



ANTI-ANEMIC ACTIVITY OF HYDRO-ALCOHOLIC LEAF EXTRACT OF *TAMARINDUS INDICA* IN PHENYLHYDRAZINE INDUCED ANEMIC RATS

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Abstract: The aim of the current study is to test the anti-anemic activity of hydro-alcoholic leaf extract of *Tamarindus indica* against phenylhydrazine induced anemia in rats. Phenylhydrazine (60mg/kg) was administered intraperitoneally for two days to induce anemia in rats. The animals were divided in to four group containing six animals each. Group I served as normal control, Group II as anemic control, Group III as reference control administered with B-complex vitamin and Group IV animals were treated with 200mg/kg, of hydro-alcoholic leaf extract of *Tamarindus indica*. All the test drugs were administered once daily for twenty eight days through oral route. On twenty ninth days blood was withdrawn, through tail puncture under phenobarbitone anesthesia and subjected to the estimation of erythrocyte, hemoglobin and percentage Hematocrit. both the hydro-alcoholic leaf extract of *Tamarindus indica* and vitamin B12 significantly increased the erythrocyte, hemoglobin and Hematocrit levels that conclude that, *Tamarindus indica* leaf extract exhibits anti-anemic activity.

Keywords: Anemia, Anti-anemic activity, *Tamarindus indica* and B-complex vitamin

Introduction: Anemia is a condition that develops once blood lacks enough healthy red blood cells or hemoglobin. Anemia affects the lives of over two billion individuals globally, accounting for over 30 percentage of the world's population that is that the most typical public health problem significantly in developing

countries occurring at all stages of the life cycle [1]. *Tamarindus indica* (T. indica) is evergreen tree that 24 m height and 7 m girth that has pale yellow and pink flowers [2]. It needs dry climate so the region it is commonly seen extends Africa to Senegal in west, Sudan and Ethiopia in east, Mozambique and Madagascar in south [3]. It is also thought that the plant came to India from Africa [2,3]. Thailand, Bangladesh, Indonesia in Asia; Mexico, Costa Rica in America are some of the countries in which this plant is mostly encountered [4].

Every part of *T. indica* plant (root, body, fruit, and leaves) not only has rich nutritional value

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and broad usage area in medicine but also has industrial and economic importance. Tamarind can be the most acidic and sweet fruit according to its growing season[5]. According to World Health Organization report, tamarind fruit is an ideal source of all essential amino acids except tryptophan (82%)[6]. Its seeds also have similar properties so it becomes an important, accessible protein source especially in countries where protein malnutrition is a common problem. According to phytochemical analysis results, *T. indica* contains phenolic compounds like catenin, procyanidin B2, epicatechin, tartaric acid, mucilage, pectin, arabinose, xylose, galactose, glucose, uronic acid and triterpen[7]. Iron deficiency is that the most typical biological process disorder in become depleted and a restricted supply of iron to numerous tissues becomes apparent. This might result in depletion of hemoprotein and iron-dependent intra- cellular enzymes taking part in several metabolic pathways [8]. Therefore, there's the requirement for proper management of micronutrient deficiencies most especially irons deficiency. Over the years, medicinal plants are recognized to be of good importance to the health of people and communities. In several developing countries, herbal medicines are assuming larger importance in primary health care.

Materials and Methods:

Plant material: The plant material is made up of the leaves of tamarind tree. The leaves were collected from medicinal garden of Modern Institute of Pharmaceutical Sciences in August 2016. The sample of plant was identified and authenticated at Rajmata Vijayraje Scindia Krishi Vishwavidhalaya, faculty of Agriculture, Indore

Preparation of Extract: The collected leaves were, shade dried then ground into coarse powder. The powder was then subjected to complete extraction by a maceration method using 90% ethyl alcohol as a solvent at room temperature for seven days. The Hydro-alcoholic extract was concentrated by vacuum

distillation to dry. The collected extract was stored in desiccators and used for further pharmacological study.

Animals: Healthy adult albino rats of Wistar strain of each sex, weighing about 150-200 g were obtained from the animal house of Modern Institute of Pharmaceutical Sciences, Indore. The rats of either sex were isolated and housed in separate cages during the course of experimental period and kept them at room temperature ($24 \pm 2^\circ\text{C}$) with a 12 : 12 h light/dark cycle. The animals were fed with normal pellet diet and provided water ad libitum. All the procedures and protocols were reviewed and approved by the Institutional Animal ethics committee of MIPS, Indore.

Anti-Anemic Activity:

Induction of Anemia: Anemia was induced in rats by intraperitoneal administration of phenylhydrazine (60mg/kg) daily for two days [9]. Rats that developed anemia with Hb concentration below 13 g/dl were recruited for the study.

Treatment of the animals: The anemic rats were randomly divided into four group's six animals each. Group I was non anemic animals (normal control) received 1ml/kg of 0.1% Carboxy methyl cellulose resolution. Group II was served as anemic control and group III served as reference control, received B vitamin sweetening (1 ml/rat) and group IV animals received 200mg/kg of *Tamarindus indica* extract through oral administration, by suspending in CMC solution. The entire test drug was administered orally, once daily for twenty eight days.

On twenty ninth days, blood was collected in EDTA coated tubes, by sinus puncture under phenobarbitone (45mg/kg, ip) anaesthesia. The following parameters like, Red blood cell count (RBC), hemoglobin (Hb) and Hematocrit percentage (HCT) were evaluated in blood [10].

Statistical Analysis: Data's were expressed as mean \pm SEM. The data were analyzed by using one way analysis of variance (ANOVA)

followed by Dunnet's 't' test. P values < 0.05 were considered as significant

Results and Discussion: Anti-anemic activity of *Tamarindus indica* leaf extract on Phenylhydrazine induced hemolytic anemia in rats was studied and the results were shown on Table 1. The anti-anemic activity of *Tamarindus indica* leaf extract was assessed by determining the red blood cell count, hemoglobin and hematocrit percentage. Phenylhydrazine decreased the RBC, Hb and % HCT as compared normal control. There was significant (P<0.001) increase in RBC and Hb with both Vitamin B₁₂ and *Tamarindus indica* leaf extract against phenylhydrazine challenge. Also there was significant (P<0.01) increase in % HCT with both Vitamin B₁₂ and *Tamarindus indica* leaf extract. This shows that *Tamarindus indica* effective anti-anemic activity against phenylhydrazine induced hemolytic anemia in rats and it has comparable effect as that of the standard drug Vitamin B₁₂.

Plants are a rich source of drugs because they produce a host of bioactive molecules, most likely acts as chemical defense against predators or infectious agents.(11) phytochemical analysis revealed the presence of large chemical groups that are: alkaloids, tannins, flavonoids, polyphenols, quinones, sterols, terpenes, cardiac glycosides, saponins and leucoanthocyanins. They have antioxidant power, promote regeneration of tissue, reduce the permeability of blood capillaries and increase their resistance to hemolysis. (12) The presence of these chemicals by their properties justifies the resistance of red blood cells of treated rats with

the extract. Indeed saponins and alkaloids have shown anti-anaemic properties. (13) Alkaloid inhibits cyclic adenosine monophosphate (cAMP) phosphodiesterase thereby accumulating cAMP. This effect stimulates phosphorylation of proteins and synthesis of proteins, which improves erythropoiesis.(14) Saponins are also known to inhibit platelet aggregation and thrombosis. Saponin containing in herbs have been successfully used in the management of liver inflammation, as tonic sedative formulas, to promote and vitalize blood circulation. Since saponins are active agents which lyse the membrane of red blood cells or other wall, it is likely that red blood cells were first lysed by the plant. Then the cells have overcome this inhibition by producing a glycosidic enzyme which cleaves some of the terminal sugars from the saponin, which causes its detoxification. This detoxification of saponins has reinforced the proper use of iron contained in the aqueous extract of *Tamarindus indica* leaf allowing to synthesize heme / haemoglobin for new red blood cells, thus leading to an improvement of Hb, RBC and PCV. Saponins especially terpene glycosides enhance the natural resistance and have the recovery powers of body. Also, flavonoids have anti-anaemic potential and veinotonic properties, which protects the blood capillaries.(15) the anti-anemia potential and haemoglobin restoring effect of aqueous extract of *Tamarindus indica* leaf as suggested by the data in this study could be attributed in part to its phytochemical constituent

Table 1: Effect of *Tamarindus indica* leaf extract on Phenylhydrazine induced hemolytic anemia in rats

S.No	Group	Drug Treatment	RBC (10 ⁶ μL ⁻¹)	Hb (g dL ⁻¹)	HCT %
1	Normal Control	(0.1% CMC)	8.89±0.60	13.49±0.51	46.88
2	Anemic Control	Phenylhydrazine (60mg/kg)	4.82±0.15	6.23±0.22	28.45
3	Reference Control	Vit B ₁₂ (1ml/rat)	8.36±0.40***	13.13±0.71***	45.22**
4	Test Control	<i>Tamarindus indica</i> (200mg/kg)	8.05±0.53***	13.05±0.75***	42.71**

Data were expressed as Mean ± SEM (n=6)

*P<0.05, ** P<0.01 and *** P<0.001 Vs Anemic Control

Conclusion: The ethanolic leaf extract of *Tamarindus indica* exhibits anti-anemic activity against phenylhydrazine induced anemia in rats. The anti-anemic effect produced by the *Tamarindus indica* leaf may be due to its high content of iron which is present in the plant.

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References:

1. Ramesh C, Lopamudra P; Prevalence of Iron-Deficiency Anemia in India: Results from a Large Nationwide Survey. *Journal of Population and Social Studies*, 2010; 19(1): 59-80.
2. Bhadoriya SS, Ganeshpurkar A, Narwaria J, Rai G, Jain AP. *Tamarindus indica*: extent of explored potential. *Pharmacogn Rev* 2011; 5(9): 73-81
3. Havinga RM, Hartl A, Putscher J, Prehsler S, Buchmann C, Vogl CR. *Tamarindus indica* L. (Fabaceae): patterns of use in traditional African medicine. *J Ethnopharmacol* 2010; 127(3): 573-588
4. World Health Organization, Micronutrient Deficiencies: Prevention and Control Guidelines, World Health Organization, Geneva, Switzerland, 2015.
5. De Caluw E, Halamov K, Van Damme P. *Tamarindus indica* L.: a review of traditional uses, phytochemistry and pharmacology. *Afrika Focus* 2010; 23(1): 53-83.
6. Glew RS, Erjagt DJ, Chuang LTS, Huang Y, Millson M, Glew RH. Nutrient content of four edible wild plants from west Africa. *Plant Foods Hum Nutr* 2005; 60(4): 187-193
7. Siddhuraju P. Antioxidant activity of polyphenolic compounds extracted from defatted raw and dry heated *Tamarindus indica* seed coat. *LWT-Food Sci Technol* 2007; 40(6): 982-990.
8. [22] Paula FS, Kabeya LM, Kanashiro A, de Figueiredo ASG, Azzolini AECS, Uyemura SA, et al. Modulation of human neutrophil oxidative metabolism and degranulation by extract of *Tamarindus indica* L. fruit pulp. *Food Chem Toxicol* 2009; 47(1): 163-170
9. Roque M, D'Anna C, Gatti C, Veuthey T; Haematological and Morphological analysis of erythropoietic regenerative responses in Phenylhydrazine induced haemolytic anaemia in mice. *Scandinavian Journal of Laboratory Animal Science*, 2008; 35: 181 – 190.
10. Ghai CL; A text book of practical physiology. Jaypee Brothers, India, 1995.
11. C. Pathirana, M.J. Gibney, T.G. Taylor, *Atherosclerosis*, 1990, 36,595–599.
12. N.Singh, P. Verma, N. Mishara, R. Nath, *Indian J. Pharmacol.*, 1991,21,99. 22.
13. T.N. Saimak, MD, MPH. Editor Melissa Conrad Stoppler MD, 2009.
14. E. Gudmand-Hoyer, H. Skovbjerg, *Scand J Gastroenterol Suppl.*, 1996, 216, 111-21. Review. PMID: 8726284.
15. Joshi Ankur, Soni Priyanka, Vyas Narendra, Khan Javed, Malviya Sapna and Kharia Anil, 2017. "Anti-anemic activity of hydro-alcoholic leaf extract of *Aegle marmelos* in Phenylhydrazine induced anemic rats", *International Journal of Current Research*, 9, (04),PP48928-48931, April 2017