic properties (15). Also, safranal and crocin have demonstrated strong antioxidant effects (16). It is well recognized that oxidative stress (OS) in uncontrolled hyperglycemia plays a key part in liver damage and pancreatic dysfunction. Superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase are the main constituents of the tissues' intrinsic anti-oxidation system (17). In this context, Yaribeygi et al. investigated how Crocin, a pharmacologically active part of Crocus sativus L., might strengthen the hepatic and pancreatic cells' antioxidant defense mechanisms. The findings demonstrated that Crocin increased the antioxidant defense system by increasing the enzymatic activities of SOD and catalase. It can also reduce oxidative stress by lowering the production of malondialdehyde (MDA), which is the primary indicator of OS, in the liver and pancreatic tissues of rats treated with diabetes (18,19).

Reducing lipid peroxidation is another way that saffron compounds enhance antioxidant status (20). It has been reported that saffron restores SOD activity and inhibits lipid peroxidation (21,22).

Materials And Methods

The study is lasted for 1 month and performed during the period from 1st of January 2025 to 1st of February 2025. Forty male Wister rats aged 8-10 weeks will included in this experiment (weighting approximately250-300 grams) and keep in the animal house at the Veterinary Medicine Faculty, Kerbala University, at 22to 24°C in a light /dark cycle of 12 on12 hours. The given food was rat chow and water which given freely. Rats stayed in the animal house for two weeks before beginning the experiments. The experimental Design includes 40 male Wister rats aged 8-10 weeks, and the groups will divided into following:

Control group : 10 male rats don’t receive any type of treatment and only injected an equal volume of vehicle. Saffron group: 10 male rats receive daily orally dose of Saffron extract 500mg/kg/bw daily for 28 days (23). Streptozotocin(STZ) group :10 male rats received only one single I/P injection of streptozotocin 65mg/kg /bw (24). Streptozotocin + Saffron group: 10 male rats receive one single I/P injection of streptozotocin 65mg/kg/ BW, followed by daily orally dose of Saffron extract 500mg/kg /bw for 28 days.

Statistical analysis

The findings were analyzed statistically using SPSS version 22.00, with one-way (ANOVA) tests employed to determine the significance of differences between the groups' results. The data was reported as Mean ± Standard Deviation (SD), and a P-value ≤ 0.05 was considered statistically significant. The LSD test was carried out to test the significant levels between means of treatments (25).

Result & discussion

The Effects of Streptozotocin and Saffron on the Body weight after 2 days of treatment show no significant differences between the Streptozotocin, Streptozotocin +Saffron, Saffron alone and control group (p≥0.05) , while after 28 days of treatment the results showed a significant decrease (p ≤ 0.05) in the body weight in Streptozotocin group when compare with control and Saffron group, a significant decrease in the body weight in streptozotocin+Saffron group when compare with control and Saffron group, and a significant increase (p≤0.05) in body weight in streptozotocin+Saffron group when compare with Streptozotocin group (table 1).

Table 1. Effects of Streptozotocin and Saffron on the Body weight after 2 days and 28 days of treatment

Periods / Groups	Weight (gm) after 2 days of treatment Mean ±SD	Weight (gm) after 28 days of treatment Mean ±SD
Control	255.4 ± 16.30337A	271.0±13.80217 A
Saffron	271.8 ± 19.48589A	258.4±20.41568 A
Streptozotocin	261.0 ± 17.20465A	205.2±8.64292 C
Streptozotocin +Saffron	271.0 ± 18.49324A	225.6±5.77062 B

The significant change between groups demonstrate by different letters (p ≤ 0.05)

In our investigation, administering STZ affected the experimental rats' glycemic control and body weight. Oral use of Saffron leaf extract reversed the glycemic indicators. The rats gained a significant weight after the therapy with Saffron leaf extract at 500 mg/kg when compare between Streptozotocin group and Streptozotocin +Saffron group (table1).

The demand for herbal remedies to reduce blood sugar levels has been a major topic of interest among patients with diabetes and researchers. Type 1 diabetes and its devastating complications impact patients' ability to live a normal life. The body weight of rats , after 2 days of treatment, show no significant differences (p ≥ 0.05) between the all four groups of experiment, while after 28 days, as anticipated, the treated streptozotocine + Saffron group's body weights were noticeably show significant increase (p ≤ 0.05) than streptozotocine group and significant decrease (p ≤ 0.05) than control and Saffron group, this is because the streptozotocine rats' poorly managed glycemic state because of Type 1 diabetes incidence which tends to affect metabolic processes. Also, treatment with saffron in group streptozotocine+ Saffron group help in regain body weight lost during the four weeks baseline period, Better glycemic control in the treated animals was likely linked to the increases in body weight, which in turn caused the streptozotocine + saffron group's rate of catabolism to decline.

Reactive Oxygen species measured in this research include (GSH,SOD and MDA). The Effects of Streptozotocin and Saffron on GSH and SOD levels show a significant decrease (p ≤ 0.05) of GSH and SOD levels in Streptozotocin group when compare with all other group, and the highest significant increase (p ≤ 0.05) of GSH and SOD levels show in Saffron group when compare with all other group, also there is a significant increase (p ≤ 0.05) in Streptozotocin +Saffron group when compare with Streptozotocin group.

The results of MDA show a significant increase (p ≤ 0.05) in Streptozotocin group when compare with all other group and the lowest significant decrease (p ≤ 0.05) of MDA levels show in Saffron group when compare with all other group, also there is a significant decrease (p ≤ 0.05) in Streptozotocin +Saffron group when compare with Streptozotocin group(table 2).

Table (2) Effects of Streptozotocin and Saffron on Reactive Oxygen Species levels

Groups	GSH (ng/L) Mean ±SD	SOD(U/ml) Mean ±SD	MDA(nmol/ml) Mean ±SD
Control	166.3±7.7802895 B	326.1±17.0555454B	13.04±1.2900304 C
Saffron	183.1±14.7363257 A	353.3±19.8737381A	10.24±1.2871151 D
Streptozotocin	97.55±11.4023816D	196.1±14.5582998D	22.90±2.3361584 A
Streptozotocin +Saffron	133.2±8.7630757C	294.2±15.6420082C	15.74±2.0864751 B

The significant change between groups demonstrate by different letters ($p \leq 0.05$)

Actually, the antioxidant properties of phenolic compounds, which are widely found in plant byproducts, are of great interest. They primarily serve as metal chelators, hydrogen donors, reducing agents, and singlet oxygen quenchers (26).

As a coloring and flavoring component, saffron (Crocus sativus), the most important medicinal food product, is a member of the Iridaceae family. These characteristics mostly relate to its contents of crocins, picrocrocins, and safranal, all of which have been shown to have health-promoting qualities (27)

Similar to some other medicinal plants, saffron is known for its natural potent antioxidant (16 , 28,) and anti- inflammatory (29, 30) properties. Based on initial phytochemical studies, it may be suggested that the antioxidant and anti-inflammatory properties of saffron extract are mostly attributed to its carotenoids, flavonoids, and anthocyanins (29).

According to several research, crocin and crocetin, two carotenoids that make up saffron, had the highest levels of radical scavenging activity, followed by safranal. (16). These components' high radical scavenging activity may be caused by their capacity to provide free radicals a single hydrogen atom. Studies on animals and in vitro have demonstrated that saffron extracts, particularly crocetin and crocin, lower plasma levels of malondialdehyde (MDA), a measure of lipid peroxidation brought on by ROS. Additionally, saffron extracts greatly boost antioxidant ability (|31,32). Additionally, other in vivo and human investigations have demonstrated the antioxidant effect of chemicals called carotenoids that were isolated from saffron. (33,34). Generally speaking, the saffron extract's exceptional antioxidant capacity might be linked to the complementary actions of its constituent compounds.

. Conclusion

This study's results show that streptozotocine caused diabetes mellitus, which is mediated by oxidative stress and disturbances in body weight. Its anti-inflammatory and antioxidant qualities, which restore body weight and lessen oxidative damage, are responsible for saffron's positive effects. These findings are consistent with earlier research and offer a physiological justification for the use of saffron as an adjuvant therapy to lessen the body's reaction to diabetes mellitus

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