# STUDIES ON SOME ANTIATHEROSCLEROTIC MEDICINAL PLANTS OF BANGLADESH

# Md Zakir Sultan\*, Mohammad Abdul Matin, Rabeya Miazi and Mohammad A. Rashid<sup>1</sup>

Centre for Advanced Research in Sciences (CARS), University of Dhaka, Dhaka-1000, Bangladesh

Keywords: Atherosclerosis, Cholesterol, Triglyceride, LDL, HDL, Medicinal plants.

#### Abstract

The study investigated the effect of some medicinal plant extracts on atherosclerosis lowering activity in mice model. The results revealed that among the extracts, garlic, ginger and cluster fig reduced cholesterol, triglycerides, and low-density lipoprotein (LDL) cholesterol, but raised the level of high-density lipoprotein (HDL) cholesterol. It is distinctly apparent that the three herb extracts out of the nine herbs studied have anti-atherosclerotic activity.

Atherosclerosis-related cardiovascular disease is one of the primary causes of morbidity and death. Atherosclerosis leads to cardiovascular disease which is the most common cause of death in Bangladesh as well as all over the world. It is also reported that a hypercholesterolemic or hyperlipidemic state also increases the level of plasma lipoproteins e.g. low density lipoprotein (LDL), triglyceride (TG) and total cholesterol (TC)] (Blankenberg *et al.* 2003, Bogani *et al.* 2007). These modifications cause the artery thickening and the arterial walls become less elastic. It is frequently described as the artery's stiffening or furring. Eventually, the wall stiffening can cause increased pulse pressure, heart block, heart attack, obesity and so on. Now-a-days, cardiac patients especially atherosclerotic patients in Bangladesh have been increasing at an alarming rate. Atherosclerosis reducing drugs can be used as prophylaxis and for the treatment of atherosclerosis and obesity (Rogers *et al.* 1986, Cai and Harrison 2000, Covas *et al.* 2006, Cullinen 2006, Zheng *et al.* 2006).

Being a developing country, many people live under the poverty level in Bangladesh. They are unable to get the benefits of modern medicines for the treatment of such diseases due to high cost of the medicines. Moreover, they have calamitous side effects. However, herbal medicines are comparatively cheaper and safe for human consumption (Anjum *et al.* 2017, 2021, 2022).

Although, plants are a major source of traditional medicines that are highly effective in treating many infectious diseases, the chemical compounds isolated from plants often do not serve as medicines. However, they can provide clues for potential new drug development. Therefore, the primary objective of this project was to study some medicinal plants and herbs that lower the levels of plasma lipoproteins, such as total cholesterol (TC), triglyceride (TG), and low-density lipid (LDL), which may help treat obesity and atherosclerosis. This will also scientifically validate the folkoric uses of these medicinal herbs.

The following nine dried plant samples were collected from the local market of Bangladesh and identified by a botanist (Table 1).

<sup>\*</sup>Author for correspondence: <zakir.sultan@du.ac.bd>. <sup>1</sup>Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Dhaka, Dhaka-1000, Bangladesh.

Local name	English name	Botanical name
Ananta Mul	Indian sarsaparilla	Hemidesmus indicus (L.) R.Br.
Bakul biz	Spanish cherry seeds	Mimusops elengi L.
Pipul	Long pepper	Piper longum L.
Labonga	Cloves	Syzygium aromaticum (L.) Merr. & L.M.Perry
Ulat Kombal	Devil's cotton	Abroma augustum (L.) L.f.
Ada	Ginger	Zingiber officinale Rosc.
Rasun	Garlic	Allium sativum L.
Dumur	Cluster fig	Ficus racemosa L.
Basak pata	Malabar nut leaf	Justicia adhatoda L.

Table 1. List of plants studied for anti atherosclerotic activities.

The plant samples were properly cleaned with water, cut into pieces, dried in an oven at  $40^{\circ}$ C for 24 hrs. Each sample (100 g) was soaked in 1.5 L methanol and left for 15 days with periodic shaking and stirring. The mixture was then filtered through cotton filter followed by Whatman filter paper No.1. The resulting filtrates were concentrated using a Buchii rotavapour at the temperature of  $40^{\circ}$ C and at reduced pressure to obtain the crude extracts.

Mice were used in the trial against atherosclerosis. Sixty albino mice, weighing an average of 40.0 g and belonging to both sexes were purchased from ICDDR'B, Dhaka, Bangladesh. The mice were housed in the Institute of Nutrition and Food Science. Animal House at Dhaka University, at 21-25°C and a 12 hrs light/dark cycle. The experiment began after the mice had been acclimated for five days.

Twelve groups were formed in which five mice were randomly assigned to each group. Group 1 was termed as control group. The mice in this group were given water and a regular diet. Group 2 termed as positive control group in which the mice with atherosclerosis caused by a high-lipid diet were given atorvastatin (1.5 mg/kg body weight/day) as a reference medication for 25 days. Group 3 to 12 were treated with plant or herb extracts. After being fed a high-lipid diet (HLD) mice with atherosclerosis were given herbal extracts (10 mg/kg body weight/day in water) for 25 days. After completion of the treatment period, mice were sacrificed and serum was collected for biochemical studies.

The procedure for measuring total cholesterol levels in a sample requires three enzymes i.e., peroxidase (PO), cholesterol esterase (CE), and cholesterol oxidase (CO) (Allain *et al.* 1974, Amundson and Zhou 1999). The amount of cholesterol was measured using a commercially available enzyme-linked immunosorbent assay kit and its protocol. The total cholesterol was calculated using the following equation:  $(A_{sample}/A_{standard}) \times C$  standard = mg/dL total cholesterol.

Serum triglycerides were determined by the method developed by Bucolo and David 1973, Fossati and Prencipe 1982. The amount of triglycerides was determined using a commercially available enzyme-linked immunosorbent assay kit and its protocol. The following equation was used for the calculation of triglycerides:  $(A_{sample}/A_{standard}) \times C_{standard} = mg/dL$  triglycerides. LDL cholesterol determination employed the technique of a unique precipitation method of LDL cholesterol by polyvinyl sulfate and then sedimentation of the precipitant (Arsman *et al.* 1984). The amount of LDL was measured by using commercially available enzymatic immunoassay kits. The following equation was used for the calculation of LDL cholesterol:  $(A_{supernatant}/A_{standard}) \times C_{standard} = mg/dL$  LDL cholesterol. This technique employed a separation process after specific precipitation by

#### STUDIES OF SOME ANTIATHEROSCLEROTIC MEDICINAL PLANTS

centrifugation (Burstein *et al.* 1980). The amount of HDL was measured by using commercially available enzymatic immunoassay kits. The following equation was used for the calculation of HDL cholesterol: ( $A_{supernatant}/A_{standard}$ ) ×C<sub>standard</sub> = mg/dL HDL cholesterol.

Among the plant extracts, garlic, ginger and cluster fig increased HDL and decreased the bad lipids. Devil's cotton and spanish cherry seeds were found that they possessed total cholesterol lowering activities (Fig. 1).

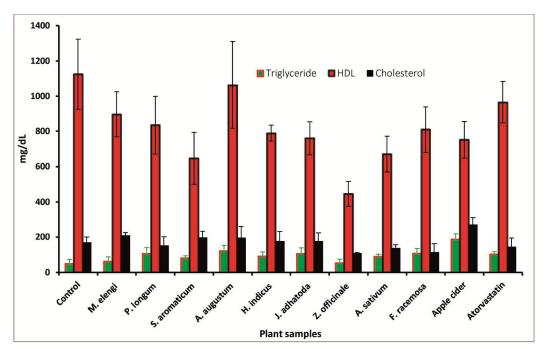


Fig. 1. Comparison of lipids levels of mice treated with herbal extracts.

The *in vivo* analysis for atherosclerosis lowering activity of the herbal extracts was performed in mice. The results revealed that among the herbs extracts, garlic, ginger and cluster fig reduced cholesterol, triglycerides, and low-density lipoprotein (LDL) cholesterol, but enhanced the highdensity lipoprotein (HDL) cholesterol level. It is distinctly apparent from the above findings that some herbs/plant extracts have anti-atherosclerotic activity. Therefore, it was concluded that these herbs/plants may be good candidate to be used in the treatment of atherosclerosis or for further chemical investigation to isolate the atherosclerosis lowering constituents.

### Acknowledgements

Financial Supports for the work from the Ministry of Science and Technology, Government of the People's Republic of Bangladesh under Allocation for Science and Technology for the fiscal year of 2020-2021 (Project No.: 39.00.0000.009.14.011.20- EAS 419) is gratefully acknowledged.

### References

- Aguilera CM, Ramirez-Tortosa MC, Mesa MD, Ramirez-Tortosa CL and Gil A 2002. Sunflower, virginolive and fish oils differentially affect the progression of aortic lesions in rabbits with experimental atherosclerosis. Atherosclerosis **162**: 335.
- Allain CC, Poon LS, Chan CS, Richmond W and Fu PC 1974. Enzymatic determination of total serum cholesterol. Clin. Chem. 20: 470-475.
- Amundson DM and Zhou M 1999. Fluorometric method for the enzymatic determination of cholesterol. J. Biochem. Biophys. Meth. **38**: 43-52.
- Anjum A, Sultan MZ, Al Mansur MA, Hasan CM and Rashid MA 2022. Assessment of antioxidant, antimicrobial, cytotoxicity and thrombolytic potential of *Erythrina fusca* Lour grown in Bangladesh. Bangladesh Pharm. J. 25: 1-8.
- Anjum A, Sultan MZ, Hasan CM and Rashid MA 2017. Antibacterial and cytotoxic constituents from Bridelia verrucosa Haines growing in Bangladesh. Dhaka Univ. J. Pharm. Sci. 16: 61-68.
- Anjum A, Kuddus MR, Sultan MZ, Al Mansur A, Hasan CM and Rashid MA 2021. Phytochemical and biological investigation of *Bridelia tomentosa* Blume growing in Bangladesh. Dhaka Univ. J. Pharm. Sci. 20: 213-218. DOI: https://doi.org/10.3329/dujps.v20i2.57171
- Arsman G, Jabs HU, Kuhnert U, Nolte W and Schriewer H 1984. LDL-cholesterol, polyvinyl sulphate method. Clin. Chim. Acta. **140**: 77.
- Blankenberg S, Barbaux S and Tiret L 2003. Adhesion molecules and atherosclerosis. Atherosclerosis 170: 191.
- Bogani P, Galli C, Villa M and Visioli F 2007. Postprandial anti-inflammatory and antioxidant effects of extra virgin olive oil. Atherosclerosis **190**: 181.
- Brand K, Page S, Rogler G, Bartsch A, Brandl R, Knuechel R, Page M, Kaltschmidt C, Baeuerle PA and Neumeier D 1996. Activated transcription factor nuclear factor-kappa B is present in the atherosclerotic lesion. J. Clin. Invest. 97: 1715.
- Breslow JL 1997. Cardiovascular disease burden increases, NIH funding decreases. Nat. Med. 3: 600.
- Bucolo G and David H 1973. Quantitative determination of serum triglycerides by the use of enzymes. Clin. Chem. **19**: 476.
- Burstein M, Scholnick HR and Morfin R 1980. Severe combined hyperlipidemia. Scand. J. Clin. Lab. Invest. **40**: 560-572.
- Cai H and Harrison DG 2000. Endothelial dysfunction in cardiovascular diseases: the role of oxidant stress. Circ. Res. **87**: 840.
- Covas MI, de la Torre K, Farre-Albaladejo M, Kaikkonen J, Fito M, LopezSabater C, Pujadas-Bastardes MA, Joglar J, Weinbrenner T, Lamuela-Raventos RM and de la Torre R 2006. Postprandial LDL phenolic content and LDL oxidation are modulated by olive oil phenolic compounds in humans. Free Radic. Biol. Med. 40: 608.
- Cullinen K 2006. Olive oil in the treatment of hypercholesterolemia. Med. Health R. I 89: 113.
- Fossati R and Prencipe L 1982. Serum triglycerides determined colorimetrically with an enzyme that produces hydrogen peroxide. Clin. Chem. 28: 2077-2079.
- Gonzalez-Santiago M, Martin-Bautista E, Carrero JJ, Fonolla J, Baro L, Bartolome MV, Gil-Loyzaga P and Lopez-Huertas E 2006. One-month administration of hydroxytyrosol, a phenolic antioxidant present in olive oil, to hyperlipemic rabbits improves blood lipid profile, antioxidant status and reduces atherosclerosis development. Atherosclerosis **188**: 35.
- Rogers KA, Hoover RL, Castellott JJJ, Robinson JM and Karnovsky MJ 1986. Dietary cholesterol-induced changed in macrophage characteristics relationship to atherosclerosis. Am. J. Pathol. **125**: 284.
- Zheng S, Qian Z, Sheng L and Wen N 2006. Crocetin attenuates atherosclerosis in hyperlipidemic rabbits through inhibition of LDL oxidation. J. Cardiovasc. Pharmacol. **47**: 70.

(Manuscript received on 30 September, 2024; revised on 7 January, 2025)