Effect of an unani formulation on lipid profile in rat

Coronary artery disease (CAD) is one of the most important cause of death all over world.^[1] Hyperlipidemia is one of the risk factors for CAD. Data show that 25-30% risk of CAD is reduced by treating hyperlipidemia. In unani pharmacopoeia several formulations with large or small number of ingredients have been described for the treatment of cardiovascular disorders. Some of the formulations are being used in unani medicine with good results. There is possibility of errors of interpretation during clinical use of such drugs. Therefore, it was considered worth while to evaluate the role of a formulation containing Bombyx mori, Nepata hindostana and Terminalia arjuna in hyperlipidemia. The study was conducted in the Department of Ilmul Advia, A.K. Tibbiya College and the Department of Pharmacology, J. N. Medical College, A.M.U., Aligarh, India. Adult albino rats of either sex weighting 150- 200 g were taken. They were provided standard diet (Lipton India) and water ad libitum. Twelve hours dark and light cycles were maintained. They were divided into 4 groups containing 6 animals each. The temperature was maintained 26 ± 2 °C.

Isoproterenol hydrochloride purchased from Sigma USA. *N. hindostana* (Badranj boya), *B. mori* (Abraisham) and bark of *T. arjuna* were procured from Dawakhana Tibiyya College, A.M.U., Aligarh and were identified by comparison for its macroscopic and microscopic characters with authentic specimens of *N. hindostana, B. mori* and bark of *T. arjuna* at the Botanical Survey of India, and the Forest Reaserch Institute, Dehradun, India. The bark of *T. arjuna* and whole plant of *N. hindostana* were dried in shade, powered in electric grinder to a coarse 20-40 mash size and extracted in soxhlet apparatus with distilled water for six hours separately. The aqueous extract of *B. mori* (raw silk cocoon) was obtained by reflex method. All three extract were separately concentrated at water bath and stored at room temperature for use.

Extracts of *Terminalia arjuna, Nepata hindostana* and *Bombyx mori* were combined in ratio of 1:2:1 respectively and freshly dissolved in distilled water on each day before administration.

The animals were divided into four groups. Group I was given distilled water and served as control. Group II were given isoproterenol 20 mg/ 100 g body weight subcutaneously twice at interval of 24 h to induce myocardial necrosis.^[2] Group III was administered unani formulation in the dose of 100 mg / 100 g of body weight, p. o. daily for 61 days (The test drugs are likely to produce optimum benefit after over 40 days of treatment). Group IV was administered unani formulation in the dose of 100 mg/100 g of body weight, p.o. daily for 60 days followed by isoproterenol subcutaneously twice at interval of 24 h. The dose was calculated in the usual manner by multiplying the unani clinical dose by factor 7 for rat.^[3] Twelve hours after second dose of isoproterenol the animals were sacrificed by cervical decapitation method. The blood was collected in plane vials, serum was separated by centrifugation and stored at 4°C till analysed.

The serum was analysed for total cholesterol and free cholesterol,^[4] triglycerides^[5] phospholipids,^[6] free fatty acids,^[7] high density (HDL),^[8] low density (LDL)^[9] and very low density lipoproteins (VLDL)^[10] by using "Diagnostic kit Syntropic: UV-VIS Spetrophotometer 117.

The results were analysed by one-way analysis of variance (ANOVA) followed by Dunnett's test. P<0.05 was considered statistically significant. The study showed significant increase in total cholesterol, free cholesterol, triglycerides, phospholipids, free fatty acids, low density (LDL) and very low density lipoproteins (VLDL) in isoproterenol treated group as compared with their respective control but the levels were statistically same in control and in unani formulation pre treated rats. Serum level of high density lipoprotein (HDL)

Table 1

The effect of unani formulation on serum lipoproteins (mg/dl) in rats

Lipid		Total cholesterol	Free cholesterol	S.triglycerides	Phospholipids	Free fatty acid [@]	LDL	VLDL	HDL
Control		60.7±6.2	86.3±7.5	21.1±2.3	53.8±8.4	40.1±5.5	38.3±1.4	23.3±2.3	17.6±2.4
Isop treated		147.5±8.4**	126.4±5.1**	75.2±6.2**	117.4±10.0**	112.2±8.6**	72.3±4.1**	39.8±2.2*	11.1±1.2*
UF		86.5 <u>+</u> 5.2	56.9 <u>+</u> 2.3	20.3±2.7	56.1±4.3	36.2±2.7	32.4±2.2	20.3±2.2	19.0±1.7
UF + Isop		82.8±6.2	67.6±3.6	27.7±5.5	63.5±4.9	44.1±4.2	38.4±2.7	28.3±3.2	17.8±1.1
One-way	F	1158	1628	1113	827.3	1338	335	121	32.1
ANOVA	df	3,20	3,20	3,20	3,20	3,20	3,20	3,20	3,20
	Ρ	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.01	<0.01

Values are mean ± SD. n = 6 in each group. *P<0.01, ** P<0.001 compared to control; Isop = Isoproterenol; UF = Unani formulation; @measured in µmole/dL.

was lowest in isoproterenol treated groups as compared with control and unani formulation pre-treated group. [Table 1] All animals of different groups were given similar diet and there was no significant change in the body weight.

Research in cardiovascular pharmacology in the past few years has been mainly focussed on hypolipidemic agents including herbal drugs. *B. mori* and *N hindostana* were reported as cardiotonic and cardioprotective drugs. *T. arjuna* is used in cardiovascular disorders.

It is well known that hyperlipidemia is one of the major risk factors for atherosclerosis. An increase in the concentration of lipids results in liberation of lisosome and trigger cells degeneration. Major component of total cholesterol is low density lipoprotein (LDL) which is directly related to CAD. It is recognised as major atherogenic lipoprotein and primary target of lipid lowering therapy. The serum low density lipoprotein (LDL) in unani formulation pre-treated group was lower as compared with isoproterenol treated group. High density lipoprotein (HDL) has preventive role in CAD, it has been shown to reduce the endothelial incorporation of lysophosphatidyl cholines (lyso-PC). The HDL level was significantly raised in unani formulation pretreated group as compared with isoproterenol treated group. Recently more emphasis is given that elevated level of LDL-C is important factor for CAD and lowering its level through diet and medication has been shown to reduce the progression of CAD. Free fatty acid level was significantly high in isoproterenol treated animals as compared to unani formulation pre-treated animals. It is reported that increased free fatty acid is associated with a rise in myocardial oxygen consumption.

Silk cocoons and bark of *T arjuna* mainly contain amino acids such as tyrosine, histidine, tryptophan, cysteine etc., which have been reported to act as antioxidants. The presence of soluble fibres and sitostanols (beta-sitosterol) could also explain the hypocholesterolaemic effect of arjuna bark. *N hindostana* and *T. arjuna* both drugs have phenolic

compounds and flavonoids abundantly, so they are potent quenchers of free radicals and also react quite rapidly with O_2 . In our study all the three ingredients of unani formulation have antioxidants compounds, which may be having protective role in hyperlipidemia.

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