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Comparison of aniseeds and coriander seeds for antidiabetic, hypolipidemic and antioxidant activities

Anason ve Kişnişin Antidiyabetik, Hipolidemik ve Antioksidan Etkinliklerinin Karşılaştırılması

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ABSTRACT

AIM: In the present study, it was aimed to investigate and to compare the antidiabetic, hypolipidemic and antioxidant activities of aniseeds (*Pimpinella anisum*) and coriander (*Coriandrum sativum*) seeds in type 2 diabetics as aniseeds and coriander seeds are rich in antioxidants and beneficial phytochemicals.
METHODS: The antidiabetic, hypolipidemic and antioxidant activities of aniseeds and coriander seeds were assessed *in vivo* by the administration of aniseed and coriander seed powder (5g/day) respectively to the selected two groups of type 2 diabetes patients for 60 days followed by the estimation of a no. of biochemical parameters viz. fasting glucose, lipid profile, enzymatic and non-enzymatic antioxidants.
RESULTS: Hyperglycemia, hyperlipidemia and oxidative stress as shown by increased lipid peroxidation, protein oxidation and increased activity of catalase (CAT) in erythrocytes, decreased serum β carotene, vitamin A, E and C observed in diabetics were countered by aniseeds and coriander seeds in the respective treated groups. In addition, decreased activities of erythrocyte antioxidant enzyme i.e. glutathione-S-tranferase (GST) and reduced glutathione (GSH) content were significantly improved in the treated-diabetics.
CONCLUSION: Both the seeds significantly influenced almost all the parameters without any detrimental effects by virtue of a number of phytochemicals, vitamins and minerals present in the seeds having therapeutic effects. The antidiabetic, hypolipidemic and antioxidant activities exhibited by the seeds are a result of the synergistic action of the bioactive compounds present in the seeds.
Key words: Type 2 diabetes, Oxidative stress, Lipid peroxidation, Protein oxidation, Antioxidant enzymes, Aniseeds, Coriander seed.

ÖZET

AMAÇ: Bu çalışmada, anason ve kişniş çekirdekleri antioksidanlarca zengin faydalı fitokimyasallar olduğundan, tip 2 diyabetiklerde anason (*Pimpinella anisum*) ve kişniş (*Coriandrum sativum*) çekirdeklerinin antidiyabetik, hipolipidemik ve antioksidan etkinliklerini araştırmak ve karşılaştırmak amaçlanmıştır.
YÖNTEM: Anason ve kişniş çekirdeklerinin antidiyabetik, hipolipidemik ve antioksidan etkinlikleri; glukoz perhizi, lipit profili, enzimatik ve non-enzimatik antioksidanlar olan biyokimyasal parametrelerin ölçümünü takiben 60 gün boyunca, tip 2 diyabetik hastalardan seçilmiş iki gruba sırasıyla anason ve kişniş çekirdeği tozunun (5 g/gün) uygulanmasıyla in vivo olarak değerlendirildi.
BULGULAR: Artmış lipit peroksidasyonu, protein oksidasyonu ve eritrositlerdeki artmış katalaz (KAT) aktivitesi, diyabetiklerdeki azalmış serum β karaten, vitamin A, E ve C sayesinde gösterilen Hiperglisemi, hiperlipidemi ve oksidatif stres, sırasıyla tedavi gruplarında anason ve kişniş çekirdekleri açısından karşılaştırıldı. Ayrıca, glutatyon-S-transferaz (GST) ve indirgenmiş glutatyon (GSH) içeriği gibi eritrosit antioksidan enzimlerinin azalmış aktiviteleri, tedavi edilmiş diyabetiklerde anlamlı olarak düzelmişti.
SONUÇ: Her iki çekirdek, tedavi edici etkilere sahip olan çekirdeklerde bulunan birçok fitokimyasallar, vitaminler ve minerallerin etkisi sayesinde, herhangi bir kötüleştirici etkileri olmadan, hemen hemen tüm parametreleri anlamlı olarak etkiledi. Çekirdekler tarafından sergilenen antidiyabetik, hipolipidemik ve antioksidan etkinlikler, çekirdeklerin içinde bulunan bioaktif bileşiklerin sinerjistik aktivitelerinin bir sonucudur.
Anahtar Kelimeler: Tip 2 diyabet, Oksidatif stres, Lipit peroksidasyonu, Protein oksidasyonu, Antioksidan enzimler, Anoson, Kişniş tohumu

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INTRODUCTION

Oxidative stress is defined as excess formation and or insufficient removal of highly reactive molecules such as reactive oxygen species (ROS) and reactive nitrogen species (RNS). Not only these are highly reactive chemical species important in ageing process, but they are also, either directly or indirectly, involved in various clinical disorders such as diabetes, atherosclerosis, reperfusion injury, cancer, etc. (1). Increased oxidative damage can result not only from more oxidative stress, but also from failure to repair or replace damaged biomolecules. Oxidative stress can result from decrease in antioxidant enzymes, depletions of dietary antioxidants and other essential constituents viz. copper, iron, zinc and magnesium (2).

Diabetes mellitus (DM) is a widespread disease with a great social impact and the human population worldwide appears to be in the midst of an epidemic of diabetes, a syndrome characterized by abnormal insulin secretion, derangement in carbohydrate and lipid metabolism, diagnosed by the presence of hyperglycemia (3). Several mechanisms may cause oxidative insult in DM (4). The increased oxidative stress as measured by indices of lipid peroxidation and protein oxidation has been shown to be increased in both insulin dependent DM (IDDM), and non-insulin dependent (NIDDM) (5). Increased oxidized low density lipoprotein (LDL) or susceptibility to oxidation has also been shown in DM (6). Type 2 diabetes patients had decreased erythrocyte-Glutathione (GSH) and increased Glutathione Disulfide (GSSG) levels (7). Advanced glycation or glycosylation end products (AGEs), the products of glycation and oxidation (glycoxidation) are increased with age, and at an accelerated rate in DM (8).

Hyperglycemia induces the polyol pathway, resulting in induction of aldose reductase and production of sorbitol (9). LDL and RBC membranes isolated from type 1 and type 2 DM patients were much more susceptible to oxidation than LDL from normal subjects (10). Protein carbonyl content is the most widely used marker of oxidative modification of proteins and suggested to be reliable marker of oxidative stress (11). Elevated carbonyl levels were detected both in type 1 and type 2 diabetes and also in experimental diabetes (5). In addition to lipid and protein oxidation, oxidative damage of DNA has been reported in diabetes patients. These changes might contribute to atherogenesis in DM and to the microangiopathic complications of the disease (12).

The introduction of insulin and later oral hypoglycemic agents, revolutionized the management of DM but none of them were unequivocally successful in maintaining normal blood glucose levels and in avoiding complications (13). Diabetics represent a population in whom oxidative stress is much higher than in the general population (14). To combat the ever-increasing oxidative stress, antioxidants may be particularly important as they diminish cumulative oxidative damage (15). Natural dietary agents such as fruits, vegetables and spices have drawn a great deal of attention from both the scientific community and the general public, owing to their putative ability to suppress oxidative stress (16). Hence, the present study was undertaken to assess and to compare the antidiabetic, hypolipidemic and antioxidant activities of aniseeds (*Pimpinella anisum*) and coriander (*Coriandrum sativum*) seeds in type 2 diabetics.

MATERIALS AND METHODS

Procurement of seeds and preparation of powder

Aniseeds (*Pimpinella anisum*) and coriander (*Coriandrum sativum*) seeds were procured from the S.P Stores, Anantapur, Andhra Pradesh, India weekly once, thoroughly cleaned to free from extraneous matter, finely powdered using electric blender, placed in air tight containers and then packed in polythene covers and used for the clinical trial.

Selection of subjects and treatment

Both male and female type 2 diabetes (non insulin dependent) patients in the age group of 40-60yrs. with no other specific complications were selected from K.M Diabetes Hospital, Anantapur, Andhra Pradesh, India on the basis of a specific questionnaire. Out of the selected subjects, 20 served as Control, 20 served as Experimental I and 20 served as Experimental II. The Experimental group I received aniseed powder and the Experimental group II received coriander-seed powder (5g per day) in 2 equal doses for a period of 60 days. Written consent was obtained from the subjects, dietary guide lines were given and all the patients were under the supervision of a diabetologist. This study was approved by the Institutional Ethical Committee.

Clinical analyses

At the initial and final stages of the experiment, fasting blood was drawn for the assay of various parameters. Fasting blood glucose (17), lipid peroxidation in plasma (18) and erythrocytes (19), protein oxidation (20), vitamin A and β-carotene (21), vit. C (22) and vit.E (23) in serum were estimated.

Activities of catalase (CAT) (24), glutathione-s-transferase (GST) (25) and reduced glutathione (GSH) (26,27) were assayed in erythrocytes.

Statistical analysis

Mean, standard deviation and paired samples-t test were conducted to assess significant difference between the data obtained before and after treatment and coefficient of variation was examined to assess significant difference between the two samples (28).

RESULTS

Fasting blood glucose

Fasting blood glucose levels in control (untreated), Experimental I treated with aniseed powder and Experimental II treated with coriander seed powder for 60 days were presented in Table 1 which indicated 11% ($p<0.001$) rise in control and 36% ($p<0.001$) decrease in aniseed-treated and 13% ($p<0.001$) decrease in coriander-treated type 2 diabetics.

Table 1. Fasting blood glucose and serum lipid profile in control and experimental diabetics (mean ± SEM).

Groups	Fasting blood glucose (mg/dl)	Cholesterol (mg/dl)	Triglyceride (mg/dl)
Control			
Initial	185.23 ± 0.7	238.03 ± 0.7	214.01 ± 0.7
Final	206.04 ±1.0*** (11)	239.04 ± 0.4	217.12 ± 0.8
Exp-I			
Initial	190.53 ± 0.8	238.11 ± 0.7	222.36 ± 0.8
Final	122.16 ± 0.9*** (36)	220.12 ± 0.6*** (8)	189.21 ± 0.6*** (15)
Exp-II			
Initial	138.81 ± 0.8	222.02 ± 0.8	214.54 ± 0.7
Final	120.80± 0.1*** (13)	189.03 ± 0.6** (15)	149.63 ± 0.7*** (30)

Exp-I Aniseed-treated group; **Exp-II** Coriander seed-treated group
The figures in parentheses indicate per cent increase /decrease over respective initial data
Comparison between initial and final: ** $p<0.01$; *** $p<0.001$

Serum cholesterol and triglycerides

Significant decrease in serum cholesterol and triglycerides i.e. 8%, 15% ($p<0.001$) in aniseed-treated and 15% ($p<0.01$), 30% ($p<0.001$)in coriander seed-treated type 2 diabetics respectively was observed at final stages of the experiment (Table 1). Table 2 shows serum lipoproteins in control, Exp I (aniseed-treated) and Exp II (coriander seed-treated) at the initial and final stages of the experiment. No much change was observed with respect to VLDL, LDL and HDL-C in control group where as significant decrease of 11% ($p<0.01$) in VLDL-C and 7% ($p<0.001$) in LDL-C levels were observed in Exp I while 31% ($p<0.001$) and 9% ($p<0.001$) decrease was noticed respectively in Exp II. Both the groups exhibited significant rise in HDL-C i.e. 34% ($p<0.001$) and 42% ($p<0.001$) respectively. With respect to VLDL-C, LDL-C and HDL-C, treatment with coriander was identified to be better compared to that of aniseeds while aniseed treatment.

Table 2. Serum lipoproteins in control and experimental diabetics (mean ± SEM).

Groups	VLDL (mg/dl)	LDL (mg/dl)	HDL (mg/dl)
Control			
Initial	43.16 ± 0.1	165.0 ± 0.5	27.6 ± 0.8
Final	43.21 ± 0.1 (1)	169.6 ± 1.1* (2)	27.3 ± 0.9 (2)
Exp-I			
Initial	44.62 ± 0.3	168.1± 0.6	26.1 ± 0.8
Final	40.11± 0.9** (11)	157.3±0.6*** (7)	35.1 ± 0.4*** (34)
Exp-II			
Initial	42.87 ± 0.2	185.0 ± 0.3	21.2 ± 0.8
Final	29.43± 0.24*** (31)	168.2 ± 0.9*** (9)	30.1 ± 0.4*** (42)

Exp-I Aniseed-treated group; **Exp-II** Coriander seed-treated group
The figures in parentheses indicate per cent increase /decrease over respective initial data
Comparison between initial and final: *$p<0.05$; ** $p<0.01$;*** $p<0.001$

Table 3. Lipid peroxidation and protein oxidation in control and experimental diabetics (mean ± SEM).

Groups	Protein oxidation	Lipid peroxidation Erythrocytes (nmol MDA/gHb)	Lipid peroxidation Plasma (nmol MDA/dl)
Control			
Initial	0.0051 ± 0.8	5.12 ± 0.6	419.81 ± 0.2
Final	0.0054 ± 1.0** (6)	10.09 ± 0.9*** (97)	420.32 ± 3.0
Exp-I			
Initial	0.0068 ± 0.4	10.83 ± 0.5	434.11 ± 0.7
Final	0.0042 ± 3.0*** (38)	5.32 ± 0.5*** (51)	341.22 ± 1.0*** (21)
Exp-II			
Initial	0.0060 ± 0.2	9.84 ± 0.9	419.01 ± 0.8
Final	0.0030 ± 1.2*** (50)	5.02 ± 0.5*** (49)	317.33 ± 7.0*** (24)

Exp-I Aniseed-treated group; **Exp-II** Coriander seed-treated group
The figures in parentheses indicate per cent increase /decrease over respective initial data.
Comparison between initial and final: ** $p<0.01$; *** $p<0.001$

Lipid peroxidation and protein oxidation

Table 3 depicts protein oxidation in serum, lipid peroxidation in erythrocytes and plasma in control, aniseed-treated and coriander seed-treated diabetics at initial and final stages of the experiment. Treatment with aniseeds decreased protein oxidation (38%) in

serum and lipid peroxidation in erythrocytes (51%, p<0.001) and plasma (21%, p<0.001) and treatment with coriander seeds also decreased protein oxidation (50%),lipid peroxidation in erythrocytes (49%, p<0.001) and plasma (24%, p<0.001) in type 2 diabetics as compared with the initial values. Contrary to this, a 97% (p<0.001) increase in erythrocyte lipid peroxidation was seen in the control group.

Serum vitamin antioxidants

Table 4 shows serum non enzymatic antioxidants (β-carotene, vitamin A, C, E)levels in control, aniseed and coriander seed-treated type 2 diabetes patients. β-carotene levels in the serum were improved by 36% and 46% (p<0.01) in aniseed and coriander seed-treated groups respectively. A slight rise was seen in the serum vitamin C levels in aniseed (9%, p<0.01) and (24%, p<0.01) in coriander-treated type 2 diabetics. The data presented in Table 4 also indicates increase of 15 % (p<0.01) and 22% (p<0.01) in serum vitamin E in aniseed and coriander seed-treated groups respectively.

Table 4. Serum non enzymatic antioxidants in control and experimental diabetics (mean ± SEM).

Groups	β carotene (µg/dl)	Vitamin A (µg/dl)	Vitamin C (µg/dl)	Vitamin E (mg/dl)
Control				
Initial	166.2 ± 0.8	23.5 ± 0.8	1.93 ± 0.6	3.12 ± 0.2
Final	141.6 ± 1.0 (15)	21.8 ± 1.0 (7)	1.88 ± 0.9 (2)	3.10 ± 1.0
Exp-I				
Initial	116.7 ± 0.4	21.7 ± 0.4	2.01 ± 0.5	3.45 ± 0.7
Final	158.3 ± 3.0** (36)	27.5± 3.0** (27)	2.20± 0.5** (9)	3.96 ± 1.0** (15)
Exp-II				
Initial	108.3 ± 0.2	20.6 ± 0.2	1.35 ± 0.9	2.98± 0.8
Final	158.5 ± 1.2** (46)	28.5 ± 1.2** (38)	1.67 ± 0.5** (24)	3.64± 0.7** (22)

Exp-I Aniseed-treated group; **Exp-II** Coriander seed-treated group
The figures in parentheses indicate per cent increase /decrease over respective initial data
Comparison between initial and final: ** p<0.01

Erythrocyte antioxidant enzymes

Table 5 shows a significant decrease i.e. 65% (p<0.001) and 59% (p<0.001) in the activity of catalase in aniseed and coriander seed-treated type 2 diabetics respectively. The data also indicates a significant increase in the GST levels (119%, 65%, p<0.001) in aniseed and coriander seed treated type 2 diabetics where as there was a decrease of 22% in the control group. Treatment with aniseeds and coriander seeds resulted in significant increase in erythrocyte glutathione i.e. 128% (p<0.001) and 278 % (p<0.001) respectively in the Exp I and Exp II diabetics.

Table 5. Serum enzymatic antioxidants and GSH in control and experimental diabetics (mean ± SEM).

Groups	Catalase (K/gHb)	Glutathione-s-transferase (µmol of CDNB-GSH conjugate/mgHb	Reduced Glutathione (IU/gHb)
Control			
Initial	5.26 ± 0.7	24.47 ± 0.6	25.75 ± 0.3
Final	9.74± 0.4*** (85)	23.05 ± 1.3 (6)	24.23 ± 0.9 (6)
Exp-I			
Initial	9.91 ± 0.4	21.65 ± 0.6	13.50 ± 0.8
Final	3.49 ± 0.2*** (65)	47.5 ± 2.6*** (119)	30.81 ± 0.4*** (128)
Exp-II			
Initial	9.98 ± 0.4	26.15 ± 0.9	7.88 ± 0.1
Final	4.12 ± 1.16*** (59)	43.12 ± 1.2*** (65)	29.79 ± 0.1*** (278)

Exp-I Aniseed-treated group; **Exp-II** Coriander seed-treated group
The figures in parentheses indicate per cent increase /decrease over respective initial data
Comparison between initial and final: *** p<0.001

DISCUSSION

Hyperglycemia is the main risk factor for developing diabetic complications and also accelerates atherosclerosis (29). It is postulated that mitochondrial glucose overload results in increased electron transfer to oxygen and formation of free oxygen radicals. This in turn activates the pathways leading to diabetic complications along with hyperglycemia (30). It is also demonstrated that acute glucose fluctuations induce oxidative stress and these fluctuations were suggested to be valuable predictors for risk of diabetic complications (31). In the present study, significant decrease in fasting blood glucose in aniseed and coriander groups indicate control over hyperglycemia and decreased oxidative stress in both the groups. However, the effect was more than double in aniseed-treated group. This is supported by significantly decreased lipid peroxidation (a marker of oxidative stress in erythrocytes and plasma) in aniseed and coriander-treated groups (Table 3).

In the present study, decreased glucose levels indicate control over oxidative stress as hyperglycemia can directly cause increased generation of reactive oxygen species as glucose undergoes autooxidation and generate OH radicals (32). In addition, glucose reacts with proteins in a non enzymatic manner leading to the development of Amadori products followed by formation of AGEs. ROS is generated at multiple steps during the process. In hyperglycemia, there is enhanced metabolism of glucose through polyol (sorbitol) pathway, which also results in enhanced production of $^{\bullet}O_2^-$ (33).

The compounds responsible for the activity of aniseeds are caffeic-acid, camphene, chlorogenic-acid, rutin, scopoletin, squalene, stigmasterol, micronutrients -ascorbic acid, minerals magnesium, manganese, copper etc. while in coriander seeds-the phytochemicals like apigenin, caffeic -acid, myristic acid, myristin, p-hydroxy-benzoic acid, palmitic acid, protocatechuic acid, isoquericitrin, gamma-terpinene, terpinen-4-ol, terpinolene, trans-anethole.

The blood sugar lowering effect exhibited by coriander seeds is attributed to the phytochemicals viz. chlorogenic acid, pectin, protocatechuic acid and rutin, the micronutrients-ascorbic acid, niacin (vitamins) and minerals-chromium, copper, magnesium and zinc present in coriander seeds. Besides, the antioxidants in coriander seeds i.e. apigenin, β-carotene, caffeic acid, camphene, gamma-terpinene, isoquercitrin, myristic acid, myristin, terpernin 4 ol, terpenoline and trans anethole etc., also contribute to the observed effect (34).

Hypercholesterolaemia occurs in diabetes with about the same frequency as in the general population, but it confers a greater risk of CHD in diabetes. Hypertriglyceridemia, hypercholesterolaemia and low serum high density lipoprotein (HDL) cholesterol levels are more common in diabetes, particularly in non-insulin-dependent diabetes. The hypertriglyceridaemia is thus exacerbated by poor diabetes control (35). Therefore, in the present study, a significant decrease in cholesterol and triglycerides could be due to hypolipidemic action of the seeds under investigation.

Type 2 diabetes and insulin resistance are commonly associated with elevated triglycerides, low HDL-C levels, dense LDL and VLDL-C (36). Low density lipoproteins (LDL) represent a major risk factor in atherogenesis after undergoing oxidative modifications. LDL oxidation is mediated by free radicals or other oxidants including extra cellular reactive oxygen species, thiols, hypochlorous aicd, metal ions, aldehydes and lipoxygenases (37). The oxidation process induces several structural and compositional modifications in the lipid of LDL (hydroperoxides, lysophosptidylcholines, oxysterols, hydroxy nonenal), and structural alterations of apo B (38). In the present study, significant decrease in LDL in both the treated groups support the antidiabetic and hypolipidemic principles present in both the samples. Hyperglycemia increases the production of free radicals which cause LDL-C oxidation. Oxidised LDL-C causes deleterious effects in the vascular walls. Besides, hyperglycemia causes glycation of LDL and glycated LDL tends to accumulate leading to atherogenesis (36). In the present study, the samples under consideration by controlling hyper glycemia would have controlled ROS generation there by decreased LDL-C oxidation. Tremendous rise in HDL observed in the study, also would have protected LDL-C from oxidation as anti atherogenic properties of HDL-C are largely related to their inhibitory effect against LDL-C oxidation. Another antioxidant mechanism of HDL may result from the ability to extract lipid peroxidation products from oxidized lipoprotein cellular membrane using oxidized erythrocytes as a cellular model. The anti atherogenic property of HDL is due to protection against the deleterious effect of ox-LDL and inhibitory effect against LDL oxidation. The antioxidant properties of HDL are also attributed in part, to the ability to chelate transition metals. HDL modulates cell proliferation and migration, and is strongly related to its cytoprotective property against apoptosis induced by ox-LDL and other pro-atherogenic agents (39).

Insulin resistance directly stimulates VLDL production (40). The elevated VLDL is a reflection of increased total triglyceride in NIDDM. Treatment with aniseeds and coriander decreased VLDL in the respective groups and is further supported by the clearance or decreased production of VLDL in the treated subjects. Generally, newly synthesized cholesterol may be used for VLDL secretion. One possibility of decrease in VLDL is that more of newly synthesized cholesterol is channeled for the synthesis of bile acids as reported by (41) in coriander-treated animals.

Lipid peroxidation appears to be highly significant consequences of oxidative stress in injured human arterial walls contributing to the development of the atherosclerotic lesions (42). Especially, polyunsaturated fatty acids (PUFAs) are highly susceptible to reactions with free radicals. Peroxidation of lipids in fatty acids may lead to a radical chain reaction. Because of these chain reactions, one substrate radical ($R^{\bullet}$) may result in the formation of many equivalents of lipid peroxides (LOOH). Diabetic red blood cells (RBCs) were shown to be more susceptible to lipid peroxidation as measured by TBARS in humans (43). Similarly, increased plasma peroxide concentrations were reported in type 2 diabetics (44). In the present study, decreased lipid peroxidation in both the treated groups in erythrocyte and plasma is attributed to the phytochemicals, present in aniseeds and coriander seeds. Most of these compounds act as radical scavengers, some of them reduce the radicals by donating hydrogen atoms, some of them also act as chain breaking agents in lipid peroxidation as a result of which a significant decrease in lipid peroxidation

was observed both in erythrocytes and plasma in treated subjects. Besides, the antioxidant property of both the seeds was further evidenced by significantly decreased serum protein oxidation (Table 3) in the treated diabetics.

Proteins are an important target for oxidative challenge. Reactive oxygen species modify amino acid side chains of proteins to form protein carbonyls and protein carbonyl content is the most widely used marker of oxidative modification of proteins and suggests to be a reliable marker of oxidative stress (11). Elevated protein carbonyl stress was detected both in type1 and type2 and also in experimental diabetes. Furthermore, the protein carbonyl content is well correlated with the complications of diabetes (5). A significant decrease in the protein oxidation in the present study, along with significant decrease in lipid peroxidation confirms the control over oxidative stress in the treated patients.

Both the groups showed rise in serum β-carotene and vitamin A levels which could have resulted in a significant decrease in lipid peroxidation (Table 4) in RBC and plasma in both the treated groups as carotenoids interact with free radicals that initiate harmful reactions such as lipid peroxidation. Carotenoids present in the samples with their highly reactive conjugated bonds act as free radical traps or antioxidants (45). Especially β-carotene, a precursor of vitamin A, a nutritional antioxidant is known to protect membrane lipids from peroxidative damage. Its antioxidant ability is attributed mainly to the scavenging of several biologically damaging free radicals or reactive oxygen species such as singlet oxygen, peroxyl radical, superoxide and nitrogen dioxide (46). The increase in free radical production with subsequent damage to the cellular processes observed in type 2 diabetes could be overcome with the supplementation of β-carotene as β-carotene traps free radicals and functions as an antioxidant (47). Hence, raised serum β-carotene and vitamin A levels in both the treated groups would have caused decrease in the lipid peroxidation in the treated groups.

Ascorbic acid is a naturally occurring major antioxidant, essential for the scavenging of toxic free radicals, both in the plasma and tissues (48). Previous studies suggest that oxidative stress is increased in diabetic patients and diabetic animal models (49). The ascorbic acid levels in plasma and tissues of diabetic patients and animals have been reported to be low (50). Supplementation of ascorbic acid decreased sorbital levels (51) and increased the stability of blood vessels (52). Therefore, decreased ascorbic acid levels in diabetes injure the blood vessels and lead to the development of diabetic complications. In the present study, raised ascorbic acid levels in the treated diabetics indicate decreased oxidative stress as ascorbic acid is a scavenger of the toxic free radicals. This increase in vitamin C could also be due to the protection of the existing vitamin C from oxidation to dehydroascorbic acid by some of the antioxidant phytochemicals present in the seeds under the investigation.

Vitamin E, the major lipid-soluble antioxidant present in all cellular membranes protects against lipid peroxidation (53) and is thought to be an important chain breaking antioxidant which plays an important role in various stages of carcinogenesis through its contribution to immuno-competence, membrane and DNA repair and decreasing oxidative DNA damage (54). Both vitamin C and E quench free radicals by providing hydrogen atoms ($H^{\bullet}$) i.e. reducing equivalents that can pair up with unpaired electrons on free radicals. In this process, vitamin E itself gets oxidized. The reduced forms of the vitamins are regenerated by reduction with GSH, ascorbate, NADH/NADPH. vit. C and GSH can regenerate or spare each other so that vitamin C may spare vit. E (55). Therefore, in the present study, increased vitamin C (the quencher of free radicals) could prevent the lipid peroxidation (Table 3) in erythrocyte and plasma in type 2 diabetics.

Catalase is a large enzyme, containing heme bound iron at its active sites, and is a major primary antioxidant defence component that primarily works to catalyze the decomposition of H_2O_2 to H_2O and shares this function with glutathione peroxidise (56). In the present study, a significant increase in the activity of catalase, shows increased rate of radical production before treatment in the groups. Treatment with aniseed and coriander seed powder indicated a significant decrease in the activity of catalase, which indicates reduced H_2O_2 production as a result of the treatment given in the groups. This can be evidenced by the decreased lipid peroxidation (Table 3) in erythrocytes after the treatment in the subjects of both the groups.

GST is a multi-functional protein found in many tissues, plays an important role in the detoxification of xenobiotics thereby protecting the cell form peroxidative damage especially in the liver and also in lungs, tissues and erythrocytes (57). Therefore, a significant increase in the levels of GST indicated protection against oxidative stress in the experimental group. This is further evidenced by remarkable increase in the lipid peroxidation in RBC and plasma (Table 3) in controls who didn't receive any treatment and significantly decreased lipid peroxidation in aniseed and coriander seed-treated groups.

Glutathione is a cellular non-enzymatic antioxidant, thus provides the cell with multiple defences not only against ROS but also against their toxic products (58). Marked alterations in antioxidant enzyme activities and tissue GSH concentration were reported in diabetes. Decreased GSH in diabetes may be caused by different pathways including 1) the increase in sorbitol synthesis causing NADPH depletion and deficiency of this limits the reduction of GSSG to GSH catalyzed by glutathione reductase, 2) decreased activity of HMP shunt enzymes which generate NADPH 3) transport of GSSG through erythrocyte membranes due to oxidative stress induced membrane damage (59). In poorly controlled diabetes, impaired glutathione system by inactivation of GPx and GR may contribute to the initiation and/or progression of diabetic complications (60). Hence, significantly increased GSH in both the treated groups in the present investigation indicates control over oxidative stress. This is further supported by significantly decreased lipid peroxidation (Table 3) in erythrocytes and plasma of the treated subjects.

CONCLUSIONS

Both aniseeds and coriander seeds decreased blood sugar but not brought down below normal, so the seeds are antihyperglycemic. The seeds also decreased serum lipids, lipoproteins and improved HDL as a result of the hypolipidemic activity. Besides, aniseeds and coriander seeds controlled lipid peroxidation, a marker of oxidative stress, improved non enzymatic and enzymatic antioxidants in type 2 diabetics and proved to be antioxidative. With respect to controlling lipid peroxidation, both are equally efficient, while with respect to decreasing fasting glucose, decreasing the activity of catalase, improving the activity of GST, aniseeds are superior to coriander, while in decreasing lipids and lipoproteins, protein oxidation, improving GSH, coriander ranks first. However, both the samples significantly influenced almost all the parameters by virtue of a number of phytochemicals, vitamins and minerals present in the seeds having both medicinal and therapeutic effects. The antidiabetic, hypolipidemic and antioxidant activities exhibited by the seeds are a result of the synergistic action of the bioactive compounds present in the seeds.

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