Can J Diabetes 42 (2018) 31–35

Contents lists available at ScienceDirect

Canadian Journal of Diabetes

journal homepage: www.canadianjournalofdiabetes.com

Original Research

The Effects of Natural Clinoptilolite and Nano-Sized Clinoptilolite Supplementation on Glucose Levels and Oxidative Stress in Rats With Type 1 Diabetes

Behnoosh Hossein Nia MSc [a], Sirous Khorram PhD [b], Hassan Rezazadeh PhD [c], Abdolrasol Safaiyan MSc [d], Ali Tarighat-Esfanjani PhD [e,*]

[a] School/Faculty of Nutrition and Food Sciences, Tabriz University of Medical Sciences, Tabriz, Iran
[b] Research Institute for Applied Physics and Astronomy, University of Tabriz, Tabriz, Iran
[c] Department of Pharmacology and Toxicology, School of Pharmacology, Tabriz University of Medical Sciences, Tabriz, Iran
[d] Road Traffic Injury Research Center, Department of Biostatistics and Epidemiology, Tabriz University of Medical Sciences, Tabriz, Iran
[e] Nutrition Research Center, Faculty of Nutrition, Tabriz University of Medical sciences, Tabriz, Iran

Key Messages

- The medical application of clinoptilolite (CLN) as an immune-enhancing and antioxidant agent was reported, but CLN and nano-sized CLN (NCLN) use in diabetes mellitus were not investigated in previous studies.
- The glucose-lowering effect of NCLN in rats with type 1 diabetes was observed in this study.

ARTICLE INFO

Article history:
Received 27 February 2016
Received in revised form
20 January 2017
Accepted 30 January 2017

Keywords:
blood glucose
diabetes mellitus
oxidative stress
rats
zeolites

ABSTRACT

Objective: Oxidative stress has a major role in development of diabetic complications. In this study we investigated whether clinoptilolite and nano-sized clinoptilolite could reduce hyperglycemia and oxidative stress in streptozotocin-induced diabetic rats and attempted to determine which intervention was more effective.
Methods: Thirty-six rats were randomly allocated to 2 groups; 1 group was randomly chosen as a diabetic group and injected with streptozotocin (60 mg/kg body weight in 0.1 mol/L sodium citrate buffer, pH 4.5) to induce diabetes. Three days after diabetes induction, each group (diabetic group and nondiabetic group) was randomly divided into 3 subgroups of 6 animals each ([1] control, [2] 1% clinoptilolite/food, [3] 1% nano-sized clinoptilolite/food). Supplementation was continued for 28 days. Blood glucose was measured 3 times, at the beginning of the study and on the 14th and 28th days. Activity of antioxidant enzymes, including glutathione peroxidase and superoxide dismutase, and levels of total antioxidant capacity, as well as malondialdehyde, were evaluated.
Results: Blood glucose and malondialdehyde were significantly elevated, but there were no statistically significant changes in superoxide dismutase, glutathione peroxidase or total antioxidant capacity in diabetic rats. In diabetic rats treated with nano-sized clinoptilolite, blood glucose decreased to near normal levels (12.4 vs. 27.5 mmol/L). No significant changes were found in the other groups. None of the oxidative stress indices showed significant changes in either the treated or untreated rats.
Conclusion: Nano-sized clinoptilolite exerted a hypoglycemic effect in streptozotocin-induced diabetic rats but had no significant influence on oxidative stress markers.

Mots clés :
Glycémie
diabète sucré
stress oxydatif
rats
zéolites

RÉSUMÉ

Objectif : Le stress oxydatif joue un rôle très important dans l'apparition des complications du diabète. Dans la présente étude, nous avons examiné si la clinoptilolite et la clinoptilolite nanométrique pouvaient réduire l'hyperglycémie et le stress oxydatif chez les rats atteints d'un diabète induit par la streptozotocine, et avons tenté de déterminer l'intervention la plus efficace.

* Address for correspondence: Ali Tarighat-Esfanjani, PhD, Nutrition Research Center, Tabriz University of Medical Sciences, Attar Nishabouri St., PO Box 14711, Tabriz, I. R. 5166614711, Iran.
E-mail address: Tarighata@tbzmed.ac.ir

1499-2671 © 2017 Canadian Diabetes Association.
The Canadian Diabetes Association is the registered owner of the name Diabetes Canada.
https://doi.org/10.1016/j.jcjd.2017.01.010

Méthodes : Trente-six rats ont été répartis de façon aléatoire dans 2 groupes. L'un des groupes a été désigné de façon aléatoire pour constituer après injection de streptozotocine (60 mg/kg de poids corporel d'une solution tampon de 0,1 mol/l de citrate de sodium, pH 4,5) le groupe de rats diabétiques. Trois jours après l'induction du diabète, chacun groupe (groupe diabétique et groupe non diabétique) a été divisé de façon aléatoire en 3 sous-groupes de 6 animaux chacun : [1] témoin ; [2] 1 % de clinoptilolite dans les aliments ; [3] 1 % de clinoptilolite nanométrique dans les aliments. La supplémentation a duré 28 jours. La glycémie a été mesurée 3 fois, soit au début de l'étude, au 14e jour et au 28e jour. L'activité des enzymes antioxydantes, dont la glutathion peroxydase et la superoxyde dismutase, ainsi que la capacité antioxydante totale et le malondialdéhyde ont été évalués.
Résultats : La glycémie et le malondialdéhyde se sont avérés significativement élevés, mais aucun changement statistiquement significatif n'a été observé dans la superoxyde dismutase, la glutathion peroxydase ou la capacité antioxydante totale chez les rats diabétiques. Chez les rats diabétiques traités par clinoptilolite nanométrique, la glycémie se situait près des concentrations normales (12,4 mmol/l *vs* 27,5 mmol/l). Aucun changement significatif n'a été observé dans les autres groupes. Aucun des indices de stress oxydatif n'a montré des changements significatifs ni chez les rats traités ni chez les rats non traités.
Conclusion : La clinoptilolite nanométrique a exercé un effet hypoglycémique chez les rats atteints d'un diabète induit par la streptozotocine, mais n'a eu aucune influence significative sur les marqueurs du stress oxydatif.

Introduction

Diabetes mellitus, which has become one of the most drastic metabolic disturbances in the world (1), is diagnosed by absolute or relevant deficiencies in insulin secretion and/or insulin action, followed by chronic hyperglycemia. Long-term complications of diabetes—including ocular, renal and cardiovascular disorders—are primarily due to hyperglycemia (2). Increasing evidence suggests that chronic hyperglycemia can cause excessive production of free radicals, particularly reactive oxygen species (ROS) (3–5). Furthermore, the role of free radicals in tissue damage of streptozotocin (STZ)–induced diabetic rats has been explained (6). On the other hand, several studies have shown that free radicals cause lipid peroxidation and aggregation of lipid peroxidation products in subjects with diabetes mellitus (7). In addition, the role of genetic susceptibility and epigenetic factors in development of diabetes and its complications has been demonstrated (8–10).

Hypoglycemic pharmacologic agents are unable to control tissue damage in diabetes; they can also cause severe side effects (6). Hence, researchers are motivated to seek remedies in traditional medicine that have milder toxicity than available synthetic drugs. Natural products from various sources, such as plants, animals and microorganisms, tend to be potential candidates for drug development (11–15). Nanotechnology might serve as a tool in diagnosis and treatment of diabetes, as well (16).

Clinoptilolites (CLNs), the most abundant natural zeolitic minerals, are hydrated natural or synthetic microporous crystals comprising AlO_4 and SiO_4 tetrahedra, linked through the common oxygen atoms (17). They are among natural remedies proposed to reduce blood sugar (18). Some CLNs are already applied in medicine as antidiarrheal, bactericidal and antifungal medications and are also used for healing wounds (19). They promote the function of the immune system by enhancing the expression of cytokines such as interleukin 1alpha, interleukin-6 and tumour necrosis factor -α (20).

Current evidence suggests that zeolites may reduce blood glucose in nondiabetic animal models (21). They may act as glucose adsorbents, so individuals with diabetes mellitus could benefit from them (22). CLNs might reduce blood glucose by preventing beta-cell destruction and thus enhancing insulin production (19).

At present, the antioxidative roles attributed to CLNs are based on their ability to diminish free radicals and lipid peroxidation levels as well as increase total antioxidant capacity (TAC) in serum. Hence, antioxidant enzymes may be potential targets of CLNs' action as well (23). In comparison with traditional CLNs, nano-sized CLNs (NCLNs) are proposed to have greater specific surface area. An increase in the catalyst surface area of CLNs increases the quantity of substances available to react and thus improves the rate of the reaction, which might positively influence process efficiency (24).

CLNs, the most abundant natural zeolitic minerals, have been widely used because of their abundance and considerably low cost. In addition, CLNs have exhibited no toxicity and cause no environmental pollution. These properties have sparked the interest of researchers. The present study was carried out to determine the efficacy of a natural CLN and NCLN in reducing blood glucose levels and oxidative stress in experimental diabetic rat models.

Methods

Natural zeolite (Afrazand Co., Tehran, Iran) is a sodium/potassium CLN with a particle size of approximately 5 µm. The chemical composition of CLN is presented in Table 1. The NCLN particles were produced by using the glow discharge plasma method, which is a novel iron-impregnated nanocatalyst for the heterogeneous Fenton process (24). Particle analysis of the NCLNs showed that generated nanoparticles' size was in the range of 30 to 40 nm in diameter.

Preparation of feeds

Animal feeds were purchased from Behparvar Company (Tehran, Iran). The standard rat food was mixed with 1% CLN or 1% NCLN powder. Tap water was added to the mixture of materials to make a dough, which was then pelleted and left to dry at room temperature.

Chemicals

STZ (Sigma Chemicals, St. Louis, MO, USA), diethyl ether and other solvents and buffers (Merck KGaA, Darmstadt, Germany) were used in this project.

Experimental animals

Male Wistar rats, aged 6 months (weight >0.25 kg), were procured from the Laboratory Animals Unit of Azad University of Marand

Table 1
Elemental composition of clinoptilolite and nano-sized clinoptilolite

	Weight (%)			Mole/ratio			
	Na	Al	Si	K	Si/Al	Na/Al	K/Al
CLN	3.58	7.07	60.33	0.72	8.28	1.25	0.15
NCLN	8.86	4.81	44.27	10.94	8.88	4.52	3.23

Al, aluminum; *CLN,* clinoptilolite; *K,* potassium; *Na,* sodium; *NCLN,* nano-sized clinoptilolite; *Si,* silicon.
Note: Adapted with permission from Table 2 in "Khataee A, Bozorg S, Khorram S, et al. Conversion of natural clinoptilolite microparticles to nanorods by glow discharge plasma: A novel Fe-impregnated nanocatalyst for the heterogeneous fenton process. Ind Eng Chem Res 2013;52;18225–33."

Branch, Tabriz, Iran. All rats were housed under climate-controlled conditions for 1 week before the study in the centre for breeding and maintenance of laboratory animals in the Faculty of Pharmacy, Tabriz University of Medical Sciences.

Animals were kept in standard metal cages under controlled conditions: a 12-hour light-dark cycle, temperature of 23°C to 25°C and relative humidity of 30% to 50%. Rats had unrestricted access to common chow (pellets) and water. Fundamental guidelines for the care and use of laboratory animals were followed. Ethical approval was obtained from the ethics committee of Tabriz University of Medical Sciences, Tabriz, Iran.

Experimental design

Thirty-six rats were randomly allocated to 2 groups, and 1 group was randomly chosen to be a diabetic group and injected with STZ (60 mg/kg body weight in 0.1 mol/L sodium citrate buffer, pH 4.5). Rats with blood glucose levels >13.87 mmol/L were considered diabetic. Three days after diabetes induction, each group (diabetic and nondiabetic) was randomly divided into 3 subgroups of 6 animals each ([1] control, [2] 1%CLN/food, [3] 1% NCLN/food). Animals were fed a diet containing 1% powdered CLN or NCLN and a normal diet for 4 consecutive weeks. At the end of the experiment (on the 28th day), animals were anesthetized with dimethyl ether, and blood samples were collected instantly from the orbital sinus of each animal in each group after a 12-hour fast. After blood collection, plasma was separated by centrifugation at 3000 rpm for 10 minutes and analyzed to estimate the amounts of TAC, malondialdehyde (MDA), glutathione peroxidase (GPX), and superoxide dismutase (SOD). Blood glucose levels were determined from tail vein blood samples, using an Accu-Chek glucometer (Roche, Mannheim, Germany) at 3 time points—at the beginning of the study and on days 14 and 28.

Biochemical analysis

Amounts of TAC and MDA and the activity of GPX and SOD enzymes were determined using commercial kits (Randox, Crumlin, County Antrim, United Kingdom) and the spectrophotometric method. All experiments were carried out at the research laboratory of the Nutrition Research Center at Tabriz University of Medical Sciences.

Statistical analysis

The potential confounding variables were adjusted between the 2 groups (diabetic and nondiabetic) in the 3 subgroups (control, CLN, NCLN) on the first day before the experiments started. Two-way analysis of variance was used for comparing MDA, TAC, SOD and GPX between diabetic factors (diabetic and nondiabetic) and treatment factors (control, CLN, NCLN). For repeated-measures data, Mauchly's W test was first checked for identity covariance matrix, and a repeated-measures test was done by using Minitab Software version 17. The results include 4 p values: the first was the group p value for comparison of the diabetic and nondiabetic groups; the second was the treatment p value for comparison of treatment factors in each group; the third was the interaction p value for recognition of the interaction effect between the diabetic factor and the treatment factor; and the fourth was the time p value for comparison of variations among the 3 assessment time points. Sidak tests were used for multiple comparisons. The level of significance was set at 0.05, and all results were expressed as means±standard error of the mean.

Results

Treatment with STZ resulted in significant elevation of blood glucose levels ($p\alpha<0.001$), compared with those in normal control rats (Table 2). A significant reduction in plasma glucose level was observed in the NCLN group when compared with untreated rats in the experimental period of 28 days ($p<0.001$), but CLN-treated diabetic rats had no significant reductions in blood glucose levels in comparison with untreated diabetic animals. As shown in Table 2, blood glucose levels decreased in the CLN-treated healthy rats during the study ($p<0.05$). Blood glucose levels differed significantly between groups across time ($p<0.001$).

Injection of STZ did not create a significant difference in TAC, SOD or GPX between the study groups (Table 3). It is clear from the data in Table 3 that STZ injection significantly increased lipid peroxidation product (MDA) in the serum of diabetic rats 4 weeks after diabetes induction ($p\alpha<0.001$). Although significant differences were not found between the experimental groups, the data reveal that CLN reduced MDA levels more than NCLN in the normal group ($p<0.05$), but MDA levels mildly increased in both CLN- and NCLN-supplemented diabetic rats.

Discussion

In this study, the STZ-treated rats were certified to be hyperglycemic 72 hours after STZ administration. Treatment with NCLN significantly decreased blood glucose levels in diabetic rats compared with the untreated diabetic rats. Besides, CLN mildly reduced blood glucose levels in nondiabetic rats. Thus it can be inferred that CLN exerts mild glycemic control in healthy rats.

Substantial evidence suggests that macrophages play an important role in development of type 1 diabetes in animal models (25). In one study it was reported that silica could almost perfectly prevent the advance of spontaneous diabetes and hyperglycemia in young BioBreeding rats (26). There are also reports that indicate a protective effect of silica against islet demolition through depletion of

Table 2
Effect of CLN and NCLN supplementation on blood glucose levels in streptozotocin-treated diabetic rats

Groups (n=6)	Treatment	Blood glucose levels (mmol/L)			Repeated-measures ANOVA p values			
		Day 0	Day 14	Day 28	Diabetes	Treatment	Interaction	Time
Nondiabetic	Control	4.1±0.26	4.2±0.29	4.4±0.26				
	CLN	4.8±0.25	4.7±0.12	4.0±0.10*				
	NCLN	4.6±0.26	4.7±0.15	4.6±0.15				
Diabetic	Control	28.7±0.80	27.6±1.71	31.2±1.60	<0.001	<0.001	<0.001	<0.001
	CLN	27.2±0.98	29.2±1.41	29.2±0.43				
	NCLN	27.5±1.24	29.8±1.68	12.4±5.53†				

Note: Data are expressed as means ± standard error of the mean.
ANOVA, analysis of variance; *CLN*, clinoptilolite; *NCLN*, nano-sized clinoptilolite.
* Sidak tests $p<0.05$ compared with nondiabetic NCLN group and time 0, 14.
† $p<0.001$ compared with diabetic control and CLN groups and time 0, 14 days.

macrophages or modification of macrophage action on T lymphocytes (27). In this study the hypoglycemic activity exhibited by NCLNs may be attributed to the presence of the compound silica. Because of the well-known adsorption properties of zeolites, they can reduce the glucose level in blood (28). Many published studies have demonstrated component adsorption of fructose, glucose, sucrose and fructooligosaccharides by NaX-type zeolites and also adsorption of glucose into zeolite beta from aqueous solution (29–31). One study of type 1 diabetes in a nonobese diabetic animal model showed that Ca^{2+}-zeolite used in combination with insulin could modify hyperglycemia through deterrence of the sodium-glucose transporter (32). It has been observed that dietary administration of CLN could alleviate nitrates' deleterious effects, including hyperglycemia and impairment of protein metabolism (33). In our study the hypoglycemic effect of NCLNs could not be due to increased absorption characteristics and catalyst surface area of CLN. The lack of a hypoglycemic effect of zeolites seems to be due to the low dose administered.

Administration of STZ may cause most of the diabetic complications associated with oxidative stress in animals (34, 35). Diabetes-induced oxidative stress is accompanied by a reduction in antioxidant status, which can increase the detrimental effects of free radicals (36). Defense mechanisms against free radicals consist of enzymes SOD, GPX and catalase, as they eliminate superoxide, hydrogen peroxide and hydroxyl radicals (37). Studies conducted with diabetic animal models, especially STZ-induced diabetic rats and mice, have demonstrated that hyperglycemia increases the rate of hydroxyl radical production, and this has been associated with the level of thiobarbituric acid (38).

Sources of oxidative stress in diabetes could originate from several pathways consisting of glycation reactions and a shift in the decreased oxygen status of the diabetic cells. Oxidative stress in diabetes could be clearly demonstrated by the extent of some specific biomarkers such as lipid hydroperoxides, protein carbonyls and DNA adducts (39). Antioxidants invert many of the effects of hyperglycemia on endothelial functions such as delayed cell replication and reduced endothelial-related relaxation.

Antioxidant enzymes play an important role in scavenging ROS to control lipid peroxidation (40).

In the present study, 28 days after STZ injection (60 mg/kg body weight), we found no significant difference in the activity of SOD and GPX or the level of TAC in STZ-induced diabetic rats when compared with the healthy animals. Our results show that the injection of STZ significantly increased the level of MDA in diabetic rats at the end of the experiment; however, the increase observed in the treatment groups can be attributed solely to either diabetes or zeolite. MDA is an aldehyde produced from lipid peroxidation, which is estimated based on its reaction with thiobarbituric acid and is used as the best existing measure of universal ROS, substantially elevated in diabetes (41). The lack of significant change in oxidative stress parameters (TAC, SOD and GPX) observed in this study may have been due to the short treatment period (28 days) (42). However, in our experimental model of diabetes mellitus, STZ administration led to a nonsignificant increase in plasma SOD activity. Some studies have shown decreased SOD activity after zeolite administration (43), while others have shown increases (44). Recently, it has been reported that pretreatment of SH-SY5Y neuronal-like cells with zeolite resulted in a significant reduction of ROS-induced cell death. On the other hand, mice exposed to zeolite at the time of initial plaque accumulation had a significant increase in SOD activity in their hippocampi compared with age-matched control mice (45). It has been reported that CLN administration (5 mg/kg) orally for 10 days in rats with partial hepatectomy decreased oxidant activity through elevation of liver tissue copper-zinc SOD activity and Glutathione (GSH) levels and reduction of plasma and liver tissue MDA levels (46). In addition, administration of 3 g of natural zeolite chabazite/phillipsite/analcime to clinically healthy nonsmokers and smokers for 4 weeks increased the levels of antioxidant enzymes able to remove ROS, including GSH peroxidase, SOD GSH reductase and reduced thiobarbituric acid reactive substance (TBARS), thus preventing or slowing down oxidative damage (23). An increased concentration of lipid-bound sialic acid in serum but a reduced amount of melanoma metastasis and lipid peroxidation in the liver have been suggested after use of zeolite for 28 days in mice with melanoma cells. Furthermore, zeolite was observed to fully suppress generation of nitric oxide (17). Effects of CLN treatment in reducing lipid peroxidation levels and recovering the levels of catalase, SOD and GPX activity during pb^{2+} toxicity have been reported (22, 47).

CLN was used in our study for the following reasons. CLN is the most abundant zeolite in Iran, and it has a total cation-exchange capacity of about 200 mEq. The exchange capacity plays an important role in the therapeutic use of zeolite, accounting for its ability to release useful elements while capturing and binding others (23).

Exact mechanisms of zeolites' antioxidant effects are not well known. Because zeolite is not absorbed into the blood from the gut, evidence suggests that the effects of zeolite may be due to an indirect interaction with biochemical systems, removal of waste and toxins from the gut, improvement of the immune system through the mucosal-related intestinal lymphoid tissue, and an increase in the bioavailability of minerals that are important co-factors for some enzymes (48, 49). In addition, it has been proposed that zeolites tend to neutralize the solutions and exchange electrons and also that zeolites, such as CLN, may modify the disorders in a redox state and arrest the generation of peroxides and free radicals through their amphoteric character, as in the CLN–iron oxidase system (46, 50). Some of the antioxidant properties of zeolites are attributed to their effect on macrophages' phagocytic function triggered after phagocytosis of zeolite particles. Phagocytic action subsequently leads to the production of cytokines such as tumour necrosis factor-α, which stimulates immunologic responses and also increases the expression of SOD (22).

Table 3
Effect of CLN and NCLN on oxidative stress parameters in serum of streptozotocin-treated diabetic rats

	Nondiabetic groups			Diabetic groups			Two-way ANOVA p value values		
Treatment	Control	CLN	NCLN	Control	CLN	NCLN	Pα	Pβ	Pθ
MDA (nmol/mL)	4.7±0.11	3.9±0.35*	4.4±0.29	4.2±0.13	5.6±0.31†	5.7±0.41†	<0.01	0.14	<0.01
TAC (mmol/L)	0.7±0.02	0.7±0.02	0.7±0.02	0.6±0.02	0.8±0.10	0.5±0.07	0.16	0.12	0.06
SOD (μg/g Hb)	1237.4±113.42	1354.8±145.19	1184.3±149.55	1568.6±141.23	1553.1±170.1	1348.5±147.13	0.06	0.42	0.83
GPX (μg/g Hb)	32.2±1.51	35.8±1.49	34.7±3.11	35.8±3.19	36.5±1.49	34.1±3.67	0.56	0.68	0.71

Note: Data are expressed as means ± standard error of the mean.
ANOVA, analysis; *CLN,* clinoptilolite; *GPX,* glutathione peroxidase: *Hb,* hemoglobin; *MDA,* malondialdehyde; *NCLN,* nano-sized clinoptilolite; Pα, diabetes p value; Pβ, group p value; Pθ, interaction p value; SOD, superoxide dismutase; TAC, total antioxidant capacity.
* $p<0.05$ compared with nondiabetic control and NCLN groups.
† $p<0.05$ Sidak test compared with diabetic control group.

This study had 2 limitations. First, we did not obtain measurements before interventions and, therefore, could not eliminate rats that had normal oxidative stress status; the lack of significant changes in the oxidative stress indices may be attributed to their normal range in both diabetic and healthy rats. Second, we could not confirm the optimal dose of CLN and NCLN supplementation because of the limited relevant investigations.

Conclusion

The findings of our study demonstrate that oral treatment with NCLN could help improve glycemic status in rats with STZ-induced diabetes, but CLN is slightly effective against oxidative stress in control/healthy rats when compared with other groups. The lack of detectable change in oxidative stress status in diabetic rats may be due to factors such as the short duration of treatment and the low dose of the intervention. Further studies are warranted to clarify antidiabetic beneficial effects of CLN and NCLN and mechanisms mediating these effects on blood glucose.

References

1. Varga ZV, Giricz Z, Liaudet L, et al. Interplay of oxidative, nitrosative/nitrative stress, inflammation, cell death and autophagy in diabetic cardiomyopathy. Biochim Biophys Acta 2015;1852:232–42.
2. Omotayo EO, Gurtu S, Sulaiman SA, et al. Hypoglycemic and antioxidant effects of honey supplementation in streptozotocin-induced diabetic rats. Int J Vitam Nutr Res 2010;80:74–82.
3. Vanessa Fiorentino T, Prioletta A, Zuo P, Folli F. Hyperglycemia-induced oxidative stress and its role in diabetes mellitus related cardiovascular diseases. Curr Pharm Des 2013;19:5695–703.
4. Tiwari BK, Pandey KB, Abidi A, Rizvi SI. Markers of oxidative stress during diabetes mellitus. J Biomark 2013;2013:378790.
5. Busik JV, Mohr S, Grant MB. Hyperglycemia-induced reactive oxygen species toxicity to endothelial cells is dependent on paracrine mediators. Diabetes 2008;57:1952–65.
6. Ashraf H, Zare S. Preventive effects of aqueous extract of Berberis integerrima Bge. Root on liver injury induced by diabetes mellitus (type 1) in rats. Iran J Pharm Res 2015;14:335–43.
7. Gordon LA, Morrison EY, McGrowder DA, et al. Effect of exercise therapy on lipid profile and oxidative stress indicators in patients with type 2 diabetes. BMC Complement Altern Med 2008;8:21.
8. Tibaut M, Petrovič D. Oxidative stress genes, antioxidants and coronary artery disease in type 2 diabetes mellitus. Cardiovasc Hematol Agents Med Chem 2016;14:23–38.
9. Sobrin L, Green T, Sim X, et al. Candidate gene association study for diabetic retinopathy in persons with type 2 diabetes: The Candidate gene Association Resource (CARe). Invest Ophthalmol Vis Sci 2011;52:7593–602.
10. Kruzliak P, Haley AP, Starcevic JN, et al. Polymorphisms of the peroxisome proliferator-activated receptor-γ (rs1801282) and its coactivator-1 (rs8192673) are associated with obesity indexes in subjects with type 2 diabetes mellitus. Cardiovasc Diabetol 2015;14:42.
11. Rashid R, Murtaza G, Khan AK, Mir S. Antioxidant and hypoglycemic effect of Otostegia aucheri methanolic extract in streptozotocin-induced diabetic male long-Evans rats. Acta Pol Pharm 2014;71:631–5.
12. Alam F, Islam MA, Kamal MA, Gan SH. Updates on managing type 2 diabetes mellitus with natural products: Towards antidiabetic drug development. Curr Med Chem 2016;23:1–37.
13. Siew Hua G, Kamal MA, Lima M, et al. Medicinal plants in management of type 2 diabetes and neurodegenerative disorders. Evid Based Complement Alternat Med 2015;2015:686872.
14. Baig M, Ahmad K, Hasan Q, et al. Interaction of glucagon G-protein coupled receptor with known natural antidiabetic compounds: Multiscoring in silico approach. Evid Based Complement Alternat Med 2015;2015:497253.
15. Tan Y, Kamal MA, Wang Z-Z, et al. Chinese herbal extracts (SK0506) as a potential candidate for the therapy of the metabolic syndrome. Clin Sci 2011;120:297–305.
16. Alam Q, ZubairAlam M, Karim S, et al. A nanotechnological approach to the management of Alzheimer disease and type 2 diabetes. CNS Neurol Disord Drug Targets 2014;13:478–86.
17. Pavelic K, Katic M, Sverko V, et al. Immunostimulatory effect of natural clinoptilolite as a possible mechanism of its antimetastatic ability. J Cancer Res Clin Oncol 2002;128:37–44.
18. Warren PW. Information about toxins and liquid zeolits (clinoptilolite). 2006. Available from: https://www.google.com/#q=Information+about+toxins+and+liquid+zeolits [Accessed 1 May 2017].
19. Pavelic K, Hadzija M. Medical applications of zeolites. In: Handbook of zeolite science and technology. New York: Dekker, 2003, pg. 1143–74.
20. Jung B-G, Toan NT, Cho S-J, et al. Dietary aluminosilicate supplement enhances immune activity in mice and reinforces clearance of porcine circovirus type 2 in experimentally infected pigs. Vet Microbiol 2010;143:117–25.
21. Januškevičius A, Januškevičiene G, Plungyte K. Klinoptilolitas suaugusiu sunu racionuose. Veteriin Zootechnika. (Vet Med Zoot) 2013;64(86):1392–2130.
22. Zarcovic N, Zarcovic K, Kralj M, et al. Anticancer and antioxidative effects of micronized zeolite clinoptilolite. Anticancer Res 2003;23:1589–96.
23. Dogliotti G, Malavazos AE, Giacometti S, et al. Natural zeolites chabazite/phillipsite/analcime increase blood levels of antioxidant enzymes. J Clin Biochem Nutr 2012;50:195–8.
24. Khataee A, Bozorg S, Khorram S, et al. Conversion of natural clinoptilolite microparticles to nanorods by glow discharge plasma: A novel Fe-impregnated nanocatalyst for the heterogeneous Fenton process. Ind Eng Chem Res 2013;52:18225–33.
25. Ihm SH, Lee K-U, Rhee BD, Min HK. Initial role of macrophage in the development of anti-β-cell cellular autoimmunity in multiple low-dose streptozotocin-induced diabetes in mice. Diabetes Res Clin Pract 1990;10:123–6.
26. Oschilewski U, Kiesel U, Kolb H. Administration of silica prevents diabetes in BB-rats. Diabetes 1985;34:197–9.
27. Oschilewski M, Schwab E, Kiesel U, et al. Administration of silica or monoclonal antibody to Thy-1 prevents low-dose streptozotocin-induced diabetes in mice. Immunol Lett 1986;12:289–94.
28. Cisneros V, Ocanto F, Linares CF. Ca2+, Mg2+ OR Fe2+ ion-exchanged cancrinite-type zeolites as possible hypoglycemiant agents. Rev. Latinoam. Quím. 2011;39:55–61.
29. Kuhn RC, Mazutti MA, Maugeri Filho F. Kinetic and mass transfer effects for adsorption of glucose, fructose, sucrose and fructooligosaccharides into X zeolite. LWT Food Sci Technol 2012;48:127–33.
30. Bai P, Siepmann JI, Deem MW. Adsorption of glucose into zeolite beta from aqueous solution. AIChE J 2013;59:3523–9.
31. Francisco M, Mlinar AN, Yoo B, et al. Recovery of glucose from an aqueous ionic liquid by adsorption onto a zeolite-based solid. Chem Eng J 2011;172:184–90.
32. Hadzija MP. Inhibition of SGLT using Ca2+-zeolite and preserve the periphery of glucose excess 2014. Eur socy Endocrinol 2014;35:P494.
33. Katsoulos P, Karatzia M, Polizopoulou Z, et al. Effects of prolonged consumption of water with elevated nitrate levels on certain metabolic parameters of dairy cattle and use of clinoptilolite for their amelioration. Environ Sci Pollut Res Int 2015;22:9119–26.
34. Kakkar R, Mantha SV, Radhi J, et al. Increased oxidative stress in rat liver and pancreas during progression of streptozotocin-induced diabetes. Clin Sci 1998;94:623–32.
35. Parihar P, Shetty R, Ghafourifar P, Parihar M. Increase in oxidative stress and mitochondrial impairment in hypothalamus of streptozotocin treated diabetic rat: Antioxidative effect of Withania somnifera. Cell Mol Biol (Noisy-Le-Grand) 2015;62:73–83.
36. Marzouk M, Soliman A, Omar T. Hypoglycemic and antioxidative effects of fenugreek and termis seeds powder in streptozotocin-diabetic rats. Eur Rev Med Pharmacol Sci 2013;17:559–65.
37. Samarghandian S, Borji A, Delkhosh MB, Samini F. Safranal treatment improves hyperglycemia, hyperlipidemia and oxidative stress in streptozotocin-induced diabetic rats. J Pharm Pharm Sci 2013;16:352–62.
38. Francés DE, Ronco MT, Monti JA, et al. Hyperglycemia induces apoptosis in rat liver through the increase of hydroxyl radical: New insights into the insulin effect. J Endocrinol 2010;205:187–200.
39. Baynes JW. Role of oxidative stress in development of complications in diabetes. Diabetes 1991;40:405–12.
40. Ceriello A, dello Russo P, Amstad P, Cerutti P. High glucose induces antioxidant enzymes in human endothelial cells in culture: Evidence linking hyperglycemia and oxidative stress. Diabetes 1996;45:471–7.
41. Qujeq D, Habibinudeh M, Daylmkatoli H, Rezvani T. Malondialdehyde and carbonyl contents in the erythrocytes of streptozotocin-induced diabetic rats. Int J Diabetes Metab 2005;13:96–8.
42. Courteix C, Eschalier A, Lavarenne J. Streptozocin-induced diabetic rats: Behavioural evidence for a model of chronic pain. Pain 1993;53:81–8.
43. Wohaieb SA, Godin DV. Alterations in free radical tissue-defense mechanisms in streptozocin-induced diabetes in rat: Effects of insulin treatment. Diabetes 1987;36:1014–18.
44. Kakkar R, Kalra J, Mantha SV, Prasad K. Lipid peroxidation and activity of antioxidant enzymes in diabetic rats. Mol Cell Biochem 1995;151:113–19.
45. Montinaro M, Uberti D, Maccarinelli G, et al. Dietary zeolite supplementation reduces oxidative damage and plaque generation in the brain of an Alzheimer's disease mouse model. Life Sci 2013;92:903–10.
46. Saribeyoglu K, Aytac E, Pekmezci S, et al. Effects of clinoptilolite treatment on oxidative stress after partial hepatectomy in rats. Asian J Surg 2011;34:153–7.
47. Basha MP, Begum S, Mir BA. Neuroprotective actions of clinoptilolite and ethylenediaminetetraacetic acid against lead-induced toxicity in mice Mus musculus. Toxicol Int 2013;20:201–7.
48. Pavelić K, Hadžija M, Bedrica L, et al. Natural zeolite clinoptilolite: New adjuvant in anticancer therapy. J Mol Med 2001;78:708–20.
49. Ivkovic S, Deutsch U, Silberbach A, et al. Dietary supplementation with the tribomechanically activated zeolite clinoptilolite in immunodeficiency: Effects on the immune system. Adv Ther 2004;21:135–47.
50. Dimirkou A, Doula MK. Use of clinoptilolite and an Fe-overexchanged clinoptilolite in Zn 2+ and Mn 2+ removal from drinking water. Desalination 2008;224:280–92.