Efficacy of Nigella sativa (Kalongi) for the Treatment of Hyperlipidemia in Sprague Dawley Rats


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Abstract

Background: To analyze the efficacy of Nigella sativa (Kalongi) for the treatment of hyperlipidemia in Sprague Dawley rats.

Methods: Twenty albino rats were divided in two groups. Both groups were fed on ad libitum diets for 02 weeks. Then cholesterol rich diet was given to all rats for 08 weeks followed by treatment with Kalonji for 06 weeks in group II but no treatment in Group I. Blood samples were collected and serum of all rats were analyzed in three phases i.e., at baseline, after cholesterol rich diet and after treatment period for serum cholesterol, serum triglycerides levels, HDLC, LDLC along with serum transaminase levels (ALT).

Results: Baseline levels of all parameters were comparable in both groups. The cholesterol rich diet affected all parameters of lipid profile. Nigella sativa (kalonji) showed significant improvement in the lipid profile of rats. The p value < 0.05 of group I and II documented that Nigella sativa (kalonji) had significantly affected the lipid profile as compared to control group. No significant difference was noted in ALT levels between two groups at the end of treatment (p > 0.05).

Conclusion: Nigella sativa reduces cholesterol and triglycerides, with adequate safety profile

Key Words: Nigella sativa(Kalongi), Hyperlipidemia

Introduction

Hypercholesterolemia is one of the major risks for heart diseases. The evidence confirms that elevated levels of low-density lipoprotein cholesterol (LDLC) are associated with increased risk of atherosclerosis and lowering LDL-C reduces cardiovascular risk. Hyperlipidemia is a well known unique and sufficient cause for the development of coronary heart disease without the episode of hypertension, diabetes, smoking, male gender and inflammation.1,2

Hyperlipidemia can be controlled through dietary modification or by drug therapies. Statins (HMG-CoA reductase inhibitors) are most commonly used. Studies are associated with side effects including gastrointestinal disorders, myalgia, arthralgia, upper respiratory infections, headache, abdominal pain, constipation and nausea. Biochemical abnormalities have occurred with simvastatin treatment with elevation of serum transaminase levels.3

Nigella sativa (Kalonji) is a flowering plant, belongs to the family Ranunculaceae and genus Nigella. It is a widely investigated herb for use as medicine. It has been traditionally used as medicine in Mediterranean region, Asia and Africa. The usefulness of Nigella sativa (Kalonji) has been mentioned in various Ahadees-e-Nabvi. The prophet (PBUH) said, "Hold onto the use of the black seed for it has a remedy for every illness except death". Different scholars reported Nigella sativa as a beneficial medicinal herb like Al-Biruni, Ibne-Sina and Greek physician Dioscorides. Nigella sativa is a very effective herb and was traditionally used for many different ailments like bronchial asthma inflammatory diseases, for the enhancement of milk production, bronchitis and rheumatism. Its various forms like the oil form and powder form are used in the treatment of different diseases. The ingredients of seeds of Nigella sativa were found effective for reducing hepatotoxicity and nephrotoxicity. Multi-systemic beneficial actions of Nigella sativa are also reported including hypcholesterolemic effects & anti-oxidant effects.4-11

Materials and Methods

In this case control study, of 16 weeks duration, twenty healthy rats, 90 - 120 days of age were included. The rats were obtained from National institute of Health, NIH, Islamabad. They were kept under standard conditions with a daily photoperiod of 12 hours light and 12 hours dark at temperature ± 5°C at animal house of NIH, Islamabad. The sample size was divided in two equal groups, 10 in each group. Group I (Control Group) did not receive any treatment during third phase of study. Group II (Experimental
Group) received Nigella sativa (1000 mg/Kg) during third phase of study.

The study was carried out in three phases. 1st and 2nd phase of the study were same for both groups. However, during third phase, the groups differed on treatment basis. During first phase, the rats of all groups were kept under optimum temperature (24±2°C) and hygienic conditions with food and water available at all times for acclimatization with the environment. In 2nd phase, the rats were given cholesterol rich diet (cholesterol powder) for 08 weeks. The diet was given ad libitum to the rats. The average per day feed of a rat is about 20 gm/day. The constituents of the routine diet includes, wheat flour (28.5%), wheat bran (28.5%), fish meal (15%), dried skimmed milk (20%), soya bean vegetable Oil (5%), molasses (2%), common salt (0.5%), vitaminera premix (0.5%). During 2nd phase, supplementation of cholesterol rich powder as 200gm/5 Kg was done in the routine diet for all rats. The average consumption of feed by a rat is about 20gm/day. In this way, 5 kg feed serves for one week for rats in study population (20 rats). The third phase comprised of treatment. During this phase, one group did not receive any treatment, second group received Nigella sativa. The duration of this phase was six weeks. At the end of each phase blood samples of all rats were taken for lipid profile and ALT estimations.

Fasting (12-14 hour fasting) blood sample of all the rats (both groups) were taken via tail vein. The blood samples were collected in plain tubes. Baseline blood samples were collected before the start of cholesterol rich diet. Samples were collected before the initiation of cholesterol rich diet (baseline), after completion of 08 weeks of cholesterol rich diet and after completion of six weeks of Nigella sativa administration.

Serum cholesterol, serum triglycerides, serum HDL and serum ALT levels were estimated in all the blood samples. Serum LDL was also estimated by using the formula:LDL=Total Cholesterol-HDL Cholesterol-TG/5

Results

Mean baseline weight of rats was 215.16±14.17gms and 241.67±14.10gms at the end of 2nd phase. There was no significant difference in weight of rats between two groups (with p value 0.973 and 0.826 respectively p>0.05) at this level. The mean weight during third phase of study was 221.5±14.51gm. During 3rd phase, there was difference in weight of rats between control vs Kalonji group (Table 1). Significant difference was observed in weight of rats between 1st vs 2nd phase of study with p value .000 (p<0.05). Insignificant difference in the weight of control group rats was observed in 2nd vs 3rd phase of study. Statistically significant difference was noted in weight of rats of Kalonji group in 2nd vs 3rd phase of study

Average baseline cholesterol level was 99.43±3.72 mg/dl (range 90 to 108 mg/dl) and 195.75±5.34 mg/dl. There was insignificant difference in the baseline values of cholesterol level between two groups in both phases. Significant difference was observed in the 3rd phase cholesterol levels between two groups. Insignificant difference was observed in control group between 2nd vs 3rd phase of study but experimental group showed significant difference in cholesterol levels between 2nd vs 3rd phase (after treatment). Average HDL level in study population at the end of 1st phase was 39.8±1.42 mg/dl. Insignificant difference in HDL levels of two groups was observed at this level. The average HDL level of rats in control group of study was 39.7±1.42 mg/dl, 20.9±1.45 mg/dl and 31.2±2.62 mg/dl during 1st, 2nd and 3rd phase respectively. Significant difference was noted in HDL level of rats between 1st vs 2nd phase of study in control group and insignificant difference in the cholesterol level of control group rats was observed in 2nd vs 3rd phase of study. Insignificant difference was noted between 1st and 3rd phase HDL levels in rats of Kalonji group.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Phases</th>
<th>Study Population</th>
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<tbody>
<tr>
<td>Weight</td>
<td>1st Phase</td>
<td>215±12.12</td>
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<tr>
<td></td>
<td>2nd Phase</td>
<td>242±13.02</td>
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<td></td>
<td>3rd Phase</td>
<td>224.5±11.57</td>
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<tr>
<td>Cholesterol</td>
<td>1st Phase</td>
<td>99.15±3.81</td>
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<tr>
<td></td>
<td>2nd Phase</td>
<td>195.75±5.34</td>
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<td></td>
<td>3rd Phase</td>
<td>130±30.34</td>
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<tr>
<td>Triglycerides</td>
<td>1st Phase</td>
<td>77.4±4.15</td>
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<tr>
<td></td>
<td>2nd Phase</td>
<td>102.7±3.58</td>
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<td></td>
<td>3rd Phase</td>
<td>84.3±8.58</td>
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<td>HDL-C</td>
<td>1st Phase</td>
<td>39.75±1.45</td>
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<td></td>
<td>2nd Phase</td>
<td>21±1.55</td>
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<td></td>
<td>3rd Phase</td>
<td>35.3±4.74</td>
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<tr>
<td>LDL-C</td>
<td>1st Phase</td>
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<tr>
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<td>2nd Phase</td>
<td>153±21±5.4</td>
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<td>77.84±33.1</td>
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<td>ALT</td>
<td>1st Phase</td>
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<tr>
<td></td>
<td>2nd Phase</td>
<td>29.8±7.19</td>
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<tr>
<td></td>
<td>3rd Phase</td>
<td>30.15±5.90</td>
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Average value of triglycerides at the end of 1st phase was 77.5±4.11 mg/dl . There was insignificant difference in baseline and 2nd phase triglyceride levels between two groups. Significant difference was noted in triglyceride levels between control group vs Kalonji group at the end of 3rd phase. Significant difference was noted in triglyceride level of rats between 1st vs 2nd phase, whereas insignificant difference in the triglycerides level was observed in 2nd vs 3rd phase of study in control group rats. Insignificant difference was noted between 1st and 3rd phase triglyceride levels of rats in Kalonji group.

Mean LDL level after completion of 1st phase was 44±3.68 mg/dl . Insignificant difference was observed in baseline and 2nd phase LDL levels of both groups. In 3rd phase, there was significant difference in LDL levels between control vs kalonji group. Significant difference was noted in LDL level of rats between 1st vs 2nd phase, whereas insignificant difference was observed in 2nd vs 3rd phase of study in control group rats. In Kalonji group, significant difference was observed in LDL levels in this group between 1st vs 2nd and 2nd vs 3rd phase of study. However, insignificant difference was noted between 1st and 3rd phase LDL levels in Kalonji group.

At completion of 1st phase, the mean ALT level of study population was 30.17±7.30 U/l . Statistically insignificant difference was observed in baseline, 2nd phase and 3rd ALT levels of control and kalonji groups.

**Discussion**

The basic mechanism for the development of coronary artery disease involves mainly retention and deposition of serum lipids especially LDL-C in the coronary arteries . This will cause the flow of blood to decrease towards the heart muscle. The available treatment involves drugs causing the lowering of cholesterol levels. However, some side effects of few of these drugs are reported which limit their usage in certain individuals.

Medicinal plants are getting the attention of medical researcher to treat different disease other than synthetic medicine. Different herbal treatments were tried for the treatment of hypercholesterolemia. The Nigella sativa commonly known as Kalonji is one such herb. In present study, its efficacy was tried for the treatment of hyperlipidemia in rats.

The comparison of baseline parameters at the end of first phase of study, including weight of the rats, serum cholesterol, serum triglycerides, serum HDL-C and serum LDL-C levels in both groups showed statistically insignificant difference (p>0.01). Hence the effects observed later during the study in each group are related to the diet, treatment etc during the study and there was no study bias in any group. Study carried out on Sprague-Dawley rats observed almost same baseline values of different parameters of lipid profile.

There was 26.5 gm increase in average weight of rats after cholesterol rich diet of eight weeks. An average increase of 38 grams in weight of the rats was observed by Matos et al. The increase in weight is relatively higher as compared to present study. The weight increase is likely due to the fact that the authors included soyabean oil and fiber content in diet which is produced relatively higher weight. In 2011, A.Rezq and Al-Khaimesy studied the effects of cholesterol diet in Sprague-Dawley rats in which sheep fat and 0.5% cholic acid was also used in addition to 1% cholesterol to induce hyperlipidemia. The authors observed 33.14 gm gain in weight in positive group of study as compared to negative control group after cholesterol rich diet.

The average cholesterol level showed 51% increase and there was an increase of 71.4% in LDL-C as compared to baseline levels. Matos et al, reported the average increase in cholesterol level and LDL-C as 56% and 78.5% respectively after cholesterol rich diet supplementation of eight weeks. The increase in cholesterol levels, LDL-C levels is relatively higher as compared to our study. The inclusion of soyabean oil in addition to cholesterol by Matos et al., in diet, has produced relatively higher values of total cholesterol and LDL-C levels. In study of Rezq and Al-Khaimesy, comparing the control negative group with positive group, who received fatty diet, the authors observed 44% increase in serum cholesterol levels, 33% and 83% increase in serum triglycerides and LDL-C levels respectively.

There is a decrease of 47% in HDL-C levels at the end of 2nd phase of our study and 34% in study of Matos et al. (2005). A.Rezq and Al-Khaimesy (2011) observed 33% decrease in HDL-C levels in positive group who received sheep fat and 1% cholesterol for six weeks as compared to negative control who received standard diet. The slight variations of HDL-C levels between our study, Matos et al. and Rezq and Al-Khaimesy is due to minor differences in study design. Moreover, the diet supplementation was continued for six weeks as compared to eight weeks in our study.

During the third phase of the present study, control group received no treatment whereas, experimental group received kalonji (1000 mg/kg/day) for 06
weeks. The treatment showed a significant change in the lipid profile of the rats. It showed 48.4% improvement (reduction) in total cholesterol levels. There was 70% and 25.3% reduction in LDL-C and triglycerides levels respectively at the end of 3rd phase. Moreover, HDL-C levels of rats in this group showed 53.5% improvement after kalonji treatment. The same dose of Nigella sativa was used by other authors\(^1\) and comparable results were found in their study in terms of reduction in total cholesterol levels, LDL-C levels and serum triglycerides levels of the rats with an increase in HDL-C levels.\(^8\) In another study of rats kalonji was added to the diet of the rats in a dose of 30mg/kg body weight, simultaneously with cholesterol rich constituents of the diet. \(^9\) It was concluded that the group with kalonji showed comparatively lesser rise in total serum cholesterol level of the rats after 20 weeks of the treatment. Similar effects of Nigella sativa on lipid profile of albino rats was observed by other authors.\(^20,21\) In another study kalonji was used as an effective herbal drug in maintaining lipid profile in normal states and also no adverse effects were observed in this study.\(^22\) The change noted after four weeks of two spoons of kalonji powder per day in mentioned study was 8.83%, 17.41% and 11.94% in total cholesterol, LDL-C and HDL-C respectively as compared to baseline levels. Although, the percent change noted after treatment was statistically significant (p <0.001), yet it differs with our study in terms of percentage. The probable reasons of difference could be the duration of treatment (04 weeks versus 06 weeks in our study).

**References**


