Hypoglycemic Effects of Nigella Sativa Extract and Endurance Training in Streptozotocin Induced Diabetic Rats

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Abstract

Objective: Diabetes is a common disease and its prevalence rate is rapidly increasing. Aim of present study was to review the hypoglycemic effects of Nigella sativa (N.sativa) extract and endurance training in streptozotocin induced diabetic rats.

Materials and Methods: In this experimental research from 50 streptozotocine induced diabetic rats, 32 rats with more than 300 mg/dL fasting blood glucose (FBS) selected and divided into four groups of 8 rats (1) endurance training, (2) N.sativa extract, (3) N.sativa extract with endurance training, and (4) control. Groups 1 and 3 trained endurance exercise 4 weeks, 5 sessions per week. Two and 3 groups received 4 weeks intraperitoneal 100 mg/kg N.sativa extract daily. For statistical analysis of data used Kolmogorov–Smirnov test, one way ANOVA and tukey post hoc tests. *P*-value ≤0.05 was considered as significant.

Results: Four weeks endurance training, N.sativa extract and N.sativa extract with endurance trainings have significant effect on reduction of FBS, insulin resistance and HbA1c of diabetic rats (*P*-value≤0.05).

Conclusion: Four weeks N.sativa extract and endurance training have hypoglycemic effects in diabetic rats but they have no interactional effects.

Keywords: Nigella Sativa, Hypoglycemic, Endurance Training

Introduction

Diabetes is a common disease and its prevalence rate is rapidly increasing (1). Diabetes is a chronic disease due to lack of insulin or reduced insulin function, which causes increased blood glucose levels and metabolic disorders (2). Increased levels of glucose over time have adverse effects on small and large vessels. Therefore, control of blood glucose levels of diabetic patients at the appropriate level can be a valuable role in diabetes induced mortality. The aim of all anti-diabetic treatments is to improve the control of blood glucose levels and maintain the health and reduce the potential complications of diabetes (3). Though, at present original and effective treatment for diabetes mellitus is using insulin and blood glucose-lowering agents, However these agents have undesirable effects, such as increased fat reserves, atrophy of adipose tissue at the injection site and hypoglycemia shocks and in long-term do not have effect on debilitating side effects of

diabetes. The cardiovascular risks due to elevated blood glucose, lipids and lipid disorders should be diagnosed and treated quickly as part of a comprehensive treatment of diabetes.[3] Several treatment modalities such as the use of herbal and synthetic drugs or change of lifestyle to treat or control of this disease is recommended to patients (4). During the recent decades in many countries the use of alternative therapies, especially herbal therapy and dietary supplements to improve a variety of diseases such as high blood lipid and glucose, has increased. One of the most important problems facing physicians as well as consumers of medicinal plants is lack of sufficient information regarding drug safety and its effect on the disease. Medicinal plants and their use have been considered in the treatment of diabetes and its complications (5). Due to the effectiveness of herbal medicine in the scientific community and its acceptance in most societies, the herbal medication is growing (1). The role of medicinal plants, with blood glucose lowering properties cannot be ignored in treatment of diabetes. N.sativa is one year old plants of the buttercup family. The Components of N.sativa can be pointed: 1) omega-6, 2) omega-3, 3) protein, 4) vitamins, 5) riboflavin, 6) niacin, 7) calcium, 8) iron, 9) copper, 10) zinc (5). N.sativa as a medicinal plant have beneficial effects on improve lipid profile and glycemic index, which has been confirmed in several studies (6-19). On the other hand regular physical activity is an important part of weight loss programs (like diet control, use of medication or injection of insulin) which can cause more glucose uptake by active muscles (20). Lack of participation in physical activity and regular exercise can involve in increase insulin resistance either directly or through weight gain. Therefore, with increasing physical activity, it is possible that metabolic syndrome related with insulin resistance in diabetic patients improve. It seems that in order to maximize reduce risk of diabetes in people with high risk, such as obese people, those with positive family history of diabetes and impaired glucose need for an appropriate level of physical activity and sport (1). Physical activity can lead to an increase in glucose uptake by active muscles. Glucose uptake in skeletal muscle during rest occurs with insulin stimulation which is impaired in diabetic patient, whereas muscle contraction through multiple distinct mechanisms and insulin sensitivity stimulates glucose transport (3). In fact, moderate intensity aerobic endurance exercise temporarily improve insulin and glucose uptake by skeletal muscle. Physical activity can respond skeletal muscle to insulin by increasing the expression or activity of proteins involved in metabolism and insulin signaling, so that aerobic endurance activities will increase the activity of glycogen synthase and increases the expression of GLUT4 transporters (2). According to these issues the goal of therapy in diabetic patients is to reach and maintain blood glucose, lipids and blood pressure to optimal levels (to prevent or delay chronic complications associated with diabetes and to improve the quality of life in these patients) in this regard, improve nutritional patterns and physical activity is recommended for diabetic people. So the aim of this study was to measure the effects of N.sativa extract and endurance training on improve glycemic indexes of diabetic rats.

Materials and Methods

In this experimental study, 50 adult male sprague dawley rats has been bought from animal breeding center of Islamic Azad University, Fars Science and Research Branch and kept in animal house of sport physiology for passes the eight-day period of adaptation (room temperature 22 ± 2 °C and 12 hours cycle of light and darkness). During this period of the time animals were free to access food and water. Then after induction of diabetes by 60 mg/kg streptozotocin (manufactured by Sigma), 32 diabetic rats with blood glucose more than 300 mg/dl selected as sample. The experimental intervention including training program and the consumption of N.sativa, started one week

after induction of diabetes. The study sample divided into four groups of 8 rats including (1) endurance training, (2) N.sativa, (3) endurance training with N.sativa and (4) control. Groups 1 and 3 performed endurance trainings four weeks and five sessions per week. Also groups 2 and 3 receive 100 mg/kg N.sativa extract intraperitoneal four weeks. At the end of four weeks after 12 hours overnight fasting, blood samples gathered. All ethical and legal aspects of this study reviewed and approved by Islamic Azad University, Fars Science and Research Branch. Method of N.sativa extraction was performed based on civilization et al study (2014) (21). For performed endurance training protocol, at first for familiar rats with endurance training, the animals ran on treadmill with speed of 8 meters per minute with zero slop for 10 minutes. At the end of the treadmill, a very weak electric shock was done to continue running forward. For prevent possible injury by electric shock, animals were conditioned by relatively poor sound or by touching their tail. Endurance training protocol consist four weeks running on treadmill without inclination (slope of zero), five sessions per week and 60 minutes per session with speed of 8 to 16 meters per minute. To warm the animals in training sessions, after placing the animals on the treadmill, the animals ran for 10 minutes at a speed of 8 meters per minute, and then training program was implemented. At the end of training sessions, in order to cool down the device, speed completely reverse to reduce the speed to zero. This program lasted about 5-7 minutes.

For statistical analysis of data used Kolmogorov–Smirnov test, one way ANOVA and tukey post hoc tests. *P*-value ≤0.05 was considered as significant.

Results

Glycemic indexes of all groups reported in table 1. The results of one way ANOVA test showed that there is significant difference in levels of fasting glucose (F=36.41, p-value: 0.001), Hb_{A1C} (F=17.07, *P*-value<0.001) and insulin resistance (F=8.00, *P*-value<0.001) of study groups, nevertheless there is no significant difference in insulin levels of study groups (F=0.34, *P*=0.79). The results of tukey post hoc test showed that four weeks endurance training, nigella sativa consumption and nigella sativa consumption with endurance training have significant effect on lowering fasting glucose, insulin resistance and Hb_{A1C} of diabetic rats (*P*-value<0.005).

Discussion

Chronic hyperglycemia associated with stress causes oxidative disorders such as nephropathy, neuropathy and cardiovascular diseases (22-23). Since the certain cure of diabetes is not possible, but diabetes can be controlled (24). The results of present study showed that four weeks N.sativa consumption caused hypoglycemic effects (significant reduction in fasting glucose, insulin resistance, and HbA1c) on streptozotocin induced diabetic rats. From N.sativa volatile oil substances such as thymol, thymoquinone and dithymoquinone obtained, which cause the main pharmacological effects of N.sativa. Anti-inflammatory, antioxidant and anti-histamine properties of N.sativa oil has various pharmacological effects such as reduction of inflammatory cytokines, reducing glucose, lipid and blood pressure (9). N.sativa also has anti-inflammatory and antioxidant properties

Table 1. Levels of fasting glucose, insulin, insulin resistance and HbA1c of research groups

Variable / Groups	Fasting glucose Mean ± SD	Insulin Mean ± SD	Insulin resistance (HOMA-IR) Mean ± SD	HbA1c Mean ± SD
Control	398.37±16.28	6.64±1.03	6.53±1.11	9.49±.86
Endurance training	285.87±22.78*	6.50±1.01	4.59±0.80*	6.70±1.04*
N.sativa	304.50±37.68 *	6.63±0.80	4.96±0.74 ¥	7.61±0.80*
N.sativa with endurance training	290.87±15.97*	6.96±0.90	5.00±0.73 ♀	6.93±0.71*

A significant difference with the control group *P*=0.001
¥ A significant difference with the control group *P*=0.006
♀ A significant difference with the control group *P*=0.007

can improve insulin secretion (to maintain the pancreatic beta cells) and reduced insulin resistance which results in control of blood glucose and diabetes (9). In order to results of this study, anti-diabetic and hypoglycemic effects of N.sativa so can use it for control of diabetes (8-19). It should be noted that the hypoglycemic effects of N.sativa seems to be dose-dependent such that significant effects were seen at higher doses (6). Many studies have confirmed the results of present study and are favorable with this study. For example, two weeks daily consumption of 100 mg/kg N.sativa decrease the glucose levels, LDL, VLDL and TG of alloxan induced diabetic rabbits (7); adding 5% N.sativa oil to the diets of streptozotocin induced diabetic rats resulted a significant reduction in glucose levels, LDL, VLDL and TG (8). A review article showed that consumption of N.sativa can lead to improve insulin resistance, blood glucose, HbA1c, hepatic gluconeogenesis, intestinal absorption and increase insulin production (9). Daily consumption of 2-3grams N.sativa for 12 weeks, significantly reduced FBS, HbA1c and insulin resistance and significantly increased pancreatic beta cell function and insulin production in diabetic patients, however, 1gram N.sativa consumption had no significant effect on noted variables (3); Regard to findings of reported studies hypoglycemic effects of N.sativa are depend on primary levels, dose and time of consumption. In order to confirm these findings 30 days daily consumption of 50 mg/kg Thymoquinone significantly reduced serum glucose, HbA1c and hepatic gluconeogenesis(11), 400 mg/kg N.sativa extract consumption significantly reduced hepatic glucose and serum glucose (6); daily consumption of 2 mg/kg N.sativa for 6 weeks improved glucose tolerance, inhibition of intestinal glucose absorption and weight loss (12); 4 months consumption of N.sativa increased phosphorylation of AMP-activated protein kinase (AMPK) and the content of GLUT4 in rats (13); one year consumption of N.sativa significantly reduced fasting glucose and HbA1c of 56 type 2 diabetic patients (14), 12 weeks daily consumption of 3grams N.sativa oil (in three separate servings) significantly decreased fasting glucose and HbA1c of 30- 60 years old type 2 diabetic patients (15); 5 mg/kg N.sativa extract consumption for 32 days significantly reduced fasting glucose in streptozotocin induced diabetic rats (16); 6 weeks consumption of 100, 200 and 400 mg/dl N.sativa significantly reduced fasting glucose in diabetic rats (17); daily intake of 2.5 ml N.sativa oil in two meals for 8 weeks significantly reduced FBS and HbA1c of healthy men with cholesterol levels between 200 to 300 mg/dl (18); 2 months consumption of 500 mg N.sativa capsules significant decreased HbA1c, FBS in men and women with metabolic syndrome (19); and mixed powder of N.sativa plant with standard food of rats (6.25%) for 2 months significantly reduced serum glucose levels of diabetic rats (5). Regarding the hypoglycemic effect of N.sativa it can be say that N.sativa with different ways can lead to improved insulin resistance and reduced serum glucose. N.sativa by regulation of liver enzymes regulates glucose metabolism and thereby reduces hepatic gluconeogenesis (9). For example, hexokinase activity increases in the liver. It also inhibits the glucose-6-phosphatase activity and fructose1 6 phosphatase which involved in gluconeogenesis. Moreover, N.sativa increases glucose-6-phosphate dehydrogenase enzyme activity in the pentose phosphate pathway inside the cell. N.sativa enzyme activates adenosine monophosphate protein kinase (AMPK) as well. Activation of AMPK inhibits gluconeogenesis and reduces hepatic glucose production. AMPK inside muscles increase synthesis and translocation translation of GLUT4 and the subsequent increased glucose uptake by muscle (9). According to mentioned studies N.sativa by increasing AMPK and GLUT4, improve FBS levels of streptozotocin induced diabetic rats.

An adaptive response to exercise in insulin resistance is improvement of glucose tolerance and insulin sensitivity by glucose transport in

skeletal muscle. This improvement (improvement in insulin action) caused by physical exercise and up regulation of specific components of glucose transport system in muscles resistant to insulin and it includes insulin receptor substrate and over expression of GLUT4 (26). Therefore, exercise can be one way of appropriate treatment in diabetic patients. The results of present study showed that 4 weeks, 5 sessions per week for 60 minutes running on treadmill with speed of 8-16 meters per minute caused hypoglycemic effects (significantly reduced fasting glucose, insulin resistance, and HbA1c) in streptozotocin induced diabetic rats. Most studies confirmed hypoglycemic effects of exercise in diabetic patients and diabetic rats (1,3,27-29). For example, it was reported that 6 months physical activity significantly reduced insulin resistance and fasting glucose levels in type 2 diabetic patients (27); 60 minutes running on treadmill with speed of 20 meters per minute, 5 sessions per week for 4 weeks significantly reduced fasting glucose, insulin, insulin resistance and HbA1c in diabetic rats (28); 45 minutes of exercise at 85- 90, 90- 95 and 95- 100 percent of an aerobic threshold in water and land for three times, significantly reduced fasting glucose of type 2 diabetic patients (29); 6 weeks, 5 sessions per week running on treadmill for 60 minutes per session has hypoglycemic effects in diabetic rats (3) as well as 6 weeks endurance training has hypoglycemic effects in sprague dawley diabetic rats (1). However, few studies have failed to show the effect of exercise on improvement of glycemic control in diabetic patients (30-33). For example 11.8 km running on treadmill for 12 weeks hadn't significant effect on fasting glucose and insulin levels of diabetic rats (30), 60 minutes pedaling on ergometer bicycle at 60% Vo_{2max}, 3 sessions per week for 12 weeks hadn't significant effect on serum glucose level and insulin of type 2 diabetic patients (31); 45 minutes walking, 3 sessions per week during 4 months, did not changes serum insulin levels, glucose and HbA1c of elderly type 2 diabetic patients (32) also 7 days aerobic exercise with 60 to 70 percent of HRR did not reduce fasting glucose and insulin of elderly men with impaired glucose tolerance (33). From possible mechanisms of endurance training which in present study induced reduction in fasting glucose levels and insulin, it can be noted that physical trainings through increasing the activity of glycogen synthase, increased oxidative and glycolytic enzyme activity and GLUT4 lead to improved insulin action in skeletal muscle. Also increase in activity of adenosine mono-phosphate kinase exercise induced leads to an increase in glucose transports (GLUT4s) in muscle cells so reduced circulation glucose levels with increasing glucose uptake by muscular cells (2).

Conclusions

In addition to these findings, results of present study showed that four weeks endurance training with N.sativa extract has hypoglycemic effects in diabetic rats but they have no hypoglycemic interaction effects. Since endurance training and N.sativa extract with the same mechanism (effect on GLUT4) lead to reduce glucose levels, likely endurance training with low dose of N.sativa don't have interaction effects on glycemic indexes. Regard to results of present study, four weeks 100 mg/kg N.sativa consumption, endurance training and endurance training with N.sativa have hypoglycemic effects in diabetic rats but their combination did not have hypoglycemic interaction effects.

References

1. Zar A, Hoseini A, Ahmadi F, Rezaei M. Effects of Ginger with Swimming Training on Blood Fat Profiles in Adult Diabetic Rats with Streptozotocin. Iranian J Nutr Scie Food Tech 2016;11:65-74.
2. Hosseini SA, Nikbakht H, Azarbayjani M.A. The Effect of Aqua Extract of Saffron with Resistance

Training on Glycemic Indexes of Streptozotocin Induced Diabetic Rats. Yasuj Med Sci Univ J 2014;18:284-94.
3. Shojae T, Hosseini SA, Hosseini SA. Review the Effect of Endurance Training on Glycemic Indexes of Streptozotocin Induced Diabetic Rats. Jahesh J 2013;15:39-46.
4. Gloria Y, David M, Ted J, Russell S. Systematic Review of Herbs and Dietary Supplements for Glycemic Control in Diabetes. Diabetes Care 2003;26:1277-1294.
5. Roghani M, Baluchnejadmojarad T, Roghani-Dehkordi F. Survey the Hypoglycemic and Hypolipidemic Effect of Chronic Oral Administration of Nigella Sativa in Diabetic Rat. Guilan Med Sci J 2007;63:26-31.
6. Bamosa AO, Kaatabi H, Lebdaa FM, Elq AM, Al-Sultanb A. Effect of Nigella Sativa Seeds on the Glycemic Control of Patients with Type 2 Diabetes Mellitus. Indian J Physiol Pharmacol 2010;54:344-54.
7. Ikram F, Hussain F. Antidiabeti Efficacy of Nigella Sativa L in Alloxan- induced Diabetic Rabbits. JMJM 2014;13:12-7.
8. Al-Logmani AS, Zari TA. Effects of Nigella Sativa L and Cinnamomum Zeylanicum Blume Oils on some Physiological Parameters in Streptozotocin-induced Diabetic Rats. Bol Latinoam Caribe Plant Med Aromat 2009;8:86-96.
9. Hadi S, Mirmiran P, Hadi V, Hosseinpour-Niazi S, Emamat S M. Effects of Nigella Sativa on Cardiovascular Risk Factors. Mazandaran Med Sci Uni J 2016;134:430-42.
10. Fararh K, Atoji Y, Shimizu Y, Shiina T, Nikami H, Takewaki T. Mechanisms of the Hypoglycaemic and Immunopotentiating Effects of Nigella Sativa L Oil in Streptozotocin Induced Diabetic Hamsters. Res Veterinary Sci 2004;77:123-9.
11. Fararh KM, Shimizu Y, Shiina T, Nikami H, Ghanem MM, Takewaki T. Thymoquinone Reduces Hepatic Glucose Production in Diabetic Hamsters. Res Vet Sci 2005;79:219-23.
12. Meddah B, Ducroc R, El Abbes Faouzi M, Eto B, Mahraoui L, Benhaddou-Andaloussi A. Nigella Sativa Inhibits Intestinal Glucose Absorption and Improves Glucose Tolerance in Rats. J Ethnopharmacol 2009;121:419-24.
13. Acevedo D, Varela E, Guerra J, Bau J, Reyna S. Nigella Sativa Influences GLUT4 through the AMPK Pathway. FASEB 2015;29:608-27.
14. Kaatabi H, Bamosa AO, Badar A, Al-Elq A, Abou-Hozaifa B, Lebda F, et al. Nigella Sativa Improves Glycemic Control and Ameliorates Oxidative Stress in Patients with Type 2 Diabetes Mellitus: Placebo Controlled Participant Blinded Clinical Trial. PLoS One 2015;10:23-31.
15. Heshmatia J, Namazib N, Memarzadehc M, Taghizadehc M, Kolahdoozd F. Nigella Sativa Oil Affects Glucose Metabolism and Lipid Concentrations in Patients with Type 2 Diabetes: A Randomized, Double-blind, Placebo-controlled Trial. Food Res Int J 2015;70:87-93.
16. Alimohammadi S, Hobbenaghi R, Javanbakht J, Kheradmand D, Mortezaee R, Tavakoli M, et al. Protective and Antidiabetic Effects of Extract from Nigella Sativa on Blood Glucose Concentrations Against Streptozotocin (STZ)-induced Diabetic in Rats: an Experimental Study with Histopathological Evaluation. Diagn Pathol 2013;8:137-42.
17. Abbasnezhad AA, Niyazmand S, Mahmoud Abadi M, Soukhtanloo M, Rezaee SA, Mousavi SM. Comparison the Effect of Hydroalcholic Extract of Nigella Sativa L Seed and Metformin on Blood Biochemical Parameters in Streptozotocin-Induced Diabetic Rats. Horizon Med Sci J 2015;20:243-8.
18. Amini M, Fallah Huseini H, Mohtashami R, Sadeqhi Z, Ghamarchehre M. Hypolipidemic Effects of Nigella Sativa L Seeds Oil in Healthy Volunteers: a Randomized, Double-Blind, Placebo-Controlled Clinical Trial. JMP 2011;40:133-8.
19. Najmi A, Nasiruddin M, Alikhan R, Haque S. Therapeutic Effect of Nigella Sativa in Patients of Poor Glycemic Control. Asian J Pharm Clinical Res 2012;5:224-28.
20. Nelson KM, Reiber G, Boyko EJ. Diet and Exercise among Adults with Type 2 Diabetes: Findings from NHANES III. Diabetes Care 2002;25:1722-8.
21. Tamadonfard Z, Sepehrara L, Jovhary H. The Effect of Nigella Sativa Extract on Learning and Spatial Memory of Adult Male Rats. JJUMS 2014;12:27-36.
22. Mahmoodi M, Sayyadi A, HosseiniZijoud S, Hajizadeh M, Kazemi Arababadi M, Asadikaram G. Survey on the Effects of Different Concentrations of Citrullus Colocynthis Fruit Powder on Some of the Blood Biochemical Factors in Normal and Diabetic Male Rats. Rafsanjan Med Sci Uni J 2012;11:11-20.
23. Roghani M, Aghaie M. The Effect Allium Ampeloprasum Feeding on Serum Level of Glucose, Triglyceride, and Total Cholesterol of Diabetic Rats. Koomesh 2007;8:73-8.
24. Roghani M, Baluchnejadmojarad T, Vaez-Mahdavi M, Vasei M. The Effect of Long-term Oral Administration of Nigella Sativum on the Contractile Reactivity of Thoracic Aorta in Diabetic Rats. Koomesh 2006;7:153-8.
25. Tadibi V, Bayat Z. Effect of Eight Weeks Aerobic Training and Drug Intervention on Quality of Life in Women with Type 2 Diabetes. Gorgan Med Sci Uni J 2012; 14:30-6.
26. Naylor LH, Davis EA, Kalic RJ. Exercise Training Improves Vascular Function in Adolescents with Type 2 Diabetes. Physio Rep 2016;4:12713-9.
27. Rowan CP, Riddell MC, Gledhill N, Jamnik VK. Community-Based Culturally Preferred Physical

Activity Intervention Targeting Populations at High Risk for Type 2 Diabetes: Results and Implications. Can J Diabetes 2016;2671:30031-4.
28. Heo M, Kim E. Effects of Endurance Training on Lipid Metabolism and Glycosylated Hemoglobin Levels in Streptozotocin-induced Type 2 Diabetic Rats on a High-fat Diet. Phys Ther Sci J 2013;25:989-92.
29. Delevatti RS, Pinho CD, Kanitz AC, Alberton CL, Marson EC, Bregagnol LP, et al. Glycemic Reductions Following Water- and Land- Based Exercise in Patients with Type 2 Diabetes Mellitus. Complement Ther Clin Pract 2016;24:73-7.
30. Sennott J, Morrissey J, Standley PR, Broderick TL. Treadmill Exercise Training Fails to Reverse Defects in Glucose, Insulin and Muscle GLUT4 Content in the db/db Mouse Model of Diabetes. Pathophysiology 2008;15:173-9.
31. Poirier P, Tremblay A, Broderick T, Catellier C, Tancrède G, Nadeau A. Impact of Moderate Aerobic Exercise Training on Insulin Sensitivity in Type 2 Diabetic Men Treated with Oral Hypoglycemic Agents: Is Insulin Sensitivity Enhanced Only in Non Obese Subjects?. Med Sci Monit 2002;8:59-65.
32. Fritz T, Wandell P, Aberg H, Engfelds P. Walking for Exercise- Dose Three Times Per Week Influence Risk Factor in Type 2 Diabetes?. Diabetes Res Clin Pract 2006;71:21-7.
33. Bloem CJ, Chang AM. Short Term Exercise Improves Beta-Cell Function and Insulin Resistance in Older People with Impaired Glucose Tolerance. J Clin Endocrinol Metab 2008;93:387-92.