



## STUDY OF HERBAL EXTRACT ON HAEMATOLOGICAL PARAMETERS IN TREATED RATS

**P. H. Rohankar**

Govt. Vidarbha Institute of Science and Humanities, Amravati

rohankarp@yahoo.com.

### Abstract:

The assessment of haematological parameters in rat can be used to determine the extent of deleterious effects of a plant extract on animal blood. Oral administration of alcoholic extract of *Achyranthes aspera* leaves show significant changes in the RBCs, Haemoglobin, platelets and lymphocytes, while other parameters are not altered significantly. This data suggests that the plant extract had no adverse effect but are safe as food and is in use as herbal medicine for other treatments by tribal people.

### Introduction:

About 80% population of the world depends on the traditional system of health care (Ahmad, 1999). Traditional medicine system still plays a major role in the delivery of primary health-care. These medicines have less side effects and man can get it easily from nature (Shah and Khan, 2006). Search of an oral agent that can control and capable of normalizing biochemical and hematological indices in human is at least as old as recorded history and was in vogue in prehistoric times.

This systems usually rely on medicinal plants for treatment, which are utilized by traditional medicinal practitioners for treatment of a diverse variety of diseases, against some of which the medicine has no known cure. Over 5000 plants are known to be used for medicinal purposes all over in the world, but only a few have been described or studied (Taylor *et al.*, 2001).

Folk medicine is the most widely used form of traditional medicine in our country. Folk medicinal practitioners believe that ailments can occur through accumulation of toxins in blood, which in turn results from undigested food or improper emptying of bowels on a regular basis. The various toxins that accumulate then pass through the blood to various organs of the body and if they pass a certain threshold, the body no longer can defend itself against diseases resulting in diseases occurring from both within and without causes. This accumulation of toxins in blood and purification of blood through ingestion of medicinal plants is quite an established concept among various traditional medicinal systems and indigenous communities throughout the Indian sub-continent.

The Amravati district of Maharashtra lies in close vicinity of Satpuda Mountains. The Melghat region and Satpuda Mountains of Amravati district are known for their rich flora. The tribals of this region have been using various plants used as blood purifier. Unfortunately, the

ethanobotanical enumerations for its effect on haematological and biochemical parameters regulatory properties were not recorded for this region. Therefore, the present study is a small effort to gain insight in the knowledge of traditional medicine of this region and establishing their haematological and biochemical parameters regulatory properties

### Material and Methods:

*Achyranthes aspera* L. (Family Amaranthaceae, Order: Caryophyllales) is a common plant of the study area abundantly found in wastelands. The plant is used in indigenous system of medicine as antiarthritic, antifertility, laxative, anti-helminthic, aphrodisiac, antiviral, anti-spasmodic, antihypertensive, anticoagulant, diuretic and anti-tumor (Anonymous, 1985). This plant is astringent, digestive, diuretic, laxative, purgative and stomachic. It is reported to contain alkaloids, flavonoids, and terpenoids. Flavonoids have shown to prevent or slows the development of some cancers (Narayana *et al.*, 2001) and mostly act as an anti-oxidant and anti-inflammatory agents.

**Preparation of extract:** The leaves of above mentioned plants were collected, shade dried, powdered and subjected to Soxhlet extraction with ethanol. The extract was evaporated to near dryness on a water bath, weighed and kept at 4 °C in refrigerator until use. The presence of various plant constituents in the plant extract was determined by preliminary phytochemical screening as per Thimmaiah (2004).

**Procurement and rearing of experimental animal:** Healthy Wistar strain female albino rats of about two month old and weighing 150- 300 gm allowed to acclimatize to laboratory environment for 15 days before experimentation.

**Acute toxicity study:** Healthy female albino rats were starved for 3- 4 hr and subjected to acute toxicity studies as per OECD guidelines No: 423 (2001). For experimentation they were

divided into 2 groups of 6 animals each and kept singly in separate cages during the experiment.

Group 1: served as the control and received distilled water.

Groups 2: received the alcohol extract of *Achyranthus aspera* leaves orally at the dose of 200 mg/kg body weight in 2 ml of distilled water respectively for 21 days via gastric intubation.

After 21 days, the rats were subsequently anaesthetized and blood sample were collected into EDTA/ lithium heparin bottles.

The haematological parameters i.e. red blood cell counts (RBC), haemoglobin (Hb) concentration, The packed cell volume (PCV), white blood cell count (WBC), mean cell volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC) were determined according to the method of Dacie and Lewis (1994). Platelet (PLT) count and WBC differentials (neutrophils, monocytes, lymphocytes, basophils and eosinophils) were analyzed according to the standard technique described by Beker *et al.*, (1998) and Cheesbrough, (2000).

All the data are expressed as mean  $\pm$  S.E. Statistical analysis was done by Student's t-test (Mahajan, 1997).

### Result:

Alcohol extract of *Achyranthus aspera* leaves at the highest dose of 2000 mg/kg body weight were used for acute toxicity activity. Clinical toxicity symptoms such as respiratory distress, salivation, weight loss and change in appearance of hair as well as mortality were not observed at any period of the experiment. This suggested that short term use for this purpose is apparently safe. Hence one tenth of highest dose were selected for present investigation.

The effects of the graded doses of the alcoholic leaves extract of *Achyranthus aspera* on haematological parameters of Wistar rats are shown in Table 1. While there was no significant difference in the effects of all the doses on the WBC, MCV, MCH, MCHC, neutrophils, monocytes and basophils, some significant differences were observed in the levels of the other haematological parameters. The levels of the RBC, Hb, platelets and lymphocytes increased significantly in comparison with the control rats. There was significant increase in the level of the PCV. In addition, the level of the eosinophils was observed to increase significantly at the dose of 200 mg/kg.

**Table 1:** Effect of the ethanolic leaves extract on haematological parameters in female rats

Parameter	Control Vehicle	<i>Achyranthus aspera</i> 200 mg/kg b. w.
	<b>RBC</b>	6.87 $\pm$ 0.29
<b>Hb (gm/ dl)</b>	13.35 $\pm$ 0.35	12.01 $\pm$ 1.76 <sup>ns**</sup>
<b>Hematocrite (%)</b>	53.12 $\pm$ 0.14	37.16 $\pm$ 1.52 <sup>***</sup>
<b>MCV(cuu)</b>	51.35 $\pm$ 0.30	64.33 $\pm$ 1.11 <sup>*</sup>
<b>MCH (pg)</b>	21.42 $\pm$ 0.98	22.83 $\pm$ 0.19 <sup>ns</sup>
<b>MCHC (gm/ dl)</b>	34.27 $\pm$ 1.21	33.13 $\pm$ 0.31 <sup>**</sup>
<b>PCV (%)</b>	31.48 $\pm$ 0.26	33.74 $\pm$ 0.52 <sup>ns</sup>
<b>WBC (ml/cmm)</b>	4032 $\pm$ 0.64	5166.6 $\pm$ 14.26 <sup>***</sup>
<b>Neutrophil (%)</b>	16 $\pm$ 0.86	22 $\pm$ 0.76 <sup>***</sup>
<b>Monocyte (%)</b>	02 $\pm$ 0.70	2.08 $\pm$ 0.26 <sup>ns</sup>
<b>Lymphocyte (%)</b>	55 $\pm$ 0.33	75.6 $\pm$ 0.76 <sup>*</sup>
<b>Eosinophil (%)</b>	03 $\pm$ 0.16	2.25 $\pm$ 0.38 <sup>**</sup>
<b>Basophil (%)</b>	02 $\pm$ 0.11	02 $\pm$ 0.27 <sup>ns</sup>
<b>Platelets (lac/ cmm)</b>	8.42 $\pm$ 0.08	10.6 $\pm$ 0.81 <sup>**</sup>

Values in means + S.E. (Standard error), n=6, \*P<0.05, \*\*P<0.01, \*\*\*P<0.001, When compared with control, ns= non-significant

### Discussion:

Phytochemicals are known to perform several general and specific functions in plants and may exhibit different biochemical and pharmacological actions in different species of animals when ingested. Their actions range from cell toxicity to cell protective effect (Trease and Evans, 1996) Similar finding was reported by Uboh *et al.*, (2010) in aqueous extract of *Psidium guajava* leaves and Dabhadkar *et al.*, (2013), while working on alcoholic extract of *Plumeria rubra* in rats.

In the present work, The acute toxicity study has revealed that the oral administration of the *Achyranthus aspera* leaves extract has non-toxic effect in Wistar rats. The assessment of haematological parameters in rats can be used to determine the extent of deleterious effect of a plant extract on animal blood (Yakubu *et al.*, 2007). It is inferred from this study that the alcoholic extract of *Achyranthus aspera* have no toxic effect on the haematological parameters which include red blood cell count, haemoglobin concentration, packed cell volume, mean corpuscular volume, mean corpuscular

haemoglobin concentration, mean corpuscular haemoglobin and red cell distribution width. This showed that the oxygen carrying capacity of the red blood cells was not compromised thus; the plant may be safe as used in traditional medicine. Some plants have been reported to cause destruction of the red blood cells which leads to anaemia and ultimately cell death (Blood and Radostits, 1989; Adedapo, 2002; Adedapo *et al.*, 2004; 2007a). These indices suggest that the extract does not possess toxic substances that can cause an anemic condition in rats. This observation corroborates with the report of Ashafa, *et al.*, (2009) and Oyedemi, *et al.*, (2010). The non-significant effect of the extract on the RBC levels may be an indication that the balance between the rate of production (erythropoiesis) and destruction of blood corpuscles was not altered. MCHC and MCH relate to individual red blood cells while Hb, RBC and PCV are associated with the total population of red blood cells. Therefore, the absence of significant effect of the extract on RBC, Hb, PCV, MCH and MCHC could mean that neither the incorporation of hemoglobin into red blood cells nor the morphology and osmotic fragility of the red blood cells was altered (Adebayo *et al.*, 2005).

The administration of the extract did not alter the level of WBC and its differentials, including basophils, eosinophils and monocytes as well as platelets, throughout the experimental period, while that of lymphocytes was only increased significantly at 200 mg/kg body weight. Similar finding was observed by Adedapo, *et al.*, (2008), while working on aqueous extract of *Acacia karroo* stem bark in rats and mice. Report about WBC counts and its differentials have pointed out that increased count of lymphocytes is supposed to be helpful in boosting immune system (Adedapo *et al.*, 2007; Mohajeri *et al.*, 2007). The platelets affect the viscosity of blood, which is correlated positively to blood pressure (Adedapo *et al.*, 2007) but, the alcoholic leaves extract of different plants does not produce any observable change in the count of blood platelets.

Mishra and Tandon (2012) studied treatment of mice with crude extract of *Hibiscus rosasinensis* flowers (500 mg/kg BW) and *Bougainvillea spectabilis* leaves (800 mg/kg BW) for a period of 30 days indicates a significant increase in the level of haemoglobin and count of RBC but a significant decline in the level of MCH and MCV in the former case. On the other hand, in *B. spectabilis* treated animals, the level of hemoglobin, RBC count and PCV declined significantly. Alterations in blood parameters

may be due to changes in cellular integrity, membrane permeability of cells or even due to exposure to toxic chemicals (Mishra and Tandon, 2012)

#### Conclusion:

This study has shown that administration of *Achyranthus aspera* alcoholic leaves extract appears to be relatively non-toxic to animals. Thus, this study may prove to be an effective and safe alternative remedy in haematological related disorders. Work is in progress on the isolation and characterization of the actual mechanism of action of the crude extract and bioactive agents.

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