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Effect of *Embilica officinalis* Fruit Extract on Haematological Profile and Serum Lipid Variables of Albino Rats

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Abstract: The effect of aqueous extract of *Emblica officinalis* fruits on some haematological and serum lipid parameters in rats during a seven day administration of the doses of 250mg/kg and 500 mg/kg body weight orally was investigated. The parameters evaluated include serum lipids, red and white blood cell indices. The results show that the extract administered significantly increased packed cell volume, haemoglobin concentration, red blood cell, MCH, MCHC, MCV and platelet count t at the dose of 250mg /kg and 500 mg/kg body weight when compared with control. Whereas the platelet was significantly increased at 250mg/kg body weight but at 500mg/kg body weight the count significantly reduced Also, the extract significantly increased white blood cell count at all doses administered when compared with control. Moreover, the extract significantly reduced ($p<0.05$, $p<0.1$) total cholesterol concentration, triglycerides and HDL-cholesterol concentration in the serum while it had no significant effect on serum LDL-cholesterol concentration at all doses administered when compared with controls. The results of this study suggest that the extract may have beneficial effect on serum cholesterol concentration and triglycerides reduction as well as in anaemia and immunity dependent disorders.

Key words: *Emblica officinalis* · Haematology · Lipids

INTRODUCTION

Herbal medicines are in great demand in the developed world for primary health care due to their efficacy, safety and lesser side effects [1]. Recently, considerable attention has been paid to utilize eco-friendly and bio-friendly plant based products [2, 3]. Herbal medicine has become a popular form of healthcare. Even though several differences exist between herbal and conventional pharmacological treatments, herbal medicine can be tested for efficacy using conventional trial methodology [4]. Several specific herbal extracts have been demonstrated to be efficacious for specific conditions [5]. The traditional preparations comprise medicinal plants, minerals, organic matter, etc. Herbal drugs constitute only those traditional medicines which primarily use medicinal plant preparations for therapy. The earliest recorded evidence of their use in Indian, Chinese, Egyptian, Greek, Roman and Syrian texts dates back to about 5000 years. The herbal medicines/traditional medicaments have, therefore, been derived from rich traditions of ancient civilizations and scientific heritage [6]. Fruits are amongst the first food items known to human beings. Fruits, whether fresh or dried, have always formed a part of the staple diet of human beings. The reason for this is that they are rich in nutrients and provide some of the essential minerals, vitamins and the like, to our body. Apart from that, they also help in curing a number of diseases [7].

*Emblica officinalis*Gaertn (commonly known in India as Amla, Syn. *Phyllanthus emblica* L.; Family: Euphorbiaceae) is regarded as ‘‘one of the best rejuvenating herbs’’ in the Ayurveda: an Indian traditional medicinal science. *Emblica officinalis* extract contains several antioxidants such as emblicanin A and B, gallic acid, ellagic acid, ascorbic acid that possesses strong antioxidative activity [8, 9]. The fruit extract has many pharmacological activities for the treatment of a number of diseases.

Several recent reports revealed that fruit extract of *Emblicaofficinalis* protect against radiation [10, 11], antiatherosclerosis [12], possess antidiabetic activity [13, 14], inhibits aging process [15], gastroprotective [16], cytoprotective and immunomodulatory [17]. Despite its

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extensive medicinal use no information is available related to its effects on haematology and lipid profile. Hence the present work investigates the effect of *Emblica officinalis* fruit extract on haematology and lipid profile of albino rats.

MATERIALS AND METHODS

Collection of Plant Material: Fresh fruits of *Emblicaofficinalis* were collected from Ranchi and the seeds were removed. The fruit was then dried in shade under 28±2°C for 6 to 7 days. Then they were crushed into coarse powdery substance by using electric grinder. The coarse powdery substance was dried again and was then sieved to get fine powder using the fine plastic sieve, which was then stored in an air tight bottle in the laboratory until required.

Extract Preparation: 50 g of the sieved powder was weighed accurately and subjected to extraction in a Soxhlet apparatus at room temperature using ~350 mL distilled water. The extract obtained was filtered, concentrated in rotary flash evaporator and maintained at 45°C the percentage yield of each extract were calculated and the dried extracts were stored in air tight containers at room temperature for further studies.

Animals: MaleAlbino rats (175-200 g) were used in the study. They were maintained under standard laboratory conditions at ambient temperature of 25±2°C and 50±15% relative humidity with a 12-h light/12-h dark cycle. Animals were fed with a commercial pellet diet and water *ad libitum*. The experiments were performed after prior approval of the study protocol by the institutional animal ethics committee of Ranchi University, Ranchi.

Experimental Design: The animals were randomly assigned into three groups of four rats each as follows:

Group 1: Received 1mL of distilled water orally

Group 2: Received 250mg/kg body weight of *E. officinalis* orally.

Group 3: Received 500mg/kg body weight of *E. officinalis* orally.

Sample Collection: By the end of each experimental period, the rats were reweighed, starved for 24 hours and sacrificed under chloroform anesthesia. 5mL of blood was collected from each animal by cardiac puncture using sterile needle and syringe. Part of the blood sample was put into test tubes and allowed to clot for 30 minutes before centrifuging at 800g (Wisperfuge, 1384, Samson, Holland) for 5 minutes. The supernatant was used for the lipid analysis. The remaining blood sample was put in an EDTA bottles for haematological determinations.

Analytical Procedure

Estimation of Lipid Profile: Estimation of total cholesterol, HDL cholesterol, LDL cholesterol and triglycerides was done by cholesterol oxidase - phenol aminoantipyrine method [18].

Estimation of Hematological Profile: The haemoglobin (Hb) level was measured by the cyanomethaemoglobin method. The Red Blood Cell (RBC) and Reticulocyte counts were determined by visual method [19]. Packed cell volume (PCV) was measured using microheamatocrit method and total White Blood Cell (WBC) count was estimated by visual method [20]. The RBC indices were calculated from the RBC count, Hb level and PCV estimations [19, 20].

Estimation of Thyroid Hormones: Estimation of serum T3, T4, TSH was done by chemiluminescence immunoassay method [21].

Statistical Analysis: All results were expressed as mean ± standard error of mean (S.E.M.). Data was analyzed using one-way ANOVA followed by Dunnett's-test. $p<0.05$ was considered as statistically significant.

RESULTS AND DISCUSSION

The effect of the oral administration of aqueous extract of *E. officinalis* on some serum lipid indices is presented in Table 1.The extract significantly reduced ($p<0.05$) serum total cholesterol concentration while it had no significant effect ($p>0.05$) on serum HDL-cholesterol concentration at all doses administered when compared with control. However, the extract significantly decreased ($p<0.05$) serum triacylglycerol concentration at the dose of 250 mg/kg as well as 500 mg/kg body ($p>0.05$) it when compared with control.

The aqueous extract of *E. officinalis* had significant effect on RBC, Hb, MCHC, MCH, PCV, MCV, neutrophils, basophils, monocytes, lymphocytes and eosinophils. The WBC was significantly elevated ($p< 0.05$) in the group treated with 250 mg/kg body where as the count

Table 1: Effect of *E. officinalis* extract on lipid profile of rats for 7 days

Parameter	Control	L.D(250 mg/kg b.wt)	H.D(500mg/kg b.wt)
Total Cholesterol (mg %)	59.83±2.31	46.51±1.88^{b,a}	37.33±2.16^{a,a}
High Density Lipid (mg %)	32±2.6	30.55±1.81^{b,a}	29.5±1.87^{b,a}
Low Density Lipid (mg %)	20±2.36	20.83±2.48^{ns}	20.83±2.31^{ns}
Triglyceride (mg %)	117.5±1.87	113.33±2.16^{a}	98.83±2.31^{a,b}

values are expressed as Mean ± SD from the experiments, where n=6, $^{a}p<0.05$,$^{b}p<0.1$, ns = non-significant relative to control.

Table 2: Effect of *E. officinalis* extract on haematological profile of rats for 7 days

PARAMETER	CONTROL	L.D(250 mg/kg b.wt)	H.D(500mg/kg b.wt)
Hematocrit (%)	39±2.60	45.83±0.44$^{a,\ ns}$	45.71±0.53^{a,ns}
Mean Corp.Vol (%)	94.33±3.31	94.9±0.26^{ns,a}	97.7±1.04^{ns,a}
Hemoglobin(g/dl)	11.8±0.31	15.95±0.32^{d,ns}	15.85±0.25^{e,ns}
Red Blood Cells(×10^6/µl)	4.26±1.08	4.58±0.04^{ns}	4.56±0.03^{ns}
Platelet(×10^3/µl)	339±2.38	275.83±2.85^{e,c}	375.66±4.17^{f,c}
White Blood Cells(×10^3/µl)	6.86±0.36	8.09±0.59^{a}	9.19±0.03^{a}
Neutrophil (%)	56.83±0.30	52.86±0.40^{e,b}	59.96±0.45^{e,b}
Lymphocyte (%)	32.46±1.25	23.76±0.48^{e,a}	29.33±0.37^{a}
Monocyte (%)	5.68±0.28	6.53±0.56^{ns}	6.83±0.40^{ns}
Eosinophil (%)	0.73±0.25	5.36±0.65^{a,ns}	5.8 ±0.9^{a,ns}
Basophil (%)	0.28±0.03	1.56±0.49^{a,ns}	1.28±0.03^{g,ns}
Mean Corp.Hemo (pg)	30.09±1.16	24.16 ±2.48^{ns}	27.86±2.32^{ns}
Mean Corp.Hemo.Conc (g/dl)	31.58±0.56	33.0. ±1.86^{ns}	34.03±0.39^{f,ns}

Values are expressed as Mean ± SD, n=6, where $^{a}p<0.001$, $^{b}p<0.005$, $^{c}p<0.05$, $^{d}p<0.0025$, $^{e}p<0.10$, $^{f}p<0.01$ and ns=non significant relative to control.

significantly decreased ($p< 0.05$) in 500mg/kg body weight. The platelet also significantly increased ($p< 0.05$) in rats treated with both the doses (Table 2).

High blood cholesterol concentration is one of the important risk factors for cardiovascular disease [22]. Thus the reduction in serum total cholesterol concentration effected by the extract is beneficial and may reduce the risk of cardiovascular disease because agents that have the ability to lower cholesterol concentration in the blood have been reported to reduce vascular resistance by improving endothelial function [22].

Emblica has the ability to lower cholesterol by the unique concerted action of both inhibiting cholesterol production and enhancing cholesterol degradation. It has also shown the amazing property of actually reducing plaque in clogged arteries caused by high cholesterol levels in some animal studies. Rabbits that had been fed a high cholesterol diet were given fresh *Emblica* juice for 60 days. Their serum cholesterol and LDL levels were lowered by 83% and 90%, respectively. Similarly, the tissue lipid levels showed a significant reduction and aortic plaques decreased in size. Consequently, the researchers suggested that *Emblica* be used as a pharmaceutical tool for patients wanting to reduce their cholesterol levels [23] Similar alterations in lipid as well as haematological profiles were reported in various other plant extracts such as *Bulbine natalensis* [24], *Bougainvillea spectabilies leaves* [22] and *Fadogia agrestis* stem [24].

Assessment of haematological parameters can not only be used to determine the extent of deleterious effect of extracts on the blood of an animal, but it can also be used to explain blood relating functions of a plant extract or its products [23]. The results obtained shows significant values of WBC, therefore it is clear that an increase in the number of WBC is a normal reaction of rats to foreign substances, which alter their normal physiological processes. Platelets play a major role in the development as well as in the stability of atherosclerotic plaques and as a consequence, anti-platelet agents have been used clinically in patients at risk for myocardial ischemia, unstable angina and acute myocardial infarction [25, 26]. Therefore the high dose (500 mg/kg body weight) of the *E. officinalis*extracts useful in reducing the platelets which in turn might be useful in reducing the cardiovascular diseases assome studies suggested various mechanisms by which flavonoid exert its antiplatelet property by lowering intracellular Ca2+ levels; alteration in the metabolism of cAMP and thromboxane A2 [27, 28]. The haemoglobin content, RBC and PCV has also significantly increased stimulate erythropoietin release in the kidney which is the humoral regulators of RBC production [29, 30].

The result of this study suggested that *E. officinalis* extracts studied showed positive haematological activities in rats and can be recommended in the management of anaemia and immunity dependent disorders as well as in regulating the cholesterol and triglyceride levels.

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