Original Article

Antidiabetic effect of hydroethanolic extract of *Crocus sativus* stigmas, tepals and leaves in streptozotocin-induced diabetic rats

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Abstract

Introduction: The present study investigated for the first time, the antihyperglycemic effect of tepals and leaves of *Crocus sativus* in streptozotocin-induced diabetic rats. The effect of these by-products were compared with that of saffron stigma and glibenclamide.

Methods: Hydroethanolic extracts (stigmas, tepals and leaves) and glibenclamide were administered orally in aqueous solution daily for 21 days. In the present study, 36 male rats were used. Six rats were used in each group. Group 1: normal control rats (N), received distilled water; group 2: diabetic control rats (D), received distilled water; group 3: treated diabetic rats (TPL), received tepal (TPL) extract; group 4: treated diabetic rats (STG), received stigma (STG) extract; group 5: treated diabetic rats (LF), received leaf (LF) extract and group 6: treated diabetic rats (GLB), received glibenclamide (GLB). Blood glucose, body weight, water intake, urine elimination, plasma triglycerides, cholesterol, urea, creatinine, aspartate amino transferase (AST) and alanine amino transferase (ALT) levels were evaluated.

Results: The present data indicated that the tepals and stigmas extracts significantly prevented decreasing body weight and protected against elevation of water intake, urine elimination, plasma triglycerides, cholesterol, urea, creatinine, AST and ALT levels in treated diabetic rats as compared to untreated diabetic rats. Leaves extract significantly prevented decreasing body weight. It decreased water intake, blood glucose, plasma triglycerides, cholesterol, creatinine, AST and ALT.

Conclusion: Based on our data, the oral administration of tepals, stigmas and leaves extract of *Crocus sativus* reduces blood glucose levels and improves control of diabetes complications.

Keywords: *Crocus sativus*; Antihyperglycemic effect; Streptozotocine; Saffron; By-products

Introduction

Diabetes mellitus is one of the principal causes of mortality and morbidity in the world (Zimmet et al., 2016). It’s a major chronic disease caused when the pancreases doesn’t secrete sufficing insulin or when the body can’t efficaciously use this insulin (Tousoulis
et al., 2012). The increase of oxidative stress and the alteration of the antioxidant defense system, are the main disorders associated with diabetes mellitus, which contributes to the initiation and progression of diabetes complications (Maritim et al., 2003). Medications used to treat diabetes are expensive and associated with undesirable side effects. For example, glibenclamide tended to increase plasma insulin and risk of hypoglycemia (Segal et al., 1997). Recently, research has focused on natural agents, such as medicinal plants, that provide effective, reliable and inexpensive treatment (Samarghandian et al., 2017). Morocco is the only country in Africa and the MENA region to produce the stigma of saffron. Moroccan surfaces grown from saffron are about 1000 ha, mainly in the mountains of the Anti-Atlas. This production area take more and more importance in other regions (Mounira et al., 2015).

*Crocus sativus* L. (saffron) belongs to the Iridaceae family. Saffron stigmas have long been used in traditional medicine, cosmetics and as a food additive for coloring and flavoring (Caballero-Ortega et al., 2007). Further, the stigma of *Crocus sativus* is traditionally used as stimulant, aphrodisiac, antidepressant, antispasmodic and eupetic to treat menstruation disorders, liver disease, physical suffering, cough, bronchospasm, asthma, heart disease, smallpox, fever and colds (Schmidt et al., 2007).

Numerous scientific studies have shown that saffron stigma has antioxidant properties (Farahmand et al., 2013), antitumor (Samarghandian and Borji, 2014), anti-inflammatory, antinociceptive (Hosseinzadeh and Younesi, 2002), antidepressant (Lopresti and Drummond, 2014), antilussive (Hosseinzadeh and Ghenaati, 2006), hypolipidemic (Sheng et al., 2006) and could improve memory and learning abilities in rats (Ghadami and Pourmotabbed, 2009; Papandreou et al., 2011). However, large amounts of saffron by-products are generated during processing of the stigmas. About 350kg of tepals, 1500kg of leaves needed to obtain only 1kg of dry stigmas (Smolskaite et al., 2011).

Tepals extracts have free radical scavenging (Hossein Goli et al., 2012; Zeka et al., 2015; Tuberoso et al., 2016), antidepressant (Moshir et al., 2006), antinociceptive, anti-inflammatory (Hosseinzadeh and Younesi, 2002) and antityrosinase activities (Kubo and Kinst-Hori, 1999; Li et al., 2004). These extracts can lower blood pressure and contractile response (Fatehi et al., 2003). They also have metal chelating properties (Sánchez-Vioque et al., 2012) cytotoxic and antifungal activity (Zheng et al., 2011; Serrano-Díaz et al., 2013). Saffron leaves are considered as source of bioactive components (Smolskaite et al., 2011). Saffron leaves extract have antioxidant and metal chelating properties (Sánchez-Vioque et al., 2012; Lahmass et al., 2018), activate proteasome in human dermal fibroblast cells from aged doners and inhibit the growth and proteasome activity of cancer cells (Sánchez-Vioque et al., 2016; Lahmass et al., 2017a).

Diabetes complications are probably linked to oxidative stress which is the result of increased blood glucose levels. Equally, the presence of bioactive molecules with a strong antioxidant properties on tepals and leaves of *Crocus sativus* thus prompted us to design the present study. For the first time, we investigated the antihyperglycemic effect of these by-products. We also examined the effect on diabetes complications. We measured plasma urea and creatinine levels to evaluate renal dysfunction. Similarly, we measured plasma alanine amino transferase (ALT) and aspartate amino transferase (AST) to study liver damage. In addition, it was reported in our laboratory and other studies that *Crocus sativus* stigma have an antihyperglycemic effect (Lahmass et al., 2017b; Samarghandian et al., 2017). For this reason we compared these by-products with *Crocus sativus* stigma and glibenclamide.

**Materials and methods**

**Plant materials**

The stigmas and tepals were harvested from a farm in Taliouine (30° 31’ 54° nord, 7° 55’ 25° 0’ oust, South of Morocco) and the leaves were collected from a farm in the region of Oujda (34° 41’ 12° nord, 1° 54’ 41° oust, East of Morocco) which the original corms were obtained from Taliouine. Saffron of both farms is cultivated without any chemical treatments. The different parts of the plant were collected manually between October and November (2016).

The identification of the plant has been done and confirmed by a professional botanist, Professor Fennane Mohammed from Scientific Institute in
Rabat, Morocco. Three specimens of the plant have been deposited at the plant section of Herbarium University Mohammed Premier, Oujda, Morocco (HUMPOM), under the voucher number (HUMPOM210).

**Chemicals**

Streptozotocin (STZ) was purchased from Sigma-Aldrich (Hambourg, Allemande), glibenclamide were obtained from Sigma chemicals (USA). All other chemicals and reagents used were of analytical grade.

**Animals**

Adult male Wistar rats (weight 150-200g) were maintained in their own cages (In the animal house of the Department of Biology, Faculty of Sciences, Oujda, Morocco) at constant temperature (21±2°C) and in a 12h light/dark cycle with free access to food (Dry rat pellets feeds provided by SONABETAIL Society, Oujda, Morocco) and water throughout the experiments. Maintenance and handling of rats were in accordance to the international conventional standard guidelines and with the Helsinki declaration for use of laboratory animals.

**Preparation of hydroethanolic extracts**

Hydroethanolic extracts were prepared as described in (Liu et al., 2013). Firstly, stigmas and the bio-residues of plant (*Crocus sativus*) were separated manually. Fresh tepals or dried and ground stigmas or leaves were macerated with ethanol/water 80/20 (v/v) for 24h in the dark at room temperature. After this first extraction, the solvent was filtered (0.45µm), and the marc was recuperated for second extraction. The procedure was repeated 3 times and the total hydroethanolic phase was subsequently evaporated under reduced pressure at 37°C. Finally, the resulting extracts were kept as solid at −20°C until use.

**STZ-induced diabetic rats**

Diabetes was induced by STZ according to method of Su et al. (2006). Briefly, STZ (60mg/kg, freshly prepared in 0.1M citrate buffer pH 4.5) was injected intraperitoneally to animals following a 16h fast for induction of diabetes. After 72h the blood was extracted from the tail vein for the measuring of fasting glucose levels using glucose oxidase method. Only those rats with blood glucose levels situated between 2 and 5g/l) were considered diabetic and taken for further experimentation.

**Experimental design**

Hydroethanolic extracts (stigmas, tepals and leaves) and glibenclamide were administered orally in aqueous solution daily using an intragastric tube for 21 days. In the present study, 36 rats (30 diabetic and 6 normal rats) were used. The rats were divided into six groups. Six rats were used in each group as follows: group 1: normal control rats (N), received distilled water (1ml); group 2: diabetic control rats (D), received distilled water (1ml); group 3: treated diabetic rats (TPL), received tepal extract (250mg/kg); group 4: treated diabetic rats (STG), received stigma extract (50mg/kg); group 5: treated diabetic rats (LF), received leaf extract (250mg/kg) and group 6: treated diabetic rats (GLB), received glibenclamide (2mg/kg) Blood glucose level and body weights were recorded at weekly intervals. Blood glucose level was determined by the GOD-POD method after prior fasting for 12h. After 21 days of treatments, blood samples were drawn from abdominal aorta into tubes containing heparin. Blood samples were centrifuged at 1300g for separation of plasma and stored at -20°C until biochemical assays. Urines were collected during 24h at the final of treatment using metabolic cages.

**Biochemical assays**

Separated plasma samples were used for determination of glucose and total cholesterol by the oxidase-peroxidase method (Trinder, 1969; Allain et al., 1974), triglycerides by the GPO-PAP method (Bucolo and David, 1973), urea by urease-GLDH method (Talke and Schubert, 1965), creatinine by Jaffe reaction without deproteinization method (Henry, 1968), AST and ALT by IFCC method (Karmen et al., 1955), using common clinical diagnostic kits.

**Statistical analysis**

The data were expressed as mean±SEM, with n=6 for each group. Normality test of data indicated that all data have normal distribution. Statistical analysis of data was performed using one-way ANOVA using GraphPad Prism 5.0 statistical software. Differences between treatment groups were analyzed by Tukey's
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Results

**Effect on body weight**

The effects of tepals, stigmas and leaves on the body weight are shown in Table 1. The untreated diabetic rats showed a significant weight loss ($P<0.001$) compared to the normal control group. Glibenclamide (GLB, 2 mg/kg/day), tepals (250 mg/kg/day), stigmas (50 mg/kg/day) and leaves (250 mg/kg/day) significantly protect against weight loss in STZ diabetic rats at 1st, 2nd and 3rd week of treatment ($P<0.001$, Table 1) compared to the untreated diabetic rats.

At the 3rd week, a significant difference ($P<0.001$) was observed on body weight between group treated with STZ induced diabetic rats (-25.8±4.98g) compared to the normal rats (+38±3.45g). However, the administration of GLB had significantly ($P<0.001$) prevented the decrease of body weight in diabetic rats (+28±2g). Also, the treatment with TPL, STG and LF induce a significant prevent ($P<0.001$) of decreased body weight in diabetic rats (+21.96±4.07g, +13±5.15g and +10.8±1.11g, respectively).

**Effect on water intake and urine elimination**

Table 2 shows the water intake and urine elimination in normal and diabetic rats after 20 days of treatment. The untreated diabetic rats showed a significant increase ($P<0.001$) of water intake and urine elimination compared to the normal group. The treatment with tepals (250 mg/kg/day), stigmas (50 mg/kg/day) and leaves (250 mg/kg/day) extracts decreased significantly water intake ($P<0.001$ and $P<0.01$, respectively) and urine elimination ($P<0.01$ and $P<0.05$, respectively). While leaves extract decreased significantly ($P<0.05$) water intake compared to untreated diabetic rats. Administration of glibenclamide (2 mg/kg/day) showed significant reduction of water intake ($P<0.001$) and urine elimination ($P<0.01$).

**Effect on hyperglycemia**

Figure 1 summarizes the levels of glucose in normal
and diabetic animals. The diabetic rats exhibited significant \( (P<0.001) \) hyperglycemia compared to the normal rats. After 3 weeks the tepals, stigmas and leaves extracts decreased blood glucose levels in the diabetic rats compared with the untreated diabetic rats (Fig. 1). Tepals, stigmas and glibenclamide significantly decreased glucose in STZ diabetic rats at 1st, 2nd and 3rd week of treatment \( (P<0.01, P<0.001 \) and \( P<0.001, \) respectively). While, leaves extract reduced significantly blood glucose level at 2nd and 3rd week of the study \( (P<0.01 \) and \( P<0.001, \) respectively) compared to untreated diabetic rats (Fig. 1).

**Effect on plasma triglycerides and total cholesterol**

Figure 2 (A and B) shows the plasma levels of total cholesterol (A) and triglycerides (B) in normal and experimental animals in each group. The STZ-diabetic rats showed a significant increase \( (P<0.001) \) in plasma cholesterol and triglycerides levels compared to the normal group. The administration of the tepals, stigmas, and leaves extracts significantly decreased plasma total cholesterol \( (P<0.01, P<0.01 \) and \( P<0.05, \) respectively) and plasma triglycerides \( (P<0.001, P<0.01 \) and \( P<0.05, \) respectively). There was a significant \( (P<0.001) \) reduction in plasma cholesterol and triglycerides of rats diabetic treated with glibenclamide compared to untreated diabetic rats.

**Effect on creatinine and plasma urea levels**

On day 21, the plasma levels of creatinine and urea in normal and diabetic animals in each group was showed in Figure 3 (A and B). Creatinine and urea levels in plasma were increased in untreated diabetic rats compared with the normal group \( (P<0.01) \). The administration of the tepals, stigmas and leaves extracts significantly decreased plasma creatinine \( (P<0.01, P<0.01 \) and \( P<0.01, \) respectively). In addition, rats treated with tepals and stigma showed a significant decrease in plasma levels of urea \( (P<0.05 \) and \( P<0.05, \) respectively). While, there was no significant decrease on plasma levels of urea in group treated with leaves. Equally, there was a significant \( (P<0.05) \) reduction in plasma creatinine and urea of rats diabetic treated with glibenclamide (Figs. 3A and B).

**Effect on plasma ALT and AST levels**

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\text{Fig. 1. Effect of } \textit{Crocus sativus} \text{ tepals (TPL, 250mg/kg), stigmas (STG, 50mg/kg) and leaves (LF, 250mg/kg) on blood glucose levels, compared to glibenclamide (GLB, 2mg/kg) in STZ induced-diabetic rats during 21 days of treatment. The results were expressed by: mean±SEM (n=6); **P<0.001 compared with normal control rats (N); *P<0.01; ***P<0.001 compared with diabetic control rats (D).} 
\]
Figure 4 (A and B) shows the plasma levels of ALT and AST in normal and experimental animals in each group. The STZ-diabetic rats showed a significant increase \((P<0.001)\) in the level of plasma ALT and AST compared with the normal control group. The administration of tepals, stigmas and leaves extracts significantly decreased plasma ALT \((P<0.001, P<0.01\) and \(P<0.05\), respectively) and plasma AST \((P<0.001, P<0.01\) and \(P<0.05\), respectively). There was a significant \((P<0.01)\) reduction in plasma ALT and AST of diabetic rats treated with glibenclamide (Figs 4A and B).

**Discussion**

The present data indicated that the tepals and stigmas extracts significantly prevented weight body loss and protected against elevation of water intake, urine elimination, blood glucose, plasma triglycerides, cholesterol, urea, creatinine, AST and ALT levels in treated diabetic rats compared with untreated diabetic rats. Moreover, leaves extract significantly prevented weight body loss. It decreased all other parameters. While, the latter shows that there was no significant decrease on urine elimination volume and plasma levels of urea.

The results observed after the injection of streptozotocin which indicate a polyphagic state and weight loss that could be due to excessive degradation of the protein tissue (Chatterjea and Shinde, 2011; Altinoz et al., 2014; Samarghandian et al., 2014) and may be due to dehydration and
catabolism of fats and proteins (Hakim et al., 1997). However, administration of tepals, stigmas and leaves extracts prevented decreasing body weight in STZ-diabetic rats. This might be due to a significant control of the state of hyperglycemia in the diabetic rats. A reduction in blood glucose levels may improve body weight in rats with streptozotocin diabetes (Kamalakkanan et al., 2003; Babu and Prince, 2004). Our results agreed with Shirali et al. who reported that the aqueous extract of saffron had a protective effect against body weight loss compared to untreated diabetic rats (Shirali et al., 2012). Moreover, Administration of plants polyphenols affect a significant increase in the body weight (Aybar et al., 2001; Patel et al., 2011).

The untreated diabetic rats drank a large amount of water and produced a correspondingly high urine volume. The treatment with tepals, stigmas and leaves extracts decreased water intake and urine elimination. Additionally, Lahmass et al. showed that stigma aqueous extract regulate water intake in tartrazine induced diabetic rats treated with saffron stigma (Lahmass et al., 2017c). Furthermore, It was indicated that polyphenols of plants reduced water intake compared to diabetic rats (Rajagopal and Sasikala, 2008; Ong et al., 2011).

Streptozotocin has been widely used for inducing diabetes mellitus in a variety of animals by affecting degeneration and necrosis of pancreatic b-cells (Merzouk et al., 2000).

Several studies indicated that the level of plasma glucose increases and the level of plasma insulin decreases in STZ-induced diabetic rats (Akbarzadeh et al., 2007).

The present results agreed with Mohajeri et al.(2008) who revealed that administration of the ethanolic saffron extract (20, 40 and 80 mg/kg) has significantly decreased blood glucose and increased plasma insulin in diabetic rats by increasing the number of β-cells in the pancreas (Mohajeri et al., 2008).

Another study also showed that saffron extract has hypoglycemic effects on healthy male rats (Arasteh et al., 2010).

Oral administration of saffron extract has significantly increased plasma insulin and reduced blood glucose levels in alloxan induced diabetic rats. In addition, Histological study showed that pancreas sections of diabetic rats treated with saffron had hypertrophy and hyperplasia of β-cells of islets of Langerhans associated with pyknosis of their nuclei (Elgazar et al., 2013).

A recent study in our laboratory suggests that saffron has curative (antidiabetic) and protective (antidiabetogenic) effect against diabetes induced by tartrazine via reducing blood glucose level and creatinine (Lahmass et al., 2017b).

Several studies indicated that crocin has an antihyperglycemic effect (Rajaei et al., 2013; Shirali et al., 2013; Asri-Rezaei et al., 2015). Besides, administration of safranal affects a significant decrease in glucose, and improve the antioxidant system (Samarghandian et al., 2013).

The anti-hyperglycemic effect of petals and leaves may be due to flavonoid compounds. Consumption of
flavonoids rich foods may reduce the risk of diabetes (Bahadoran et al., 2013). This class of compounds protect from diseases like obesity, diabetes and their complications (Vinayagam and Xu, 2015), regulate the enzymes and hormones and help to maintain blood glucose levels. Improve insulin secretion, glucose uptake, lipid profile and immune system (Hanhineva et al., 2010; Hajjaghaalipour et al., 2015). Kaempferol derivatives were the most abundant flavonoids on tepals and leaves (Goupy et al., 2013; Sánchez-Vioque et al., 2016; Tuberoso et al., 2016). Oral administration of kaempferol to diabetic rats has resulted a significant reduction in plasma glucose and an increase in plasma insulin, back to near normal levels in plasma lipid peroxidation products, enzymatic, and non-enzymatic antioxidants (Al-Numair et al., 2015). Additionally, kaempferol improved insulin secretion and synthesis in β-cells, protected pancreatic beta-cells from hyperglycemia-induced apoptosis and dysfunction (Zhang and Liu, 2011).

Diabetic patients suffer with hyperlipidemia. Furthermore, it was demonstrated that insulin deficiency in DM caused a lot of perturbation in metabolic processes. Consequently, cholesterol storage problems result an increased lipids such as TC and TG levels (Goldberg, 1981). Our results confirm this notion as the treatment with streptozotocin (60 mg/kg, i.p.) caused a significant increase of plasma TC and TG levels. In fact, treatment with stigmas, tepals and leaves significantly decreased the cholesterol and triglycerides levels compared to untreated STZ-diabetic rats. Shirali et al. (2013) showed that crocin in stigma extract has significantly decreased the levels of triglyceride and total cholesterol in the diabetic rats by improving insulin resistance in the diabetic rats (Shirali et al., 2013).

In addition, Samarghandian et al. (2013), indicated that safranal inhibits elevation of the plasma lipid level by controlling oxidative and nitrosative stress (Samarghandian et al., 2013). The aqueous extract of Crocus sativus tepals has been shown to improve dyslipidemia and insulin resistance in obese rats fed a high-fat diet (Hoshyar et al., 2016).

The beneficial effect of the tepals and leaves extract may be due to their richness on polyphenols and flavonoid compounds. The elevation of production of ROS caused an increase of oxidative stress on the kidney. That is the result of increased uptake of glucose (Forbes et al., 2008). It was demonstrated that the increase of plasma urea and creatinine levels in diabetics are linked to renal dysfunction (Almdal et al., 1986).

Our data showed that stigma, tepals and leaves extracts decreased the plasma creatinine level in diabetic rats. Also, rats treated with extracts of tepals and stigma showed a significant decrease in urea plasma levels. Well, there was no significant decrease on plasma levels of urea in the group treated with the leaves.

Also, Kianbakht and Hajiaghaee (2011) suggested that saffron may have anti-hyperglycemic effect without renal toxicities in the alloxan - diabetic rats. It has also been reported that crocin administration could be considered as a valuable adjunct therapy for prevention against renal complication due to diabetes by its potential role in defense against free radicals (Rajaei et al., 2013). A protective effect of tepals and leaves of Crocus sativus against the increase of plasma creatinine and urea levels, may be due to bioactive compounds. Various studies suggest that polyphenols compounds of plants may protect against renal dysfunction in diabetes models by decreasing oxidative stress or decreasing blood glucose (Sabu and Kuttan, 2002; Kumar et al., 2011; Shivanna et al., 2013).

A specific indicator of liver damage is the elevation of ALT and AST activities (El-Demerdash et al., 2001). This elevation caused an increase on ketogenesis and gluconeogenesis observed in diabetes. These enzymes (ALT and AST) degrade the amino acids into P-keto acid, following metabolism pathways via the Krebs cycle (Maiti et al., 2004). Our results showed that administration of stigma, tepals and leaves decreased significantly those enzymes levels in plasma compared with untreated STZ-diabetic rats. The present data agreed with Altinos et al. who revealed that crocin (active constituent of saffron stigma) decreased serum ALT and AST levels compared to untreated STZ-induced diabetic rats. It may protected the liver tissue by decreasing the oxidative stress (Mohammad et al., 2011; Altinoz et al., 2014).

The effect of the tepals and leaves extract may be due to polyphenols and flavonoids compounds. Flavonoids compounds effectively protected the liver
against STZ-induced damage in rats. Therefore, application of flavonoids compounds decreased serum ALT and AST levels compared to untreated STZ-induced diabetic rats. There are reports indicating that several medicinal plants such as agyanom mixture, bolex bitters and remedia mixture can reduce these liver enzymes markers (Girish et al., 2009; Akande et al., 2010). Polyphenolic compounds and flavonoids can improve metabolism and protect cells against destructive effect of free radicals by increasing antioxidant enzymes capacity such as catalase, superoxide dismutase and glutathione peroxidase (Baer-Dubowska et al., 1998; Prakash et al., 2007).

### Conclusion

Diabetes complications are probably linked to oxidative stress which is the result of increased blood glucose levels. Based on our data, the oral administration of tepals, stigmas and leaves extract of *Crocus sativus* reduces blood glucose levels and improves control of diabetes complications that are hyperglycemic results. In addition, the antidiabetic effect of these by-products may be due to the presence of bioactive molecules with antioxidant properties which may exert various health benefits.

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### Conflict of interest

The authors declare no conflict of interest.

### References


Baer-Dubowska W, Szaefer H, Krajka-Kuzniak V. Inhibition of murine hepatic cytochrome P450 activities by natural and synthetic phenolic compounds. Xenobiotica 1998; 28: 735-43.


Farahmand SK, Samini F, Samini M, Samarghandian S. Safranal ameliorates antioxidant enzymes and...


Papandreu MA, Tsachaki M, Eftihimopoulos S, Cordopatis P, Lamari FN, Margarity M. Memory enhancing effects of saffron in aged mice are correlated with antioxidant protection. Behav Brain Res 2011; 219: 197-204.


Vinayagam R, Xu B. Antidiabetic properties of dietary flavonoids: a cellular mechanism review. Nutr Metab
Antidiabetic effect of *Crocus sativus* by-products

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(Lond) 2015; 12: 60.


Zheng CJ, Li L, Ma WH, Han T, Qin LP. Chemical constituents and bioactivities of the liposoluble fraction from different medicinal parts of *Crocus sativus*. *Pharm Biol* 2011; 49: 756-63.