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Effect of *Raphanus Sativus* (Radish) Leaf Extract and High Doses of Atorvastatin on Lipid Profile

Obaid Anwar[1], Hafiz Muhammed Imran Aziz[2], Asad Mahmood Khan[3], Rehana Ramzan[4], Saleha Shafi[5], Zobia Mushtaq[6]

1 *Associate Professor, Department of Pharmacology, Abwa Medical College, Faisalabad Pakistan*
Idea conception, Data Collection, approval of final draft

2 *Assistant Professor, Department of Pharmacology, Bakhtawar Amin Medical College, Multan Pakistan*
Idea conception, Data Collection, approval of final draft

3 *Professor, Department of Pharmacology, Sahiwal Medical College, Sahiwal Pakistan*
Manuscript writing, approval of final draft

4 *Ph.D. Scholar, Department of Biochemistry, Government College University, Faisalabad Pakistan*
Data Analysis, Manuscript Writing

5 *Department of Pharmacy, The University of Faisalabad, Faisalabad Pakistan*
Data Collection, Approval of final draft

6 *Professor, Department of Pharmacology, Gujranwala Medical College, Gujranwala Pakistan*
Literature Search

CORRESPONDING AUTHOR
Dr. Asad Mahmood Khan
Professor, Department of Pharmacology, Sahiwal Medical College, Sahiwal, Pakistan
Email: drasadmahmood@gmail.com

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ABSTRACT

Background: Hyperlipidemia is a key component in a devastating chain reaction that ultimately results in mortality among the elderly. Atorvastatin is the most commonly advised statin to patients suffering from hyperlipidemia. *Raphanus Sativus (R Sativus)* commonly used in salads contains many antioxidants. **Objective:** To determine the effect of *Raphanus Sativus* (Radish) leaf extract and high doses of Atorvastatin on Lipid Profile. **Study Design:** Randomized control trail (RCT). **Settings:** Pharmacology department, Faisalabad Medical University, Faisalabad Pakistan. **Duration:** 3 Months from July 2020 to September 2020. **Methods:** Ethanolic extract of *R Sativus* was screened for beneficial effects on lipid profile by atorvastatin. **Results:** Findings were compared with both the normal and experimental control by assessing the different components of the lipid profile, such as the LDL, HDL, triglycerides, and serum cholesterol. There was a substantial decrease in weight observed in all groups compared to the negative control group A. **Conclusion:** Ethanolic extract of leaves of radish used in three increasing doses has a significant effect on lipid profile in albino rabbits when used along with atorvastatin.

Keywords: *LDL, HDL, Atorvastatin, Raphanus sativus.*

INTRODUCTION

Traditional medicine is becoming more and more important economically on a global scale. In emerging nations, national governments and healthcare professionals are beginning to value complementary and alternative medicine more and more.[1] Approximately 85% of people worldwide utilize herbal remedies to cure and prevent illness. Both developed and emerging nations are seeing an increase in demand. There are several chemical components in both the plant and the fruits.[2]

Medicinal plants are those that include components, or precursors thereof, that may be used to create effective medications for medical conditions. Since ancient times, all societies have used medicinal plants as a source of treatment.[3] It has been shown that over 5000 plants have medicinal uses. The elderly guy has protected his health by using plants as their primary source of defense against a variety of ailments. Many agricultural plants are being used in folk medicine as a remedy for a variety of illnesses.[4] Numerous papers and articles have been produced as a result of recent studies on the use of natural ingredients to cure illness. The growing number of studies devoted to drug development based on natural products might be explained by their potential as future therapeutic candidates that can reduce side effects and undesirable effects by using components that work synergisticall.[5] Thus, a key component of safe and successful hyperlipidemia treatment approaches is the screening of natural products by looking into their bioactive components.[6]

In Korea, radish, is a popular Cruciferous vegetable. Radish contains alkaloid compounds, phenolic

compounds, glycosides, tannins, carbohydrates, saponins, flavonoids, volatile oil and amino acids[7]. The methanolic, aqueous, anhydrophobic radish extracts or certain phytochemicals found in radishes, such as anthocyanins, phenolic acids, and isothiocyanates. Consequently, it has also shown possible nutritional and health-promoting qualities, including anti-inflammatory, anti-hypertensive, and protective effects on vascular smooth muscle cells, as well as anti-diabetic and anti-cancer effects.[8] Given its potent properties against oxidants and inflammation, radish may be the finest natural option to find new medications against hyperlipidemia. Furthermore, radish is inexpensive and simple to grow and prepare. It is thus quite likely to be used in medicine once its effectiveness is established.[9] The purpose of this research was to assess the impact on lipid parameters in albino rabbits of three escalating dosages of *R Sativus* leaf extract combined with high doses of atorvastatin.

METHODS

This Randomized control trail (RCT) was conducted in Pharmacology department, Faisalabad Medical University, Faisalabad from July 2020 to September 2020. Fresh Raphanus sativus leaves weighing one hundred kg were bought from the local vegetable market in Faisalabad. Using given keys, plant sample was taxonomically identified and validated (Herbarium voucher no. 173-14-1) at the University of Agriculture (UAF), Faisalabad, Pakistan. Fresh leaves were properly cleansed with distilled water to eliminate any dust or foreign material. For ten days, leaves were dried in the shade at room temperature. They were then dried for six hours in a hot air oven at 60°C before being ground in an electric grinder. After passing powder through a mesh sieve, it was extracted with 80% ethanol and filtered using filter paper. The extracts were concentrated in a hot air oven set at 37°C, lyophilized using a freeze drying equipment and then stored in an airtight container in a freezer set to -20°C.[10] Atorvastatin with a purity of 99% was acquired from Stand-Pharm Pharmaceuticals (PVT) Ltd. 2 grammes of it were dissolved in 100 milliliters of distilled water, resulting in a concentration of 20 milligrams of atorvastatin per 1 milliliter of distilled water for administration. All reagents used were of analytical quality. A total of 50 mature male rabbits, aged 6-8 weeks and weighing between 1-1.5 kg, were acquired from the local market in Faisalabad, Pakistan. The rabbits were placed in separate iron cages, each labelled and suitable dimensions inside Clinical Medicine and Surgery department at the UAF, Faisalabad. The cages were maintained at a constant room temperature of 22±2°C and provided with enough ventilation for the duration of the experiment. The rabbits were provided with seasonal forage and grain, and had unrestricted access to water. Before the experimentation, the rabbits had a one-week acclimatization period. The rabbits had a fasting period of 16-18 hours before to the administration of test extracts.

Table 1: Study Design

Group	No. of rabbits	Treatment
A (Normal control)	10	Routine diet throughout the experiment for 28 days.
B (Positive control)	10	Treated daily with atorvastatin (20mg/kg as hepatotoxic drug P.O. for 28 days.
C (Experimental group I	10	Treated daily with 20mg/kg atorvastatin + 100mg/kg *R. sativus* leaf extract P.O. for 28 days.
D (Experimental group II)	10	Treated daily with 20mg/kg atorvastatin + 200mg/kg *R. sativus* leaf extract P.O. for 28 days.
E (Experimental group III)	10	Treated daily with 20mg/kg atorvastatin + 400mg/kg *R. sativus* leaf extract P.O. for 28 days.

Biochemical Study: By using spectrophotometric assay kits, the serum cholesterol levels including LDL, HDL and serum triglycerides were determined with automatic chemistry analyzer at Allied hospital Faisalabad, Pakistan.

Data was entered and analyzed using SPSS 20.0. Mean ± SE was calculated for numerical variables, which included the serum cholesterol levels including LDL, HDL and serum triglycerides. Paired sample t test was applied at baseline and post treatment groups to see any significant impact on serum lipids level. P-value ≤ 0.05 was considered as statistically significant.

RESULTS

Table 2 shows serum cholesterol level of rabbits on 0 and 28 days. Results showed atorvastatin significantly reduced serum cholesterol level in 28 days. While for groups using *R. sativus* extract, highly significant difference was noted in serum cholesterol of rabbits after 28 days in group using group C, D and E.

Table 2: Effect of R. sativus extract on Serum Cholesterol of atorvastatin treated rabbits.

Groups	Day 0	Day 28
A (normal)	33.90±2.228	32.80±1.960
B (At. 20mg/kg)	39.00±2.226	18.40±2.617
C (At. 20mg/kg + R. S 100mg/kg)	49.10±3.990	23.60±1.565
D (At. 20mg/kg + R. S 200mg/kg)	38.90±3.579B	19.20±1.356C
E (At. 20mg/kg + *R. S* 400mg/kg)	46.00±5.177	43.50±4.922
P-value	0.41	0.000

Table 2 shows serum Triglycerides level of rabbits on 0 and 28 days. Results showed atorvastatin significantly reduced serum triglycerides level in 28 days. While for groups using *R. sativus* extract, highly significant difference was noted in serum triglycerides of rabbits after 28 days in group C, D and E.

Table 3: Effect of R. sativus extract on Triglycerides of atorvastatin treated rabbits.

Groups	Day 0	Day 28
A (normal)	122.80±3.054	122.50±3.191
B (At. 20mg/kg)	127.90±4.848	43.20±3.596
C (At. 20mg/kg + *R. S* 100mg/kg)	123.00±6.481	71.40±6.208
D (At. 20mg/kg + *R. S* 200mg/kg)	119.40±9.453	71.20±5.081
E (At. 20mg/kg + *R. S* 400mg/kg)	123.90±5.311	119.60±5.905
P-value	0.914	0.000

Table 4 shows the serum LDL level of rabbits on 0 and 28 days. Results showed atorvastatin significantly reduced serum LDL level in 28 days. While for groups using *R. sativus* extract, highly significant difference was noted in serum LDL of rabbits after 28 days in group C, D and E.

Table 5: Effect of R. sativus extract on LDL of atorvastatin treated rabbits

Groups	Day 0	Day 28
A (normal)	78.70±4.006A	79.10±2.779AB
B (At. 20mg/kg)	66.80±3.952B	74.20±1.806BC
C (At. 20mg/kg + *R. S* 100mg/kg)	52.40±4.129C	89.00±4.544A
D (At. 20mg/kg + *R. S* 200mg/kg)	57.30±2.741BC	86.50±4.719A
E (At. 20mg/kg + *R. S* 400mg/kg)	54.90±2.157C	64.90±5.699C
P-value	0.231	0.001

Table 5 demonstrate serum HDL level of rabbits on 0 and 28 days. Results showed atorvastatin significantly reduced serum HDL level in 28 days. While for groups using *R. sativus* extract, highly significant difference was noted in serum HDL of rabbits after 28 days in group C, D and E.

Table 5: Effect of R. sativus extract on HDL of atorvastatin treated rabbits

Groups	Day 0	Day 28
A (normal)	47.10±1.370	48.80±1.665
B (At. 20mg/kg)	47.60±1.621	57.30±1.674
C (At. 20mg/kg + *R. S* 100mg/kg)	46.10±1.941	56.30±0.831
D (At. 20mg/kg + *R. S* 200mg/kg)	48.10±1.969	56.00±2.113
E (At. 20mg/kg + *R. S* 400mg/kg)	48.30±1.606	54.50±1.797
P-value	0.899	0.006

DISCUSSION

Hyperlipidemia is a crucial component in a complex sequence of events that ultimately results in life-threatening diseases, particularly in older individuals. The decrease in blood cholesterol levels might potentially mitigate or prevent cardiovascular risks, hence potentially saving lives. Existing synthetic hypolipidemic medications are linked to several adverse effects.[11] Statins are globally used to reduce cholesterol levels, with atorvastatin being the most often prescribed medication for this purpose.[12] There is an increasing interest in nutraceuticals, such as nutrients, herbals, and dietary supplements, as an alternative to contemporary medicine. These substances provide health advantages.[10]

Raphanus sativus leaves are rich of flavonoids. They are used as antioxidants and thus conserve cellular integrity of the live cells.[13] Being very common and economical, ethanolic extract of leaves of radish was used in three increasing doses to assess its properties against the on lipid profile in albino rabbits. There was a decrease in lipid profile as observed on weekly basis over a period of 28 days as compared to negative control group A which showed an increase in weight. This decrease was observed in all treated groups including the positive control group B. So, our study showed that ethanolic extract of leaves of radish used in three increasing doses has a significant effect on lipid profile in albino rabbits. Our results are in accordance with a study by MF Abdelhameed *et al* using *R. Sativus* seed oil in rat fed on high-fat diet.[14] Their research demonstrated a substantial reduction ($p \leq 0.01$) in levels of cholesterol, triglycerides, and total lipids in the HFD+ radish oil group at various time intervals. The findings of another study unequivocally show that the hot water extract of R. sativus reduces harmful lipids in the blood and liver, specifically total cholesterol (TC) and triacylglycerol (TG). At the same time, it increases the elimination of cholesterol through faeces in both rats with normal cholesterol levels and rats with artificially induced high cholesterol levels.[15]

CONCLUSION

Ethanolic extract of leaves of radish used in three increasing doses has a significant effect on lipid profile in albino rabbits when used along with atorvastatin.

LIMITATIONS

This was animal-based study with limited sample size so the results of this study cannot be generalized without further studies.

SUGGESTIONS / RECOMMENDATIONS

The administration of atorvastatin lead to decrease lipid profile however, this can only be proven by further and comprehensive studies involving human subjects.

CONFLICT OF INTEREST / DISCLOSURE

No.

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