



Cardio-protective and anti-cancer therapeutic potential of *Nigella sativa*

Hammad Shafiq¹, Asif Ahmad^{1,2*}, Tariq Masud¹, Muhammad Kaleem¹

¹ Department of Food Technology, PMAS Arid Agriculture University Rawalpindi, 43600 Pakistan

² Corn, Soybean and Wheat Quality Labs., 205 Williams Hall, 1680 Madison Avenue, Wooster, OH 44691 USA

ARTICLE INFO	ABSTRACT
<p>Article type: Review article</p> <hr/> <p>Article history: Received: May 17, 2013 Accepted: Oct 10, 2014</p> <hr/> <p>Keywords: Anti-diabetic Cancer Cardiovascular <i>Nigella sativa</i></p>	<p><i>Nigella sativa</i> is the miraculous plant having a lot of nutritional and medicinal benefits, and attracts large number of nutrition and pharmacological researchers. <i>N. sativa</i> seed composition shows that it is the blessing of nature and it contains and many bioactive compounds like thymoquinone, α-hederin, alkaloids, flavonoids, antioxidants, fatty acids many other compounds that have positive effects on curing of different diseases. Several medicinal properties of <i>N. sativa</i> like its anti-cancer, anti-inflammatory, anti-diabetic, antioxidant activities and many others are well acknowledged. However, this article focuses on activity of <i>N. sativa</i> against cardiovascular diseases and cancer. For gathering required data the authors went through vast number of articles using search engines like Science direct, ELSEVIER, Pub Med, Willey on Line Library and Google scholar and the findings were classified on the basis of relevance of the topic and were reviewed in the article. <i>N. sativa</i> is rich source of different biologically active compounds and is found effective in controlling number of cardiovascular diseases and various cancers both <i>in vivo</i> and <i>in vitro</i> studies.</p>
<p>► Please cite this paper as: Shafiq H, Ahmad A, Masud T, Kaleem M. Cardio-protective and anti-cancer therapeutic potential of miraculous herb <i>N. sativa</i>. Iran J Basic Med Sci. 2014; 17:967-979.</p>	

Introduction

Various herbal plants are being used for treatment of various ailments since ancient time. In the modern world, herbal plants are still receiving considerable attention as indicated by the annual growth of the herbal plants based industry in developed countries that is growing at rate of 7-15 % annually (1). In the developing countries, large segment of population (about 80%) uses herbal medicines for curing different basic medical problems (2). The reason behind their wide spread use is that they are considered as effective, safe, reliable, non-toxic, easily available and affordable as compared to modern allopathic medicines (3). There is a growing trend of modern research in this era to explore the therapeutic potential and medicinal uses of different plant species, *Nigella sativa* is among one of such plants. It is also regarded as miracle herb with rich religious and historical back ground (4).

N. sativa is a small shrub and annual flowering plant which belongs to the family Ranunculaceae. It bears white, yellow, pink and purplish delicate flowers containing 5 to 10 petals (5). The fruit is an inflated large capsule which bears large number of black seeds when ripped, the seeds are known as black seeds or black cumin in English, Habbat el Baraka or Habbah Sawda in Arabic (6). They are

commonly called as kalonji in Pakistan and India (7). *N. sativa* plant is commonly grown in Middle Eastern and Western Asian countries including Syria, Lebanon, Pakistan, India and Afghanistan (8).

N. sativa oil and seeds have been widely used to management of different diseases within centuries and regarded as important drug in traditional medical system in Asian and Middle East countries (Ayurveda, Unani, Arabic and Chinese medicines) (10, 11) and also recommended for regular use in Tibb-e-Nabwi (12).

This article is an effort to gather the published work of researchers against the cardiovascular disease (CVD) and anti-cancer activity with reference to *N. Sativa* and its active components, so to enhance our awareness about the potential health benefits of this miracle herb and to further arouse the interest of researchers to explore its health benefit and discover new novel drugs from *N. sativa* for treatment of these two notorious diseases. As these two ailments are the leading causes of deaths in low, as well as high income countries. CVDs are the chief cause of deaths worldwide (13). Low and middle income countries populations are most affected from cardiovascular diseases. It is estimated that over 80% of the CVD deaths occur in developing countries (14) and deaths due to CVD may touch a figure of

*Corresponding author: Asif Ahmad. Corn, Soybean and Wheat Quality Labs, 205 Williams Hall, 1680 Madison Avenue, Wooster, OH 44691 USA. Tel: +1 330-439-9793; email: ahmad.179@osu.edu

23.3 million in next 15 years (15). Cancer is the second major cause of the deaths worldwide and account for about 8.2 million deaths in 2012 (16) that may rise up to 22 million in next 20 years (17).

Materials and Methods

The needed information was gathered using systematic literature searches on Science direct, ELSVIER, Pub Med, Willey online Library and Google scholar. Key words used for searching of data include *N. sativa*, *N. sativa* cardio-protective effect, *N. sativa* anti-diabetic activity, *N. sativa* anti-cancer activity and thymoquinone. About 400 relevant research and review articles were studied during the year 2013-2014. The literature used as reference is collected from last two decades but majority of paper used as reference are from 2000-2014.

The results of findings were interpreted and categorized based on the relevance to topic. First of all the traditional use of *N. sativa* is presented followed by the different studies that describes the effect of *N. sativa* on different health complications including cardiovascular, anti-diabetic and anti-cancer activities. Structure of different bioactive compounds is drawn and possible mechanism of action against different diseases is presented in this manuscript.

Traditional uses

N. sativa seeds and it's oil, were traditionally used from centuries to cure various ailments in different parts of world particularly Asia, Middle East and Africa (18, 19). *N. sativa* holds special importance to Muslims as Prophet Muhammad once stated that "*N. sativa* can heal every disease except death" (20). *N. sativa* seeds are commonly used as spice and food preservative and are considered to have abortifacient, anodyne, anthelmintic, appetizing, carminative, deodorant, diaphoretic, digestive, diuretic, emmenagogue, expectorant, febrifuge, galactagogue and purgative effects (1, 21). In folk medicines it is used to treat conditions and diseases like asthma, bronchitis, cough,

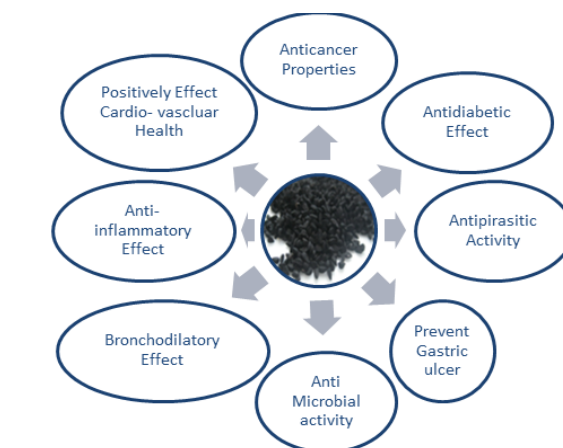


Figure 1. Health Benefits of the *Nigella sativa*

diarrhea, dysentery, dyspepsia, fever, flatulence, jaundice, paralysis, piles and other disorders related to cardio-vascular, digestive, immune, liver, respiratory and kidney systems (4, 22, 23). Its seed are rich source of many biologically active components that are known to have anti-inflammatory, anti-fungal, hypoglycemic, anti-hypertensive and anti-histaminic effects (24-27). A tincture made from *N. sativa* seed is used to cure worms and skin eruptions (4). Due to its so many beneficial health effects it is called as "Habbatul barakah" in Arabic meaning the seed of blessing (28). Some of the health benefits of *N. sativa* are depicted in Figure 1.

Chemical composition of *N. sativa*

N. sativa chemical composition is very diverse and consists of range of different components including carbohydrates, proteins, fats, oils, fibers, vitamins, minerals (Cu, Fe, P and Zn etc.), alkaloids, saponins and many other biologically active compounds (29). Major alkaloids identified in *N. sativa* are: pyrozol alkaloid (e.g. nigellicine and nigellidine) and isoquinoline alkaloids (e.g. nigellicimine and nigellicimine-N-oxide) as depicted in Figure 2 (30).

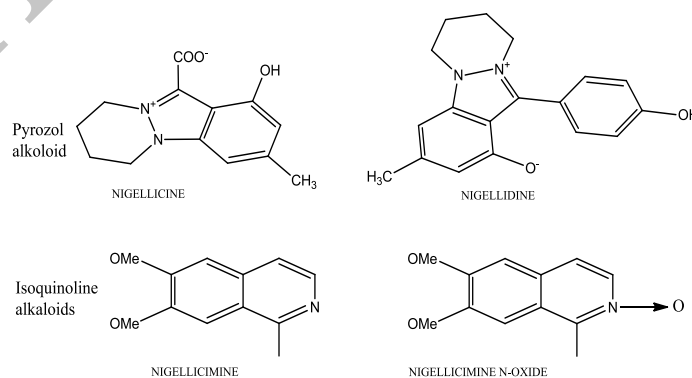


Figure 2. Alkaloids in *Nigella sativa*

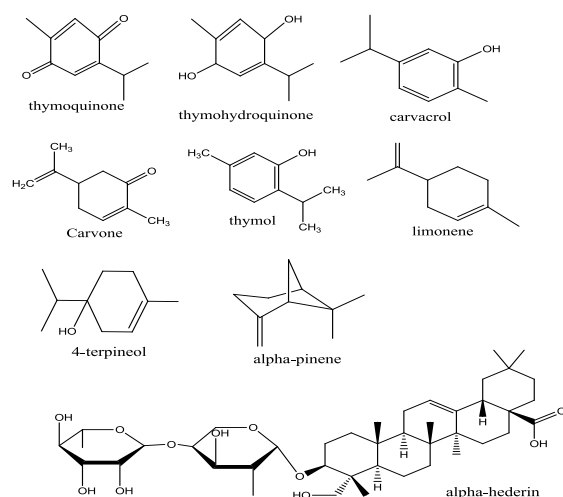


Figure 3. Biologically active compounds found in *Nigella sativa*

Range of biologically active compounds (Figure 3) found in *N. sativa* that includes: thymoquinone (TQ), thymohydroquinone, dithymoquinone (DIM), 4-terpineol, carvacrol, carvone, t-anethol, α -pinene, thymol, α -hederin, limonene and many other compounds found in trace amount (10). Most of the pharmacological properties of *N. sativa* are result of its quinone constituents (30).

The seed is also rich in fatty acids including oleic acid, linolenic acid, linoleic acid, eicodadienoic acid, arachidic acid, palmitoleic acid, palmitic acid, stearic acid and myristic acid (31, 32).

Cardiovascular Health Benefits of *N. sativa*

Cardiovascular diseases (CVD) refer to groups of diseases related to heart and blood vessels. The reasons of cardiovascular diseases are very diverse. Hypertensions, atherosclerosis, LDL, high cholesterol level are among the major causes of CVD (33). CVD are increasing at very rapid pace worldwide. Although older people are more vulnerable to cardiovascular diseases but these may also occur at early stages of life (34). The cardiovascular health benefits of the *N. sativa* are discussed below.

Effect of *N. sativa* on lipid profile

Cholesterol rich diet, oxidative stress and hypercholesterolemia can lead to development of the atherosclerosis. It is CVD in which elasticity of artery walls is reduced and they become hardened, which can lead to heart attack. Elevated serum cholesterol, LDL and triglycerides level are major cause of this ailment (35). Studies have reported that *N. sativa* has favorable effects on the lipid profile and it significantly reduces serum cholesterol LDL and triglycerides levels (7, 36-38).

In a study, to investigate the effect of the thymoquinone (TQ) consumption on the serum lipid profile of the rabbits fed upon cholesterol rich diet, results showed that TQ consumption significantly

reduced total cholesterol, LDL, triglycerides and thiobarbituric acid-reactive substances concentrations, while increased HDL-cholesterol concentration (39). Similar effects were observed in other studies using *N. sativa* oil and powder (36-38, 40). *N. sativa* oil was observed to decrease serum cholesterol level by 15.5% and triglyceride level by 22 % in normal rats (41).

Promising results was also reported in human studies, it was estimated that a dose of 1 gm. *N. sativa* powder per day for a period of two months resulted in noteworthy decrease in LDL-cholesterol, triglycerides levels and increase in HDL-cholesterol level in hypercholesterolemic patients (42). Similar results were also reported in another study on the hypercholesterolemic patients in which *N. sativa* consumption was found to be associated with lowering of bad cholesterol level thus helpful in normalization of lipid profile in patients, preventing heart problems (43, 44)

Effects *N. sativa* on the lipid profile was thought to be the combined effect of various components, like TQ, sterols and flavonoids rather than any single component (25). Various mechanisms have been proposed to explain hypolipidemic effect of *N. Sativa*, including inhibition of new cholesterol synthesis or stimulation of bile acid secretion, both actions were known to reduce serum cholesterol levels (44, 45). Another reported mechanism based on antioxidant activity of *N. sativa* which prevent non-enzymatic lipid peroxidation (46).

Anti-diabetic effect of *N. sativa*

Diabetes mellitus (or simply diabetes) refers to group of chronic metabolic disease in which patient suffers from high blood sugar level. Untreated diabetes can lead to many serious complications particularly cardiovascular diseases and kidney failure (47). Diabetes can occur due to either pancreas were not making enough insulin (Type I diabetes mellitus) (48) or due to inability of cells to respond appropriately to insulin produced by the pancreas (Type II diabetes mellitus) (49). About 347 million people worldwide are suffering from diabetes; and the numbers are continuously increasing (14). Around 80% of the diabetic deaths occur in developing and low income countries (15). Many researchers have reported that *N. sativa* has anti-diabetic and hypoglycemic activity.

Oxidative stress is thought to play an important part in the pathogenesis of the diabetes mellitus, as oxidative stress can decreases the efficiency of pancreatic β cell and in turn effects the insulin production (50). *N. sativa* and its components are effective against diabetes as they decrease oxidative stress and thus preserve the pancreatic beta cell integrity (51). The protective effect of *N. sativa* seeds/oil on the pancreatic β cell was demonstrated in number of studies, in which it was reported that no ultra-structural changes in pancreatic cells occur

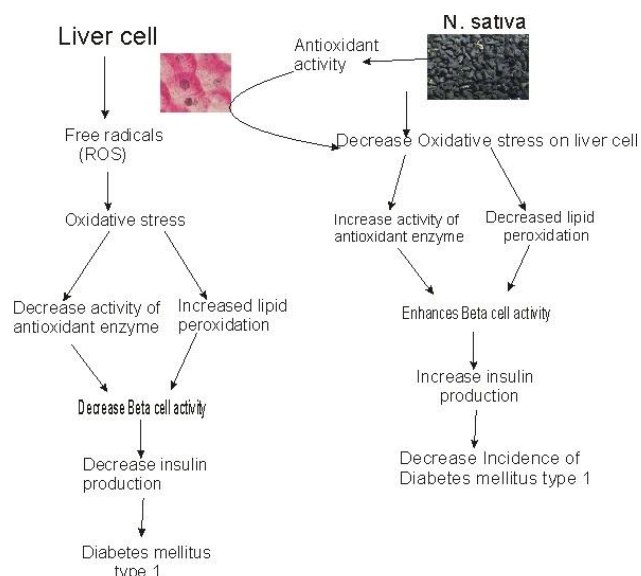


Figure 4. Mechanism of anti-diabetic effect of *N. sativa*

in STZ induced diabetic mice's which received *N. sativa* seed /oil treatment while Islet cell degeneration occur in mice's which do not received *N. sativa* seed/oil treatment (52-55). Antidiabetic mechanism of *N. sativa* is elucidated in Figure 4.

N. sativa oil and aqueous extract are equally good in controlling serum glucose and insulin response. Positive effect of the *N. sativa* extract/oil intake on the parameters like serum insulin, super dismutase (SOD), serum glucose and malondialdehyde (MDA) levels were demonstrated in number of studies (53, 56-59). TQ content of *N. sativa* were considered as the major constituent responsible antidiabetic activity of the herb (60-62). It was found that the effectiveness of *N. sativa* in diabetic patients is further increased if it is used along with α -lipoic acid and L-carnitine. The combination of all three component was shown to have a great impact on carbohydrate metabolism in STZ induced diabetic rats and their consumption was found to enhance insulin production and C-peptide level (63).

Effect of *N. sativa* on diabetes type II

Diabetes type II accounts for more than 90% of all the diabetes cases (49). A study planned to evaluate the potential effects of the TQ on the activities of the two main enzymes (i.e. hexokinase and glucose 6-phosphate dehydrogenase) involved in carbohydrates metabolism in rats in which diabetes was induced by injecting STZ-nicotinamid. The results showed that TQ consumption (at the rate of 80 mg/kg body weight) is effective against diabetic conditions and this amount also restore the activities of enzymes near to normal thus enhancing glucose consumption by the tissues and reducing the risk of type-II diabetes (64). To show the effect of *N. sativa* on the insulin resistance syndrome a study was planned with sixty patients. The patients were divided into two groups of thirty

patients each. Patients of the both groups were advised to take a fixed dose of statin and metformin tablets for six weeks period. The patients of second group were additionally given 2.5 ml of *N. sativa* oil two times a day during the therapy. The results showed more improvement in serum cholesterol and fasting blood glucose of the patients of group 2 as compared to group 1 thus indicating therapeutic potential of *N. sativa* against insulin resistance syndrome (62).

In a similar study *N. sativa* seeds were used as adjuvant therapy in patients suffering from type II diabetes mellitus along with their basic medication. The subjects were divided into 3 different dose groups and were given 1, 2 and 3 gm. *N. sativa* seeds / day for period of 3 months. Results showed that dose of 2 gm. per day resulted in noteworthy reduction in fasting blood glucose level, glycosylated hemoglobin (HbA1c) and cause increase in β -cells functionality. 1 gm. per day dose resulted in improvement in all parameters but that was not statistically significant while dose of 3 gm. per day doesn't cause any additional benefit as compared to 2 gm. per day dose. So the dose of 2 gm. *N. sativa* seed / day can be beneficial adjuvant therapy in type 2 diabetes mellitus (65).

Endothelial dysfunction

It is a pathological disorder of the endothelium that play role in the pathogenesis of numerous cardiovascular disorders and can result from hypercholesterolaemia, hypertension, obesity, septic shock, diabetes and smoking as well (78, 79). In a recent study, to determine the result of TQ on the age related endothelial dysfunction, it was found that TQ recovers endothelial function at least in part, through inhibition of oxidative stress and regulation of the angiotensin system (79).

Effect of *N. sativa* consumption on the blood pressure and heart rate

Different studies showed that *N. sativa* positively effects the elevated heart rate and blood pressure and its consumption is associated with lowering of elevated heart rate and blood pressure (80-84). Consumption of *N. sativa* was found effective in normalizing the elevated heart rate of the cadmium treated rats (81). While in another study *N. sativa* was found effective in reducing the diabetes-induced disturbances in heart rate of the rabbits in which diabetes was induced by injecting alloxan (84).

The human study designed to access the effect of *N. sativa* on the blood pressure of the patients suffering from mild hypertension, shows that *N. sativa* extract consumption favorably effects the elevated blood pressure (85). Similar results were found in another human study in which Qidwai, *et al* (86) reported that *N. sativa* seeds have favorable effect on the high blood pressure.

Table 1. Effect of *Nigella sativa* on lipid profile and its ant diabetic effects

Study objectives	Results	References
To evaluate the hypolipidemic effects of NS in menopausal women	Significant improvement in the lipid profile was observed in menopausal women	Ibrahim, et al (67)
To investigate the effect of the poly herbal formulation (NS sugar powder) on diabetes and serum lipid profile	Significant reduction in fasting blood glucose and increase in HDL and decrease in TG was observed in Wistar rats	Alam, et al (69)
To study the effect of NS on lipid profile of the hypercholesterolemic rabbits	NS positively effects all the lipid variables in rabbits	Asgary, et al (70)
To explore the effect of NS on hypolipidemic and hypocholesterolemic condition	Significant reduction in the TG, TC and LDL-c was observed in rats	Ali, et al (71)
To study the effect of NS on hypertriglyceridemia	Significant reduction in serum TG level was observed in human	Nasir, et al (72)
To study NSO treatment effect upon the regeneration of pancreatic islets of langerhans in diabetic rats	Positive effect on the regeneration of the islets of langerhans was observed in rats	Sobhi, et al (76)
To evaluate the repairing ability of the NSO on the damaged pancreatic tissue in diabetic rats	Positive effect was observed on the morphology of the Langerhans islets in diabetic rats	Hmza, et al (77)
To examine the anti-diabetic potential of the NS fixed oil and NS essential oil	Reduction in MDA level and antioxidants damage and modulation of lipid profile was observed in rat	Sultan, et al (66)
To evaluate the anti-mutagenic effect of NS on the experimentally induced diabetes type - II	NS decreased the frequency of micronuclei in the erythrocytes of bone marrow and enhanced the antioxidant status in the treated diabetic Wistar rats	Sheikh, et al (68)
To evaluate the effect diet fortified with NS and Fenugreek on diabetic rats	Positive effects on the serum Glucose and lipid profile, thyroid hormones, kidney and liver functions was observed in rats	Mahmoud (73)
To study the effects of NSO and TQ on cardiovascular risk parameters in experimental hyperlipidemia	High antioxidant activity of NS and protective effect of NS was observed in rats	Shafeeqe (74)
To Study of the effects of NS hydroalcoholic extract on glucose concentrations in diabetic rats	NS extract cause significant reduction in FBG level and protect the great deal of the pancreatic islet cells of rats	Alimohammadi, et al (75)

MDA malondialdehyde, NS *N. sativa*, NSO *N. sativa* oil, HDL high density lipoprotein, TC total cholesterol, TG Triglyceride, LDL-c low density lipoprotein, FBG fasting blood glucose

Anti-cancer activity of the *N. sativa*

Cancer is a rapidly growing health problem and posing a serious challenge to health professionals and researchers. American cancer society has reported that more than 0.5 million deaths occur due to cancer during 2013 in USA alone and the numbers are really threatening in under-developed world (87). With the wide spread and devastating effect of cancer and its high economic load, there is need for identifying natural and cheap products with anti-carcinogenic activity. Many studies depicts that risk of cancer occurrence can be reduced by the consumption of many vegetables and fruits (88-90). *N. sativa* is one of them who showed promising anti-cancer activity in number of studies (10, 62, 91-94).

Ibn-Sina (428 Hijri) was aware of the antitumor effect of the *N. sativa* and was probably the first known physician who used *N. sativa* in the tumors treatment (10). The anti-cancer effect of *N. Sativa*, with regard to modern time was perhaps first revealed when improvement in activity of natural killer cells was detected in cancer patients who were

receiving multimodality immunotherapy program in which *N. sativa* was one of the element (95). Later on, number of studies was done by many researchers to study the anti-cancer effect of *N. Sativa* seed and its extracts using both *in vivo* and *in vitro* models. Anti-cancer activity of *N. sativa* against different types of cancers is discussed below.

Lung cancer

Lung cancer is the leading cause of cancer deaths worldwide. It caused 1.59 million deaths in 2012 i.e. about 20% of total cancer deaths (16), while American cancer society estimated that of all the cancer deaths in 2014 about 27% will occur due to lung cancer (87).

The diet supplemented with *N. sativa* and honey was shown to have protective effect against lung, colon and skin cancers (96). *N. sativa* seed extract showed cytotoxicity against number of cancer cell line including Lewis lung sarcoma (LL/2) (97). Kumara and Huat (98) demonstrated *N. sativa* extract exhibit anti-cancer activity against the LL/2 tumor cell line that are subcutaneously implanted in BDF1 mice. TQ,

extracted from the *N. sativa* seed, at concentration of 100 μ M showed significant anti-cancer activity against the lung cancer cell line and demonstrated to inhibit cancer cell proliferation by about 90% (99).

N. sativa oil showed significant inhibitory effect against the human lung cancer cell line A-549 with IC₅₀ value of 43 μ g/ml (100). Later on in a recent study, Al-Sheddi, *et al* (101) also reported that *N. Sativa* oil and seed extract significantly reduces human lung cancer cells viability and alter the cellular morphology of A-549 cells in a concentration dependent manner.

Breast cancer

Breast cancer is the most common cancer in the woman's and caused more than 0.5 million deaths in 2012 (16). It is the number one cause of the cancer deaths in women's of less developed countries while second major cause of the cancer deaths in woman's of developed countries (102).

In a study, the effect of the aqueous and alcoholic extracts of *N. Sativa* on MCF-7 (breast cancer cell line) was accessed and the results showed the *N. Sativa* extracts are effective in inactivating MCF-7 cells (103). In another study MCF-7 (breast cancer cell line) was exposed to the aqueous and alcoholic extracts of *N. Sativa* in combination with the H₂O₂ (oxidative stressor). Survivability of the MCF-7 cells was measured under different concentrations and combinations using standard cell culture technique and results showed that *N. sativa* extracts alone or in were combination with H₂O₂ are effective against MCF-7 cells and affect their survivability and can provide a promising treatment for breast cancer therapy (104).

To evaluate the protective potential of the melatonin, retinoic acid and *N. sativa* seeds against breast cancer 7,12-di-methylbenzene (α) anthracene (a well-known carcinogenic substance that induce mammary carcinoma in rats) was injected in rats. At the end of study period, serum and tissues of animals were evaluated for the factors like markers of tumorigenicity, endocrine derangement, apoptotic changes, and markers of oxidative stress. Then results showed that frequency of mammary carcinoma was quite low in rats (33%) which start receiving melatonin, retinoic acid and *N. sativa* treatment 14 days before the intake of DMBA as compared to rats which received treatment after the intake of DMBA (56%) while rats which do not received any treatment showed highest frequency (60%). The results showed that melatonin, retinoic acid and *N. sativa* are effective in reducing the carcinogenic effects of DMBA (105). Similar anti cancerous results were reported for TQ derivative against MCF-7 breast cancer cell line (106).

In a more recent study, the anti-cancer activity of the supercritical-CO₂ extract of *N. sativa* against MCF-7 breast cancer cell line was investigated and results showed that supercritical-CO₂ extract of *N. sativa* have

significant anti-proliferative activity against MCF-7 cells and can be used to treat breast cancer (107).

Colon cancer

Colorectal cancer caused more than 0.69 million deaths in 2012 (16) and it is second major cause of cancer related deaths in US (87). To evaluate the preventive effect of *N. Sativa* oil on colon cancer aberrant crypt foci were induced in Fischer rats using 1,2-dimethylhydrazine. The rats were divided into four groups, first group served as a control group, second group received oil at post-initiation stage, third group received oil at the initiation stage while fourth group received 0.9% saline and oil from beginning till end of study. At the end of study i.e. 16 weeks, rats were sacrificed and results showed that *N. sativa* oil treatment in the post-initiation stage (group two) significantly reduced the total number of aberrant crypt foci however, treatment at initiation stage (group three) did not produce significant inhibitory effect. So it was concluded from the study that *N. sativa* oil has the potential to prevent colon carcinogenesis in the post-initiation stage (108). Gali-Muhtasib, *et al* (109) studied the effect of TQ on the HCT-116 (colon cancer cells line) and found that TQ is effective against colon cancer cells and trigger apoptosis in colon cancer cells in time and dose dependent manner thus inhibits the growth of cancer cells. However, TQ was not found effective against the human colon carcinoma cell line HT-29 in a study by Rooney and Ryan (110).

Renal cancer

A study was designed to evaluate the therapeutic effect of the *N. sativa* against ferric nitrilotriacetate (Fe-NTA) induced renal carcinogenesis in Wistar rats. Fe-NTA induction caused number of changes in the normal metabolic processes of the kidney. A dose of 50 and 100 mg *N. sativa* crushed seeds / kg body weight showed significant restoration of normal metabolic processes thus preventing the cancerous effect of Fe-NTA (111) In a recent study the anti-cancer activity of the *N. sativa* seed hydro-alcoholic extract was evaluated against the human renal carcinoma cells and results shows that *N. sativa* seed extract significantly inhibit the growth of human renal carcinoma cells (112).

Anti-cancer activity of the *N. sativa* decoction

To evaluate the anti-carcinogenic potential of the decoction, composed of *N. sativa* seed, *S. glabra* rhizome and *H. indicus* root, hepato-carcinogenesis was induced in male Wistar rats using dimethyl nitrosamine. The rats were given decoction after the initiation of carcinogenesis for 10 weeks, results showed significant reduction in the number and area of dimethyl nitrosamine mediated glutathione S-transferase placental form positive foci, number of

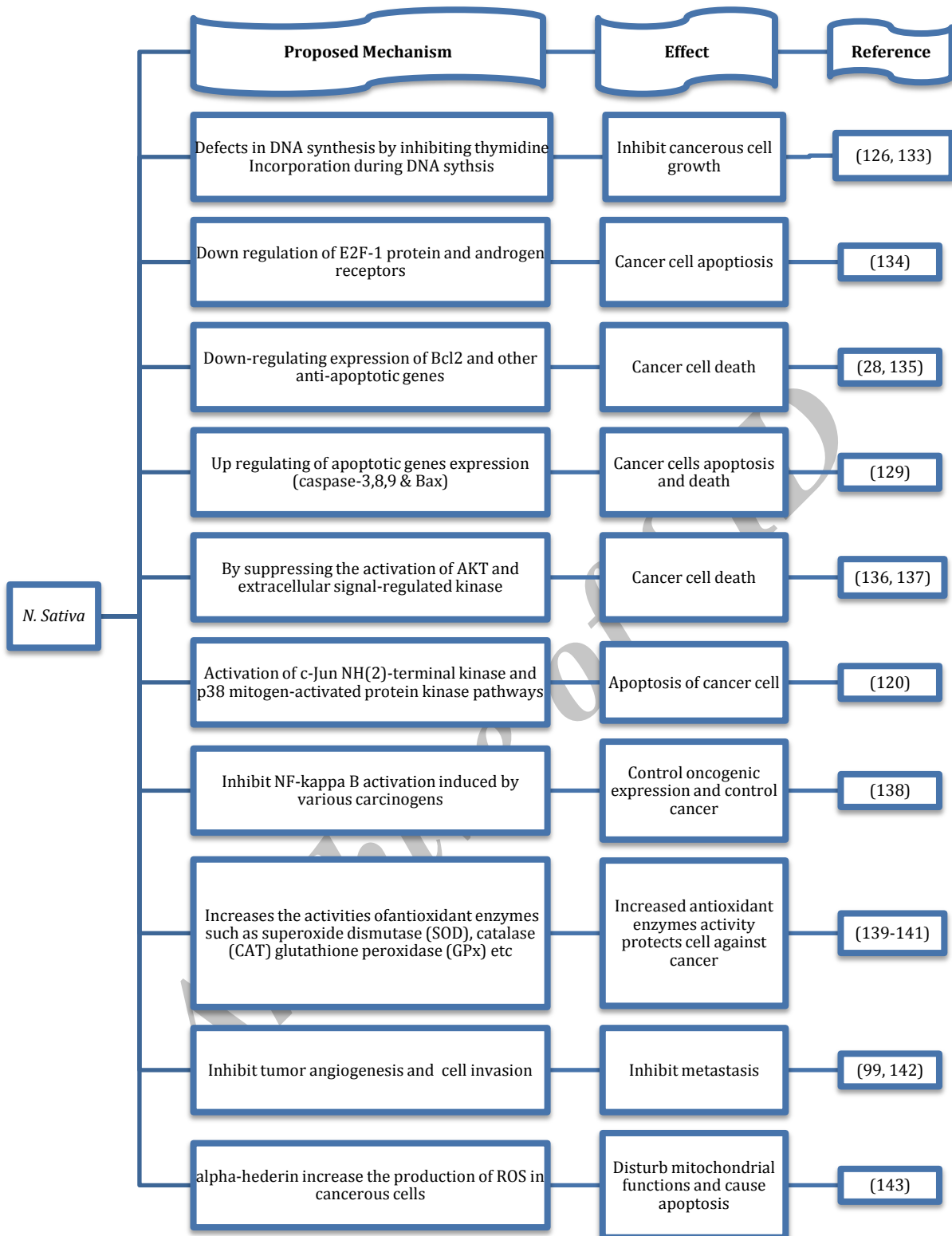


Figure 5. Different proposed mechanisms for anti-cancer activity of *N. sativa*

cells/cm² of foci and the staining intensity of foci in the liver as compared to controls (113). Later on, same decoction was shown to have the protective effect against dimethyl nitrosamine mediated carcinogenic changes in a study of 16 months (114). The anti-cancer effect of the same decoction was

evaluated against the human hepatoma (HepG2) cell line and it was found that decoction has strong dose dependent anti-cancer activity against HepG2 cancer cell line (115). Anti-carcinogenic effect of the same decoction was further evaluated by the Samarakoon, *et al* (116) who studied the anti-cancer effect of the

decoctions made by aqueous and ethanol extracts of the *N. sativa*, *S. glabra* and *H. indicus* against human hepatoma (HepG2) cell lines using MTT and SRB assays. Results depicted that both the extracts showed strong cytotoxicity to HepG2 cells. However, aqueous extract showed quite higher cytotoxic potential as compared to the ethanol extract.

Miscellaneous studies

Delay in the onset of papilloma and reduction in their number per mouse was observed, in mice in which skin cancer was induced by application of croton oil, by the topical application of *N. sativa* extract (117). Proliferation and apoptosis in pancreatic ductal adenocarcinoma (PDA) cells was observed by the administration of TQ (118). Anti-cancer effect of TQ against the pancreatic cancer was also demonstrated by other researchers (119, 120). Shafi, *et al* (121) studied the effect of *N. sativa* organic (Hexane, methanol and chloroform) extracts on the human epithelial cervical cancer (HeLa) cell line and found that all the three extracts induced apoptosis in HeLa cells and effectively killed them. Hasan, *et al* (122) found that *N. sativa* extract caused 88.3% inhibition in proliferation of the SiHa (human cervical cancer cells line) at a concentration of 125 µl/ml.

Thymoquinone (TQ) anti-cancer activity

Probably cytotoxic activity of the TQ was first reported against Ehrlich ascites carcinoma, Dalton's lymphoma ascites and Sarcoma-180 by Salomi, *et al* (123). Later on TQ and DIM, both extracted from *N. Sativa*, were reported to have cytotoxic activity against several human cancer cell lines, that were resistant to etoposide and doxorubicin anti-cancer drugs (124). Soon after that, TQ was found effective against benzo-(α)-pyrene induced forestomach carcinogenesis in rats whose consumption (0.01% in drinking water) had shown to reduce the onset and multiplication of the benzo-(α)-pyrene induced forestomach cancer (125). Likewise, antitumor potential of the TQ was investigated against the 20-methylcholanthrene induced fibrosarcoma in mice and results showed that consumption of TQ, a week in advance and after 20-methylcholanthrene treatment, can considerably delay the onset of fibrosarcoma tumor (126).

It was reported that TQ showed antitumor activity against the SW-626 colon cancer cell line which was equivalent to 5-fluorouracil a well-known drug used in treatment of colon cancer (127, 128). Anti-cancer activity of TQ was also observed against the human leukemia (HL-60) cells (129). Furthermore, TQ also showed significant antitumor activity against hepatocellular carcinoma (HepG2) cell line in a dose-dependent manner (130, 131). In another study, TQ is showed to inhibit growth of the panel of human colon cancer cells (Caco-2, DLD-1 HCT-116, HT-29 and

LoVo), without any cytotoxic effect on the normal human intestinal FHs74Int cells (132).

Possible mechanisms of *N. sativa* anti-cancer activity

Anti-cancer activity of any compound is due to two main reasons i.e. it either kills cancer cells or hinder any alteration in the genetic material of the normal cells. Several mechanisms have been proposed for the anti-cancer activity of the *N. sativa* that are summarized in Figure 5.

Conclusion

N. sativa is really a seed of blessing and proved to provide protection against the two most notorious ailments i.e. cancer and cardiovascular health problems. It is rich in different phytochemicals and nutritionally essential components. Health benefits of the *N. sativa* seed, oil and extracts have been shown in both *in vivo* and *in vitro* types of studies. In detail research of this herb and chemical modifications in the molecular structure of *N. sativa* active components can lead to the discovery of many novel medicines. *N. sativa* can also be used in combination with already recognized drugs. We hope that this article will increase the awareness and the interest of researchers to investigate the potential health benefits of *N. sativa*.

References

1. Paarakh PM. *Nigella sativa* Linn.-A comprehensive review. Indian J Natu Prod Res 2010; 1:409-429.
2. Grover J, Yadav S. Pharmacological actions and potential uses of *Momordica charantia*: a review. J Ethnopharmacol 2004; 93:123-132.
3. Mills S, Bone K. Principles and practice of phytotherapy. Modern herbal medicine: Churchill Livingstone; 2000.
4. Ahmad A, Husain A, Mujeeb M, Khan SA, Najmi AK, Siddique NA, *et al*. A review on therapeutic potential of *Nigella sativa*: A miracle herb. Asian Pacific J Trop Biomed 2013; 3:337-352.
5. Al-Khalaf MI, Ramadan KS. Antimicrobial and Anti-cancer Activity of *Nigella sativa* oil-A Review. Aus J Basic Appl Sci 2013;7:505-514.
6. Ismail MYM. Therapeutic Role of Prophetic Medicine Habbat El Baraka (*Nigella sativa* L.)-A Review. World Appl Sci J 2009;7:1203-1208.
7. Gilani A, Jabeen Q, Khan M. A review of medicinal uses and pharmacological activities of *Nigella sativa*. Pak J Biol Sci 2004;7:441-451.
8. Zohary D, Hopf M, Weiss E. Domestication of Plants in the Old World: The origin and spread of domesticated plants in Southwest Asia, Europe, and the Mediterranean Basin: Oxford University Press; 2012.
9. Sharma NK, Ahirwar D, Jhade D, Gupta S. Medicinal and pharmacological potential of *Nigella sativa*: A review. Ethnobot Rev 2009; 13:946-955.
10. Randhawa MA, Alghamdi MS. Anti-cancer activity of *Nigella sativa* (black seed)—A review. Am J Chin Med 2011; 39:1075-1091.

11. Nasir A, Siddiqui MY, Mohsin M. Therapeutic Uses of Shoneez (*Nigella sativa* Linn.) Mentioned in Unani System of Medicine-A Review. *Int J Pharm Phytopharmacol Res* 2014; 4: 47-49.
12. Al-Bukhari MI, Sahih Al-Bukhari. The Collection of Authentic Sayings of Prophet Mohammad (peace be upon him), Division 71 on Medicine. 2nd ed. Ankara, Turkey: Hilal Yayinlari; 1976.
13. Alwan A. Global status report on noncommunicable diseases 2010: World Health Organization; 2011.
14. WHO. Cardiovascular diseases 2013 [25/4/2014]. Available from: <http://www.who.int/diabetes/facts/en/>.
15. Mathers CD, Loncar D. Projections of global mortality and burden of disease from 2002 to 2030. *PLoS Med* 2006; 3:e442.
16. WHO. Cancer fact sheet 2013 [25/4/2014]. Available from: <http://www.who.int/mediacentre/factsheets/fs297/en/>.
17. Reuters T. Cancer cases worldwide likely to rise to 22 million new cases in 2 decades 2014 [29/4/2014]. Available from: [cancer-cases-worldwide-likely-to-rise-to-22-million-new-cases-in-2-decades-1.2521156](http://www.reuters.com/article/cancer-cases-worldwide-likely-to-rise-to-22-million-new-cases-in-2-decades-1.2521156).
18. Warriar P, Nambiar V, Ramankutty C. Indian medicinal plants: a compendium of 500 species: Orient Longman Ltd. Chennai. 1996; 3:38-90.
19. Yarnell E, Abascal K. *Nigella sativa*: holy herb of the middle East. *Altern Complement Ther* 2011; 17:99-105.
20. Ramadan MF. Nutritional value, functional properties and nutraceutical applications of black cumin (*Nigella sativa* L.): an overview. *Int J Food Sci Technol* 2007; 42:1208-1218.
21. Goreja WC. Black seed: nature's miracle remedy: TNC International Inc; 2003.
22. Padhye S, Banerjee S, Ahmad A, Mohammad R, Sarkar FH. From here to eternity-the secret of Pharaohs: Therapeutic potential of black cumin seeds and beyond. *Canc ther* 2008; 6:495-510.
23. Sharma P, Yelne M, Dennis T, Joshi A, Billore K. Database on medicinal plants used in Ayurveda. The University of Virginia; 2000. accessed from: <http://agris.fao.org/aos/records/US201300132829>.
24. Chakravarty N. Inhibition of histamine release from mast cells by nigellone. *Ann Allergy* 1993; 70:237-242.
25. Ali B, Blunden G. Pharmacological and toxicological properties of *Nigella sativa*. *Phytother Res* 2003; 17:299-305.
26. Khan M, Ashfaq M, Zuberi H, Mahmood M, Gilani A. The *in vivo* antifungal activity of the aqueous extract from *Nigella sativa* seeds. *Phytother Res* 2003; 17:183-186.
27. Al-Ghamdi M. The anti-inflammatory, analgesic and antipyretic activity of *Nigella sativa*. *J Ethnopharmacol* 2001; 76:45-48.
28. Khan MA, Chen HC, Tania M, Zhang DZ. Anti-cancer activities of *Nigella sativa* (black cumin). *Afr J Tradit Complement Altern Med* 2011; 8:226-232.
29. Nergiz C, Ötles S. Chemical composition of *Nigella sativa* L. seeds. *Food Chem* 1993; 48:259-261.
30. Khan MA. Chemical composition and medicinal properties of *Nigella sativa* Linn. *Inflammopharmacol* 1999; 7:15-35.
31. Nickavar B, Mojab F, Javidnia K, Amoli MR. Chemical composition of the fixed and volatile oils of *Nigella sativa* L. from Iran. *Zeitschrift Fur Naturforschung C* 2003; 58:629-631.
32. Cheikh-Rouhou S, Besbes S, Hentati B, Blecker C, Deroanne C, Attia H. *Nigella sativa* L.: Chemical composition and physicochemical characteristics of lipid fraction. *Food Chem* 2007; 101:673-681.
33. Yusuf S, Reddy S, Öunpuu S, Anand S. Global burden of cardiovascular diseases part I: general considerations, the epidemiologic transition, risk factors, and impact of urbanization. *Circulation* 2001; 104:2746-2753.
34. Mendis S, Puska P, Norrving B. Global atlas on cardiovascular disease prevention and control: World Health Organization; 2011.
35. Sabzghabae AM, Dianatkah M, Sarrafzadegan N, Asgary S, Ghannadi A. Clinical evaluation of *Nigella sativa* seeds for the treatment of hyperlipidemia: a randomized, placebo controlled clinical trial. *Med Arch* 2012; 66:198-200.
36. El-Dakhakhny M, Mady NI, Halim MA. *Nigella sativa* L. oil protects against induced hepatotoxicity and improves serum lipid profile in rats. *Arzneimittelforschung* 2000; 50:832-836.
37. Amarouch H, Zaoui A, Cherrah Y, Alaoui K, Mahassini N, Hassar M. Effects of *Nigella sativa* fixed oil on blood homeostasis in rat. *J Ethnopharmacol* 2002; 79:23-26.
38. Al-Naqeeq G, Al-Zubairi AS, Ismail M, Amom ZH, Esa NM. Antiatherogenic potential of *Nigella sativa* seeds and oil in diet-induced hypercholesterolemia in rabbits. *Evid Based Complement Alternat Med* 2011; 2011:213628.
39. Nader MA, El-Agamy DS, Suddek GM. Protective effects of propolis and thymoquinone on development of atherosclerosis in cholesterol-fed rabbits. *Arch Pharmacol Res* 2010; 33:637-643.
40. Le PM, Benhaddou-Andaloussi A, Elimadi A, Settaf A, Cherrah Y, Haddad PS. The petroleum ether extract of *Nigella sativa* exerts lipid-lowering and insulin-sensitizing actions in the rat. *J Ethnopharmacol* 2004; 94:251-259.
41. Zaoui A, Cherrah Y, Alaoui K, Mahassine N, Amarouch H, Hassar M. Effects of *Nigella sativa* fixed oil on blood homeostasis in rat. *J Ethnopharmacol* 2002; 79:23-26.
42. Bhatti IU, Rehman FU, Khan M, Marwat S. Effect of prophetic medicine kalonji (*Nigella sativa* L.) on lipid profile of human beings. An *in vivo* approach. *World Appl Sci J* 2009; 6:1053-1057.
43. Tasawar Z, Siraj Z, Ahmad N, Lashari MH. The effects of *Nigella sativa* (Kalonji) on lipid profile in patients with stable coronary artery disease in multan, Pakistan. *Pak J Nutr* 2011; 10:162.
44. Bamosa AO, Ali B, Sawayan S. Effect of oral ingestion *Nigella sativa* seeds on some blood parameters. *Saudi Pharm J* 1997; 5:126-129.
45. Bamosa AO, Ali BA, al-Hawsawi ZA. The effect of thymoquinone on blood lipids in rats. *Indian J Physiol pharmacol* 2002; 46:195-201.
46. Kanter M, Coskun O, Budancamanak M. Hepatoprotective effects of *Nigella sativa* L and *Urtica dioica* L on lipid peroxidation, antioxidant enzyme systems and liver enzymes in carbon tetrachloride-

- treated rats. *World J Gastroenterol* 2005; 11: 6684-6688.
47. Sowers JR, Epstein M. Diabetes mellitus and associated hypertension, vascular disease, and nephropathy an update. *Hypertension* 1995; 26:869-879.
48. Daneman D. Type 1 diabetes. *The Lancet* 2006; 367:847-858.
49. Anonymous. Guidelines N. Anaemia management in people with chronic kidney disease (CKD). Clinical Guidance. N.C.c.f.C. Conditions, London: Royal College of Physicians; 2006. p. 1-172.
50. Stadler K. Oxidative stress in diabetes. *Adv Exp Med Biol* 2012; 771:272-287.
51. Shabana A, El-Menyar A, Asim M, Al-Azzeh H, Al Thani H. Cardiovascular benefits of black cumin (*Nigella sativa*). *Cardiovascular Toxicol* 2013; 13:9-21.
52. Kanter M, Akpolat M, Aktas C. Protective effects of the volatile oil of *Nigella sativa* seeds on β -cell damage in streptozotocin-induced diabetic rats: a light and electron microscopic study. *J Molecular Histol* 2009; 40:379-385.
53. Abdelmeguid NE, Fakhoury R, Kamal SM, Al Wafai RJ. Effects of *Nigella sativa* and thymoquinone on biochemical and subcellular changes in pancreatic β -cells of streptozotocin-induced diabetic rats. *J Diabetes* 2010; 2:256-266.
54. Kanter M, Coskun O, Korkmaz A, Oter S. Effects of *Nigella sativa* on oxidative stress and β -cell damage in streptozotocin-induced diabetic rats. *Anat Rec A Discov Mol Cell Evol Biol* 2004; 279:685-691.
55. Rchid H, Chevassus H, Nmila R, Guiral C, Petit P, Chokairi M, et al. *Nigella sativa* seed extracts enhance glucose-induced insulin release from rat-isolated Langerhans islets. *Fundam Clin Pharmacol* 2004; 18:525-529.
56. Al-Hader A, Aqel M, Hasan Z. Hypoglycemic effects of the volatile oil of *Nigella sativa* seeds. *Pharm Biol* 1993; 31:96-100.
57. Fararh K, Atoji Y, Shimizu Y, Takewaki T. Insulinotropic properties of *Nigella sativa* oil in Streptozotocin plus Nicotinamide diabetic hamster. *Res Vet Sci* 2002; 73:279-282.
58. Kaleem M, Kirmani D, Asif M, Ahmed Q, Bano B. Biochemical effects of *Nigella sativa* L seeds in diabetic rats. *Indian J Exp Biol* 2006; 44:745-748.
59. Alenzi F, El-Bolkiny Y-S, Salem M. Protective effects of *Nigella sativa* oil and thymoquinone against toxicity induced by the anti-cancer drug cyclophosphamide. *Br J Biomed Sci* 2010; 67:20-28.
60. Hawsawi ZA, Ali BA, Bamosa AO. Effect of *Nigella sativa* (black seed) and thymoquinone on blood glucose in albino rats. *Ann Saudi Med* 2001; 21:242-244.
61. Marles RJ, Farnsworth NR. Antidiabetic plants and their active constituents. *Phytomedicine* 1995; 2:137-189.
62. Abu Khader MM. Thymoquinone: a promising antidiabetic agent. *Int J Diabetes Developing Countries* 2012; 32:65-8.
63. Salama RH. Hypoglycemic effect of lipoic acid, carnitine and *Nigella sativa* in diabetic rat model. *Int J Health Sci* 2011; 5:126-134.
64. Pari L, Sankaranarayanan C. Beneficial effects of thymoquinone on hepatic key enzymes in streptozotocin-nicotinamide induced diabetic rats. *Life Sci* 2009; 85:830-834.
65. Bamosa AO, Kaatabi H, Lebda FM, Elq A-MA, Al-Sultan A. Effect of *Nigella sativa* seeds on the glycemic control of patients with type 2 diabetes mellitus. *Indian J Physiol Pharmacol* 2010; 54:344-354.
66. Sultan MT, Butt MS, Karim R, Zia-Ul-Haq M, Batool R, Ahmad S, et al. *Nigella sativa* Fixed and Essential Oil Supplementation Modulates Hyperglycemia and Allied Complications in Streptozotocin-Induced Diabetes Mellitus. *Evid Based Complement Alternat Med* 2014; 2014:826380.
67. Ibrahim RM, Hamdan NS, Mahmud R, Imam MU, Saini SM, Rashid SN, et al. A randomised controlled trial on hypolipidemic effects of *Nigella Sativa* seeds powder in menopausal women. *J Transl Med* 2014; 12:82.
68. Sheikh T, Joshi D, Patel B, Modi C. Protective role of *Nigella sativa* against experimentally induced type-II diabetic nuclear damage in Wistar rats. *Veterinary World* 2013; 6:698-702.
69. Alam S, Reddy SK, Baig A, Reddy MK, Mohiuddin M, Reddy MV, et al. Evaluation of antidiabetic and anti-lipidemic potential of kalongi sugar powder water extract in stz induced diabetic rats. *Int J Pharm Pharm Sci* 2013; 5:94-96.
70. Asgary S, Ghannadi A, Dashti G, Helalat A, Sahebkar A, Najafi S. *Nigella sativa* L. improves lipid profile and prevents atherosclerosis: Evidence from an experimental study on hypercholesterolemic rabbits. *J Functional Foods* 2013; 5:228-234.
71. Ali SA, Asghar F, Nafees M, Tayyab M. Effect of *Nigella Sativa* (Kalonji) on Serum Lipid Profile. *ANNALS* 2012; 18:224-228.
72. Nasir A, Siddiqui MY, Mohsin M. Efficacy of Saboos-e-Asaphgol (*Plantago ovata*) and Kalonji (*Nigella sativa*) in the Management of Hypertriglyceridemia. *Int J Pharm India* 2013; 2:560-568.
73. Mahmoud MY. Effect of High Protein Diet Containing Fortified Bread with Fenugreek and *Nigella sativa* Seeds on Rats Suffering from Diabetes. *Pak J Nutr* 2013; 12:736-747.
74. Shafeeque A. Effects of volatile oil and nonsaponifiable fractions from *Nigella sativa* oil and its constituent, thymoquinone against cardiovascular risk parameters in experimental hyperlipidemia. *Indian ETD Repository @ INFLIBNET* 2013.
75. 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. *Diagnostic Pathol* 2013; 8:137.
76. Sobhi W, Khettal B, Belmouhoub M, Atmani D, Duez P, Benboubetra M. Hepatotoxicity and Langerhans islets regenerative effects of polar and neutral lipids of *Nigella sativa* L. in nicotinamide/streptozotocin-induced diabetic rats. *Pteridines* 2013; 22:97-104.
77. Jamal A, Hmza A, Omar E, Adnan A, Osman MT. *Nigella sativa* Oil Has Significant Repairing Ability of Damaged Pancreatic Tissue Occurs in Induced Type 1 Diabetes Mellitus. *Global J Pharmacol* 2013; 7:14-19.
78. Vanhoutte P, Shimokawa H, Tang E, Feletou M. Endothelial dysfunction and vascular disease. *Acta Physiologica* 2009; 196:193-222.

79. Idris-Khodja N, Schini-Kerth V. Thymoquinone improves aging-related endothelial dysfunction in the rat mesenteric artery. *Naunyn Schmiedebergs Arch Pharmacol* 2012; 385:749-758.
80. Zaoui A, Cherrah Y, Lacaille-Dubois M, Settaf A, Amarouch H, Hassar M. Diuretic and hypotensive effects of *Nigella sativa* in the spontaneously hypertensive rat. *Therapie* 1999; 55:379-82.
81. Demir H, Kanter M, Coskun O, Uz YH, Koc A, Yildiz A. Effect of black cumin (*Nigella sativa*) on heart rate, some hematological values, and pancreatic β -cell damage in cadmium-treated rats. *Biol Trace Elem Res* 2006; 110:151-162.
82. El Tahir KE, Ashour MM, Al-Harbi MM. The cardiovascular actions of the volatile oil of the black seed (*Nigella sativa*) in rats: elucidation of the mechanism of action. *Gen Pharmacol* 1993; 24:1123-1131.
83. Khattab MM, Nagi MN. Thymoquinone supplementation attenuates hypertension and renal damage in nitric oxide deficient hypertensive rats. *Phytother Res* 2007; 21:410-414.
84. Meral I, Donmez N, Baydas B, Belge F, Kanter M. Effect of *Nigella sativa* L. on heart rate and some haematological values of alloxan-induced diabetic rabbits. *Scand J Lab Anim Sci* 2004; 31:49-53.
85. Dehkordi FR, Kamkhah AF. Antihypertensive effect of *Nigella sativa* seed extract in patients with mild hypertension. *Fundam Clini Pharmacol* 2008; 22:447-452.
86. Qidwai W, Hamza HB, Qureshi R, Gilani A. Effectiveness, safety, and tolerability of powdered *Nigella sativa* (kalonji) seed in capsules on serum lipid levels, blood sugar, blood pressure, and body weight in adults: results of a randomized, double-blind controlled trial. *J Altern Complement Med* 2009; 15:639-644.
87. American Cancer Society. Cancer Facts and Statistics 2014 [25/4/2014]. Available from: <http://www.cancer.org/research/cancerfactsstatistics/index>
88. Servan-Schreiber D. Anti-cancer: a new way of life. *J Alternative Complementary Med* 2009; 15:805-6.
89. Wang S, Meckling KA, Marcone MF, Kakuda Y, Tsao R. Can phytochemical antioxidant rich foods act as anti-cancer agents? *Food Res Int* 2011; 44:2545-2554.
90. Wargovich MJ. Anti-cancer properties of fruits and vegetables. *HortSci* 2000; 35:573-575.
91. Ait Mbarek L, Ait Mouse H, Elabbadi N, Bensalah M, Gamouh A, Aboufatima R, et al. Anti-tumor properties of blackseed (*Nigella sativa* L.) extracts. *Brazilian J Med Biol Res* 2007; 40:839-847.
92. Gali-Muhtasib H, Roessner A, Schneider-Stock R. Thymoquinone: a promising anti-cancer drug from natural sources. *Int J Biochem Cell Biol* 2006; 38:1249-1253.
93. Rooney S, Ryan M. Effects of alpha-hederin and thymoquinone, constituents of *Nigella sativa*, on human cancer cell lines. *Anticanc Res* 2005; 25:2199-2204.
94. Al-Ali A, Alkhawajah AA, Randhawa MA, Shaikh NA. Oral and intraperitoneal LD50 of thymoquinone, an active principle of *Nigella sativa*, in mice and rats. *J Ayub Med Coll Abbottabad* 2008; 20:25-27.
95. El-Kadi A, Kandil O, editors. Effects of *Nigella sativa* (the black seed) on immunity. Proceeding of the 4th International Conference on Islamic Medicine, Kuwait Bull Islamic Med; 1986.
96. Mabrouk GM, Moselhy SS, Zohny SF, Ali EM, Helal TE, Amin AA, et al. Inhibition of methylnitrosourea (MNU) induced oxidative stress and carcinogenesis by orally administered bee honey and *Nigella* grains in Sprague Dawely rats. *J Exp Clinical Canc Res* 2002; 21:341-346.
97. Swamy S, Tan B. Cytotoxic and immunopotentiating effects of ethanolic extract of *Nigella sativa* L. seeds. *J Ethnopharmacol* 2000; 70:1-7.
98. Kumara SS, Huat BT. Extraction, isolation and characterisation of antitumor principle, α -hederin, from the seeds of *Nigella sativa*. *Planta medica* 2001; 67:29-32.
99. Jafri SH, Glass J, Shi R, Zhang S, Prince M, Kleiner-Hancock H. Thymoquinone and cisplatin as a therapeutic combination in lung cancer: In vitro and in vivo. *J Exp Clin Canc Res* 2010; 29:87.
100. Bourgou S, Pichette A, Marzouk B, Legault J. Bioactivities of black cumin essential oil and its main terpenes from Tunisia. *South Afr J Bot* 2010; 76:210-216.
101. Al-Sheddi ES, Farshori NN, Al-Oqail MM, Musarrat J, Al-Khedhairi AA, Siddiqui MA. Cytotoxicity of *Nigella Sativa* Seed Oil and Extract Against Human Lung Cancer Cell Line. *Asian Pacific J Canc Prev* 2014; 15:983-987.
102. Globocan. Fact sheet [27/4/2014]. Available from: http://globocan.iarc.fr/Pages/fact_sheets_cancer.aspx.
103. Farah IO, Begum RA. Effect of *Nigella sativa* (N. *sativa* L.) and oxidative stress on the survival pattern of MCF-7 breast cancer cells. *Biomed Sci instrum* 2002; 39:359-364.
104. Farah IO. Assessment of cellular responses to oxidative stress using MCF-7 breast cancer cells, black seed (N. *Sativa* L.) extracts and H2O2. *Int J Envir Res Publ Health* 2005; 2:411-419.
105. El-Aziz MA, Hassan HA, Mohamed MH, Meki AR, Abdel-Ghaffar SK, Hussein MR. The biochemical and morphological alterations following administration of melatonin, retinoic acid and *Nigella sativa* in mammary carcinoma: an animal model. *Int J Exp pathol* 2005; 86:383-396.
106. Effenberger K, Breyer S, Schobert R. Terpene Conjugates of the *Nigella sativa* Seed-Oil Constituent Thymoquinone with Enhanced Efficacy in Cancer Cells. *Chem biodivers* 2010; 7:129-139.
107. Baharetha HM, Nassar ZD, Aisha AF, Ahamed MB, Al-Suede FS, Kadir MO, et al. Proapoptotic and Antimetastatic Properties of Supercritical CO2 Extract of *Nigella sativa* Linn. Against Breast Cancer Cells. *J Med Food* 2013; 16:1121-1130.
108. Salim EI, Fukushima S. Chemopreventive potential of volatile oil from black cumin (*Nigella sativa* L.) seeds against rat colon carcinogenesis. *Nutr Canc* 2003; 45:195-202.
109. Gali-Muhtasib H, Diab-Assaf M, Boltze C, Al-Hmaira J, Hartig R, Roessner A, et al. Thymoquinone extracted from black seed triggers apoptotic cell death in human colorectal cancer cells via a p53-dependent mechanism. *Int J Oncol* 2004; 25:857-866.

110. Rooney S, Ryan M. Modes of action of alpha-hederin and thymoquinone, active constituents of *Nigella sativa*, against HEp-2 cancer cells. *Anticancer Res* 2005; 25:4255-4259.
111. Khan N, Sultana S. Inhibition of two stage renal carcinogenesis, oxidative damage and hyperproliferative response by *Nigella sativa*. *Eur J Canc Prev* 2005; 14:159-168.
112. Tabasi N, Mahmoudi M, Rastin M, Sadeghnia HR, HosseinPour Mashhadi M, Zamani Taghizade Rabe S, et al. Cytotoxic and apoptogenic properties of *Nigella sativa* and thymoquinone, its constituent, in human renal cell carcinoma are comparable with cisplatin. *Food Agric Immunol* 2015; 26:138-156.
113. Iddamaldeniya SS, Wickramasinghe N, Thabrew I, Ratnatunge N, Thammitiyagodage MG. Protection against diethylnitrosoamine-induced hepatocarcinogenesis by an indigenous medicine comprised of *Nigella sativa*, *Hemidesmus indicus* and *Smilax glabra*: a preliminary study. *J Carcinogenesis* 2003; 2:1-6.
114. Iddamaldeniya SS, Thabrew M, Wickramasinghe S, Ratnatunge N, Thammitiyagodage MG. A long-term investigation of the anti-hepatocarcinogenic potential of an indigenous medicine comprised of *Nigella sativa*, *Hemidesmus indicus* and *Smilax glabra*. *J Carcinogenesis* 2006; 5:1-7.
115. Thabrew MI, Mitry RR, Morsy MA, Hughes RD. Cytotoxic effects of a decoction of *Nigella sativa*, *Hemidesmus indicus* and *Smilax glabra* on human hepatoma HepG2 cells. *Life Sci* 2005; 77:1319-1330.
116. Samarakoon SR, Thabrew I, Galhena PB, De Silva D, Tennekoon KH. A comparison of the cytotoxic potential of standardized aqueous and ethanolic extracts of a polyherbal mixture comprised of *Nigella sativa* (seeds), *Hemidesmus indicus* (roots) and *Smilax glabra* (rhizome). *Pharmacognosy Res* 2010; 2:335-342.
117. Salomi MJ, Nair SC, Panikkar KR. Inhibitory effects of *Nigella sativa* and saffron (*Crocus sativus*) on chemical carcinogenesis in mice. *Nutr Canc* 1991; 16:67-72.
118. Chehl N, Chipitsyna G, Gong Q, Yeo CJ, Arafat HA. Anti-inflammatory effects of the *Nigella sativa* seed extract, thymoquinone, in pancreatic cancer cells. *HPB* 2009; 11:373-381.
119. Banerjee S, Kaseb AO, Wang Z, Kong D, Mohammad M, Padhye S, et al. Antitumor activity of gemcitabine and oxaliplatin is augmented by thymoquinone in pancreatic cancer. *Canc Res* 2009; 69:5575-5583.
120. Torres MP, Ponnusamy MP, Chakraborty S, Smith LM, Das S, Arafat HA, et al. Effects of thymoquinone in the expression of mucin 4 in pancreatic cancer cells: implications for the development of novel cancer therapies. *Mol Canc Therapeutics* 2010; 9:1419-1431.
121. Shafi G, Munshi A, Hasan TN, Alshatwi AA, Jyothy A, Lei DK. Induction of apoptosis in HeLa cells by chloroform fraction of seed extracts of *Nigella sativa*. *Cancer Cell Int* 2009; 9:29.
122. Hasan TN, Shafi G, Syed NA, Alfawaz MA, Alsaif MA, Munshi A, et al. Methanolic extract of *Nigella sativa* seed inhibits SiHa human cervical cancer cell proliferation through apoptosis. *Natur Prod Commun* 2013; 8:213-216.
123. Salomi N, Nair S, Jayawardhanan K, Varghese C, Panikkar K. Antitumor principles from *Nigella sativa* seeds. *Canc Lett* 1992; 63:41-46.
124. Worthen DR, Ghosheh OA, Crooks P. The in vitro anti-tumor activity of some crude and purified components of blackseed, *Nigella sativa* L. *Anticancer Res* 1997; 18:1527-1532.
125. Badary O, Al-Shabanah O, Nagi M, Al-Rikabi A, Elmazar M. Inhibition of benzo (a) pyrene-induced forestomach carcinogenesis in mice by thymoquinone. *Eur J Canc Prev* 1999; 8:435-440.
126. Badary OA, El-Din AMG. Inhibitory effects of thymoquinone against 20-methylcholanthrene-induced fibrosarcoma tumorigenesis. *Canc Detect Prev* 2001; 25:362-368.
127. Norwood A, Tucci M, Benghuzzi H. A comparison of 5-fluorouracil and natural chemotherapeutic agents, EGCG and thymoquinone, delivered by sustained drug delivery on colon cancer cells. *Biomed Sci Instrum* 2006; 43:272-277.
128. Norwood A, Tan M, May M, Tucci M, Benghuzzi H. Comparison of potential chemotherapeutic agents, 5-fluorouracil, green tea, and thymoquinone on colon cancer cells. *Biomed Sci Instrum* 2005; 42:350-356.
129. El-Mahdy MA, Zhu Q, Wang QE, Wani G, Wani AA. Thymoquinone induces apoptosis through activation of caspase-8 and mitochondrial events in p53-null myeloblastic leukemia HL-60 cells. *Int J Canc* 2005; 117:409-417.
130. Ahmed WA, Hassan SA, Galeb FM, El-Taweel MA, Abu-Bedair FA. The in vitro promising therapeutic activity of thymoquinone on hepatocellular carcinoma (HepG2) cell line. *Global Veterinaria* 2008; 2:233-241.
131. Hassan S, Ahmed W, M.Galeb F, El-Taweel M, A.Abu-Bedair f. In vitro challenge using thymoquinone on hepatocellular carcinoma (HepG2) cell line. *Iran J Pharm Res* 2010; 7:283-90.
132. El-Najjar N, Chatila M, Moukadem H, Vuorela H, Ocker M, Gandesiri M, et al. Reactive oxygen species mediate thymoquinone-induced apoptosis and activate ERK and JNK signaling. *Apoptosis* 2010; 15:183-195.
133. Salomi M, Nair SC, Panikkar K. Inhibitory effects of *Nigella sativa* and saffron (*Crocus sativus*) on chemical carcinogenesis in mice. *Nutr Cancer* 1991; 16:67-72.
134. Kaseb AO, Chinnakannu K, Chen D, Sivanandam A, Tejwani S, Menon M, et al. Androgen receptor- and E2F-1-targeted thymoquinone therapy for hormone-refractory prostate cancer. *Canc Res* 2007; 67:7782-7788.
135. Koka PS, Mondal D, Schultz M, Abdel-Mageed AB, Agrawal KC. Studies on molecular mechanisms of growth inhibitory effects of thymoquinone against prostate cancer cells: role of reactive oxygen species. *Exp Biol Med* 2010; 235:751-760.
136. Yi T, Cho S-G, Yi Z, Pang X, Rodriguez M, Wang Y, et al. Thymoquinone inhibits tumor angiogenesis and tumor growth through suppressing AKT and extracellular signal-regulated kinase signaling pathways. *Mol Canc Therapeutics* 2008; 7:1789-1796.
137. Xuan NT, Shumilina E, Qadri SM, Goetz F, Lang F. Effect of thymoquinone on mouse dendritic cells. *Cellular Physiol Biochem* 2010; 25:307-314.

138. Sethi G, Ahn KS, Aggarwal BB. Targeting nuclear factor- κ B activation pathway by thymoquinone: role in suppression of antiapoptotic gene products and enhancement of apoptosis. *Mol Canc Res* 2008; 6:1059-1070.
139. Ismail M, Al-Naqeep G, Chan KW. *Nigella sativa* thymoquinone-rich fraction greatly improves plasma antioxidant capacity and expression of antioxidant genes in hypercholesterolemic rats. *Free Radical Biol Med* 2010; 48:664-672.
140. Ebru U, Burak U, Yusuf S, Reyhan B, Arif K, Faruk TH, et al. Cardioprotective effects of *Nigella sativa* oil on cyclosporine A-induced cardiotoxicity in rats. *Basic Clin Pharmacol Toxicol* 2008; 103:574-580.
141. Barron J, Benghuzzi H, Tucci M. Effects of thymoquinone and selenium on the proliferation of mg 63 cells in tissue culture. *Biomed Sci Instrum* 2007; 44:434-440.
142. Bawadi H. A, Bansode R. R, Losso J. N. Thymoquinone in the control of hypoxia-induced angiogenic disease biomarkers: Insight into the mechanism of action in vitro, IFT Annual Meeting 2004; Las Vegas NV USA, 2004: 7-8
143. Swamy SM, Huat BT. Intracellular glutathione depletion and reactive oxygen species generation are important in α -hederin-induced apoptosis of P388 cells. *Mol Cell Biochem* 2003; 245:127-139.

Archive of SID