

INFLUENCE OF AN AQUEOUS FRUIT EXTRACT OF *SOLANUM TORVUM* SWARTZ ON PHENYLHYDRAZINE INDUCED HEMOLYTIC ANEMIA IN WISTAR RATS

Gnangoran Boua, N. *, Coulibaly Yédiotiaire, S., N'dia Kouadio, F., Yapo Angoué, P.
Laboratory of Physiology, Pharmacology and Pharmacopeia; UFR-SN, University Nangui Abrogoua; 02
LP 801 Abidjan 02 (Côte-d'Ivoire).

*boua_gn@yahoo.fr

Abstract

Solanum torvum (Solanaceae) is a medicinal plant used in Côte d'Ivoire to treat anemia. The objective of this study is to evaluate the therapeutic efficacy of a total aqueous fruit extract of *S. torvum* in the treatment of anemia. Thus, Hemolytic anemia was induced using intraperitoneal injection of phenylhydrazine (PHZ, 40 mg/kg b.w.) for two successive days (D₀ and D₁) to rats in three batches of six each. From D₂ to D₂₀, these animals received orally distilled water (positive control), 250 mg/kg body weight (b.w.) of a total fruit aqueous extract of *S. torvum* (TAS_t) and Hemafer® (reference anti-anemia) at 5 mg/kg b.w. Six rats of another batch (negative control) received NaCl 9 ‰ for two successive days (D₀ and D₁) and distilled water (10 mL/kg) from D₂ to D₂₀. Blood samples were taken from the retro-orbital sinus of the eye in all rats on days D₀, D₂, D₈, D₁₄ and D₂₀ for erythrocyte parameters determination. The results revealed that oral administration of TAS_t at 250 mg/kg b.w. and Hemafer® 5 mg/kg b.w. in rats pretreated with phenylhydrazine did not cause any significant change in body weight. On the other hand, TAS_t and Hemafer® quickly normalized the level of erythrocytes, the hemoglobin concentration, the hematocrit level, the Mean Corpuscular Volume (MCV), the Mean Corpuscular Hemoglobin Concentration (MCHC) and the Mean Corpuscular Hemoglobin (MCH) deteriorated by PHZ compared to the rats of the positive control group. In conclusion, this study showed that the fruits of *Solanum torvum* have properties similar to that of Hemafer® in rats, which could justify the traditional use of this plant in the treatment of anemia.

Keywords: Anemia, erythrocyte parameters, phenylhydrazine, *Solanum torvum*

Introduction

Anemia is a real public health problem. It mainly affects pregnant women and young children. This pathology has major drawbacks on the health which are stunted growth, disruption of the child's mental, cognitive development and productivity reduction at work (1). In addition, anemia is associated with an increased risk of morbidity and mortality in pregnant women and young children (2). Worldwide, approximately 35% of the population suffers from anemia (3). The most affected regions are South Asia, Central and West Africa. In central and west Africa, the prevalence of anemia is estimated to 80% in children, 52% in non-pregnant women and 61% in pregnant women (4). In Côte d'Ivoire, the study done by (5) in three areas of Abidjan (Abobo, Cocody and Yopougon) showed that the prevalence of anemia in school children aged 5 to 11 years is 14.2% in boys and 16.1% in girls. Another research work done by (6) in, also, three on for populations' health care.

In Côte d'Ivoire, among the many medicinal plants used to treat anemia, *S. torvum* was chosen for this study because it is frequently used by the population. But, there is no scientific proof to support this claim. The objective of this work is to study the potential of the fruit aqueous extract

Methods

Plant

The fruits of *Solanum torvum* (Solanaceae) were harvested in August 2018 in Progréagui, a town located at 15 kilometers from Méagui (Côte d'Ivoire). They were identified and authenticated by botanists of Nangui Abrogoua University (Abidjan, Côte d'Ivoire).

Animals

24 albino rats (12 females and 12 males) weighing between 119 and 196 g, of the species *Rattus norvegicus* were used. These rats were 10 to 12 weeks old and are from the Laboratory of Physiology, Pharmacology and Pharmacopoeia of Nangui Abrogoua University. They had free access to tap water and food (granules) and were acclimated in cages for 14 days in a room with temperature of 25 ± 2 °C and a photoperiod of 12h (12 hours of light and 12 hours of darkness)

according to the guidelines of the Laboratory of Physiology, Pharmacology and Pharmacopoeia. Phenylhydrazine which reduce the morphology of the red blood cell and its function nction (7) (9).

The prevention and treatment of hemolytic anemia in pregnant women and young children is of a major importance. Treatments based on the use of immunosuppressants or corticosteroids, erythropoietin injections, blood transfusion or even bone marrow transplantation are expensive and often have disturbing side effects (10). Thus, traditional medicine can be an alternative solution for populations' health care.

In Côte d'Ivoire, among the many medicinal plants used to treat anemia, *S. torvum* was chosen for this study because it is frequently used by the population. But, there is no scientific proof to support this claim. The objective of this work is to

Preparation of the fruit total aqueous extract of *Solanum torvum*

The fruit total aqueous extract of *Solanum torvum* (Solanaceae) was prepared according to a method described by (11).

The fruits of *Solanum torvum* were, first, washed with distilled water and crushed using a mortar and pestle. After drying under air conditioning at 15 °C for five days, they were finely pulverized using an electric grinder (Culati, France). 500 g of the *S. torvum* fruit powder was boiled at 100 °C for 15 minutes in 1000 mL of distilled water. The decocted product obtained was filtered through cotton wool and then through Whatman No.1 filter paper. A volume of 500 mL of hot distilled water was added to the residue and boiled, a second time, at 100 °C for 10 minutes. This solution was also filtered. The filtrates were evaporated and dried at 45 °C using an oven (Friucell, Germany) for 48 hours. 117.78 g of powder was obtained, that correspond to a yield of 23.56%. The powder of the total aqueous fruit extract of *Solanum torvum* (TAS_t) was stored in the refrigerator at 7 °C until use

Experimental design

Induction of hemolytic anemia

The method used in this study was that described by (12) (13). Thus, Anemia was induced in rats using phenylhydrazine (PHZ) dissolved in distilled water and administered to rats

intraperitoneally at a dose of 40 mg/kg b.w. for two successive days (D_0 and D_1).

Treatment of the rats

24 rats were divided into four groups of six rats each with three females and three males. The different groups of rats received daily the following treatments:

Group 1 (normal control) received NaCl 9‰ for two successive days (D_0 and D_1) and distilled water (10 mL/kg) from D_2 to D_{20} by oral route;

Group 2 (positive control) were intraperitoneally administered with PHZ (40 mg/kg b.w.) for the first two days (D_0 and D_1) and distilled water (10 mL/kg b.w.) orally from D_2 to D_{20} ;

Group 3 (Hemafer®) were intraperitoneally administered with PHZ (40 mg/kg b.w.) for the first two days (D_0 and D_1) and before the oral administration of the standard solution (Hemafer®) (5 mg/kg b.w.) from D_2 to D_{20} ;

Group 4 (TASt) received by intraperitoneal route 40 mg/kg b.w. of PHZ the first two days (D_0 and D_1) and the aqueous fruit extract of *Solanum torvum* (250 mg/kg b.w.) orally from D_2 to D_{20} .

Blood collection

The rats were fasted for ten hours and anesthetized with an overdose of ether. Venous blood samples were, then, taken from the retro orbital sinus of the eye of each rat using a sterile Pasteur pipette according to the technique described by (14). The collected blood was put into tubes containing EDTA. The dosage of the various parameters was carried out on days D_0 , D_2 , D_8 , D_{14} and D_{20} . The blood samples were used for the determination of hematological parameters such as red blood cells (GR), hemoglobin level (HGB), hematocrit (Hte), Mean Corpuscular Volume (MCV), Mean Corpuscular Hemoglobin Concentration (MCHC) and Mean Corpuscular Hemoglobin (MCH) using an automatic counter (Sysmex XP 300, France) as described by (15).

Data analysis

The statistical data analysis and graphs were performed using Graph Pad Prism 5.01 software (San Diego, California, USA). The results were expressed as an average followed by the standard error on the mean ($M \pm SEM$). One-way analysis of variance (ANOVA1) was carried out with Tukey's post-hoc test by determining the significant differences between the different parameters in

groups of rats. The significance threshold was set at $p < 0.05$.

Results

Effect of the total aqueous fruit extract of *Solanum torvum* on the rats' body weight

The treatment of the rats with phenylhydrazine (PHZ) induced a non-significant decrease ($p > 0.05$) of their body weight compared to normal control rats during 20 days of experimentation. In addition, the results show that the body weight of rats receiving PHZ and treated with either the total aqueous fruits extract of *S. torvum* at 250 mg / kg b.w. or Hemafer® at 5 mg/kg b.w. increased but not significantly ($p > 0.05$) compared to the group of rats having received only PHZ (Figure 1).

Effect of the total aqueous fruit extract of *Solanum torvum* on the rats' erythrocyte parameters

Erythrocyte levels

Figure 2 shows the influence of the total aqueous fruit extract of 250 mg/kg b.w. of *S. torvum* and 5 mg/kg b.w. of Hemafer® on the erythrocytes level in rats. Phenylhydrazine (PHZ) administered to rats by intraperitoneal route induces a significant reduction ($p < 0.01-0.001$) on D_2 , D_8 , D_{14} and D_{20} in the number of erythrocytes compared to the normal control rats' group. On the other hand, the treatment of groups of rats having received PHZ with 250 mg/kg b.w. of TASt and 5 mg/kg b.w. of Hemafer® leads to a significant increase ($p < 0.05-0.001$) in the number of erythrocytes on D_8 and D_{14} compared to the group of rats treated with PHZ only. On D_{20} , the results do not reveal any significant variation ($p > 0.05$) of the erythrocytes level compared to the group of rats having received PHZ only.

Hemoglobin concentration

The PHZ induces a significant reduction ($p < 0.05-0.001$) in the concentration of rats' hemoglobin on D_2 , D_8 and D_{14} when compared to that of the normal control group. However, no significant variation of this parameter was observed at D_{20} . The rats pretreated with PHZ before the gavage of 250 mg/kg b.w. of TASt or 5 mg/kg b.w. of Hemafer® show a significant increase ($p < 0.01-0.001$) in their hemoglobin concentration on D_8 and D_{14} compared to PHZ treated rats' group. On D_{20} , no change ($p > 0.05$) was observed in the hemoglobin

concentration between the rats post-treated with TAST and Hemafer® compared to the group of rats which received only PHZ (Figure 3).Text

Hematocrit level

PHZ administered to rats significantly reduced ($p < 0.001$) the hematocrit level only on D_2 ($p < 0.001$) and D_{20} ($p < 0.05$) compared to normal control rats' group. The administration of 250 mg/kg b.w. of TAST and 5 mg/kg b.w. of Hemafer® in rats pre-treated with PHZ induced no significant change ($p > 0.05$) in the hematocrit level on D_8 , D_{14} and D_{20} compared to the rats' group treated with PHZ (Figure 4).

Mean Corpuscular Volume (MCV)

PHZ induces a significant increase of MCV in the rats ($p < 0.01-0.001$) on D_8 , D_{14} and D_{20} compared to the control group rats. However, no variation in the MCV was observed on D_2 . The extract and 5 mg/kg b.w. of Hemafer® administered to rats induce a significant decrease in this parameter ($p < 0.01-0.001$) on D_8 followed by a significant increase ($p < 0.05-0.01$) on D_{14} compared to rats treated with PHZ only. No significant change in MCV was noticed, on D_{20} , in PHZ-pretreated rats with 250 mg/kg b.w. of TAST and 5 mg/kg b.w. of Hemafer® compared to the group of rats treated with PHZ only (Figure 5).

Mean Corpuscular Hemoglobin Concentration (MCHC)

The results reveal a significant increase ($p < 0.001$) in MCHC on D_2 and a significant reduction ($p < 0.001$) on D_8 in the group of rats only treated with PHZ compared to the control rats' group. However, the PHZ had no significant influence ($p > 0.05$) on this parameter on D_{14} and D_{20} . The treatment of rats with the extract (250 mg/kg b.w.) and Hemafer® (5 mg/kg b.w.) induced a significant increase in MCHC on D_8 ($p < 0.001$) and D_{14} ($p < 0.05$) compared to the group of rats treated with PHZ only. On D_{20} , no significant variation ($p > 0.05$) was observed in rats treated with 250 mg/kg b.w. TAST and Hemafer® compared to those who received PHZ only (Figure 6).

Mean Corpuscular Hemoglobin (MCH)

The intraperitoneal administration of the PHZ to rats increased significantly ($p < 0.01-0.001$) the MCH from D_2 to D_{14} compared to control rats. However, PHZ had no significant effect ($p > 0.05$) in

MCH on D_{20} . The extract does not cause any significant influence ($p > 0.05$) on the MCH of the pretreated rats' group at all studied periods. So does Hemafer. However, on D_{14} , the extract and Hemafer® induced a significant increase ($p < 0.05-0.001$) in this parameter compared to rats having received PHZ (Figure 7).

Discussion

The results showed that the treatment of rats with a total aqueous fruit extract of *Solanum torvum* (TAST) at 250 mg/kg b.w. did not significantly increase the body weight of the rats compared to the rats which received phenylhydrazine. In this study, the effect of TAST is relatively similar to that of Hemafer® (5 mg kg b.w.), a standard anti-anemic drug. Therefore, TAST would have no effect on food consumption and the amount of food absorbed. The results indicated that this extract did not promote appetite in anemic rats. Therefore, TAST would not influence the weight gain of anemic rats. Other studies have also shown no influence on the body weight gain of anemic rats after oral administration of the aqueous extract of the leaves of *Jatropha tanjorensis* at a dose of 200 mg/kg b.w. for 21 days (16). However, our results are, contrary to those of (17). In fact, these authors showed that PHZ- anemic rats treated with an ethanolic leaves extract of *Moringa oleifera* (300 and 600 mg/kg b.w. for 21 days) induced a significant increase in body weight compared to rats only treated with phenylhydrazine. Regarding the erythrocyte parameters, the results revealed that the PHZ induced in the rats a significant decrease in the level of erythrocytes, in hemoglobin concentration, in hematocrit level on day 2 (D_2). However, significant increases in MCHC and MCH are noted compared to control rats. This reduction would confirm induction of hemolytic anemia in rats. From day 2 (D_2) to day 20 (D_{20}), the effects of the Phenylhydrazine (PHZ) in the rats were gradually reduced over time. This observation is thought to be related to the acute effect of PHZ (18) (19). These authors noted the reversibility of PHZ-induced anemia after discontinuation of the administration.

From D_8 , oral treatment of anemic rats with 250 mg/kg b.w. of the total aqueous fruit extract of *Solanum torvum* (TAST) resulted in a significant increase in the level of erythrocytes and the concentration of hemoglobin, but the extract does

not significantly affect the hematocrit level compared to rats treated only with PHZ. In addition, the effect of TAsT is substantially similar to that of the standard antianemic drug used in this study (Hemafer®) during the different treatment periods. The extract (TAsT, 250 mg/kg b.w.) and Hemafer® (5 mg/kg b.w.) improve the disturbances occurred by the PHZ in rats. These results would indicate that the total aqueous fruit extract of *Solanum torvum* and Hemafer® would have stimulated hematopoiesis via erythropoietin. Erythropoietin is a hormone which regulate the production of red blood cells. It increased the number of sensitive erythroblasts in the bone marrow that are converted to reticulocytes and later to mature erythrocytes (20). Similar results were obtained after treatment of anemic rats with oral administration of an aqueous extract of the sheath of *Sorghum bicolor* at doses of 200 and 300 mg/kg b.w. (21) and that of the leaves of *Trema guineensis* at 200 mg/kg b.w. (13). In addition, the total aqueous fruit extract of *S. torvum* resulted in fluctuating values of MCV, MCHC and MCH deteriorated by PHZ.

However, the values obtained, in this study, are within the limits of the references (22). ETAsT induced a normalization of MCH, MCHC and MCV. It means that TAsT normalizes the size of the erythrocytes, the charge and the hemoglobin content. Our results are different from those of (19). They revealed in their work a significant decrease in MCHC and MCV during different periods in PHZ anemic rats treated with 300 mg/kg b.w. of an aqueous extract of the chalice of *Hibiscus sabdarifa* for 15 days.

During this study, TAsT was efficient against anemia induced by phenylhydrazine. This result revealed that the total aqueous extract of the fruits of *Solanum torvum* contains bioactive molecules capable of repairing the damages caused by PHZ. Previous work has revealed the presence of minerals, proteins, fats, carbohydrates and vitamins in the fruits of *Solanum torvum* (23). Other researchers have also mentioned the presence of steroids, saponins, alkaloids, flavonoids, tannins and glycosides in the fruits of this plant (24) (25). In fact, amino acids, proteins, vitamins B, E and iron stimulate erythropoiesis which is involved in the synthesis of hemoglobin, the formation and

maturation of red blood cells (26). In addition, tannins, saponins, flavonoids and alkaloids have anti-anemic properties: They promote tissue regeneration, reduce the permeability of blood capillaries and increase their resistance to hemolysis (27). The effect of extract of the fruits of *Solanum torvum* on the hemolytic anemia induced by phenylhydrazine in rats could be partly attributed to its phytochemical constituents.

Conclusion

The intraperitoneal injection of phenylhydrazine in rats caused alteration of erythrocyte parameters by inducing hemolytic anemia. The treatment of rats with an oral administration of the total aqueous fruit extract of *Solanum torvum* at the dose of 250 mg/kg b.w. or Hemafer® (5 mg/kg b.w) quickly normalize the erythrocyte indices disturbed by phenylhydrazine. In addition, the effect of the extract is practically similar to that of the anti-anemic standard drug. This study confirms and validates the traditional therapeutic use of *Solanum torvum* fruits in the treatment of anemia.

Acknowledgement

The authors are thankful to Dr. OUSSOU N'Guessan Jean-Baptiste (Lecturer and Researcher at the Nangui Abrogoua University's Laboratory of physiology, pharmacology and pharmacopoeia, Côte d'Ivoire) for his invaluable help in translating this manuscript and also, all the staff of the laboratory for their encouragement during these investigations.

Compliance with ethical standards

Competing Interests. The authors state no competing interests

References

1. UNICEF/UNU/WHO, 2001. Iron deficiency anaemia: assessment, prevention, and control. Genève, Organisation mondiale de la Santé, (WHO/NHD/01.3. Available at: http://www.who.int/nut/documents/ida_assessment_prevention_control.pdf.
2. Oski FA. Iron deficiency in infancy and childhood. N Engl J Med. 1993; 329: 190-193.
3. McLean E, Cogswell M, Egli I. Worldwide prevalence of anaemia, WHO Vitamin and Mineral Nutrition Information System, 1993-2005. Public Health Nutr. 2009; 12: 444-454.

4. Stevens G, Finucane M, De-Regil L et al. Nutrition impact model study group (Anaemia). Global, regional, and national trends in haemoglobin concentration and prevalence of total and severe anaemia in children and pregnant and non-pregnant women for 1995-2011: A systematic analysis of population-representative data. *Lancet Glob Health* 2013; 1:16-25.
5. Kokoré BA, Bleyere, MN, Ehile EE, Yapo PA. L'anémie nutritionnelle chez les écoliers de trois communes du district d'Abidjan en Côte d'Ivoire. *Antropo*. 2013; 29: 49-56.
6. Bléyé NM, Kouadio HJ, Koné M, Sawadogo D, Yapo AP. Comparison during pregnancy of iron metabolism between adolescent and adult women in Côte d'Ivoire. *Appl Sci Reports* 2014; 1 (1): 16-23.
7. Asobayire SF, Adou P, Davidsson I, Cook DJ, Hurrell RF. Prevalence of iron deficiency with and without concurrent anemia in population groups with high prevalence of malaria and other infections: a study in Côte d'Ivoire. *Am J Clin Nutr*. 2001; 74: 776-782.
8. Kalenga MK, Nyembo MK, Nshimba M, Foidart JM. Anémie associée au paludisme et aux helminthiases intestinales à Lubumbashi. *Santé pub*. 2003; 15: 413-421.
9. Idu M, Igbafe G, Erhabor J. Anti-anaemic activity of *Jatropha tanjorensis* Ellis & Saroja in Rabbits. *J Med Pl Stud* 2014; 2 (1) : 64-72.
10. Movaffaghi Z, Hasanpoor M. Effect of therapeutic touch on blood hemoglobin and hematocrit level. *J Holist Nurs*. 2006; 24:41-48.
11. Zirihi GN, Kra AKM, Guédé-Guina F. Evaluation de l'activité Antifongique de *Microglossa pyrifolia* (Lamarck) O. Kuntze (Asteraceae) « PYMI » sur la croissance in vitro de *Candida albicans*. *Rev Méd Pharm. Afr*. 2003; 17: 11-18.
12. Naughton BA, Moore E, Bush ME, Rabbit DM, Domfest B. Hemostatic alterations associated with phenylhydrazine-induced haemolytic anemia. *Eur J Clin Invest*. 1995; 25, 722-727.
13. Droucoula GC, Kouakou SL, Miezian BAP, Tchimon DP, Yapi HF. Antianemic properties of *Trema guineensis* aqueous and ethanolic extracts. *Eur J Biotechnol Biosci*. 2017; 5: 20-24.
14. Kraus AL, Baker HJ, Lindsey JR, Weisbroth SH. Research methodology-the laboratory rat. Academic Press 1980; 1:5-19.
15. Smith CN, Neptan DA, Irons RD. Effect of sampling site and collection method on variations in baseline clinical pathology parameters in Fisher-344 rat: II. *Clin. hematol. fundam. appl. toxic*. 1986; 7: 658-663.
16. Ameloko UJ. Anti-anaemic effect of aqueous and methanolic leaf extracts of *Jatropha tanjorensis* on phenylhydrazine induced haemolytic anaemic rats. Thesis, department of biochemistry, faculty of science, Ahmadu Bello University (Zaria, Nigeria) 2015; 119p.
17. Nku-Ekpang, O-AT, Nwaehujor CO, Ofem OE, Ezekiel JI. Effect of Moringa oleifera Lam. Ethanol Leaf Extract on Hematology in Phenylhydrazine-induced Anemic Albino Wistar Rats. *American J Pharm Sci*. 2015; (3) : 67-73.
18. Redondo PA, Alvarez, AI, Diez C. Physiological response to experimentally induced anemia in rats: a comparative study. *Laboratory Anim Sci* 1995; 45(5): 578-583.
19. Sènou M, Tchogou AP, Dougnon TV et al. Efficiency of *Sorghum bicolor* extract in the treatment of induced anemia on Wistar rats. *Int J Biosci*. 2016 ; 8(4) :62-71.
20. Sanchez- Elsner JR, Ramirez F, Rodriguez-Sanz E et al. A cross talk between hypoxia and TGF-beta orchestrates erythropoietin gene regulation through SPI and Smads. *J Mol Bio* 2004; 36 (1): 9-24.
21. Ogwumike OO. Hemopoietic effect of aqueous extract of the leaf sheath of *Sorghum bicolor* in albinos rats. *Afr J Biomed Res* 2002 ; 5: 69-71.
22. Descat F. Hématologie du rat : hémogramme et myélogramme. Thèse médecine vétérinaire de Toulouse, Tou 3-4011, France 2002 ; p 91
23. Leung WTW, Butrun RR, Chang FH. Food composition table for use in East Asia, Depart. health, educ. and welfare, Bethesda, United states 1972; 1: 334-344.

24. George K, Patrick A, Terrick A. Immunomodulatory and erythropoietic effects of aqueous extract of the fruits of *Solanum torvum* Swartz (Solanaceae). *Pharmacognosy Res* 2011; 3(2): 130-134
25. Fang A, Shulin W, Danhua Y, Yuewen G, Shuhua W. Attenuation of oxidative stress of erythrocytes by plant-derived flavonoids, orientin and luteolin, Hindawi. *Publ. corpor. Evidence based CAM* 2016; 1: 8-20.
26. Mathé G, Boivin P, Caen J, Turpin F, Griscelli C, Florentin I, Subtil E. Sang tissus hématopoiétique et lymphoïdes et leurs relations avec l'immunité in: *Sémiologie médicale* Eds Flammarion Médecine-Sciences, Paris; France, 1981; 63-80.
27. Singh N, Verma P, Mishara N, Nath R. A comparative evaluation of some antistress agents of plant origin. *Indian J Pharmacol.* 1991; 21:99.

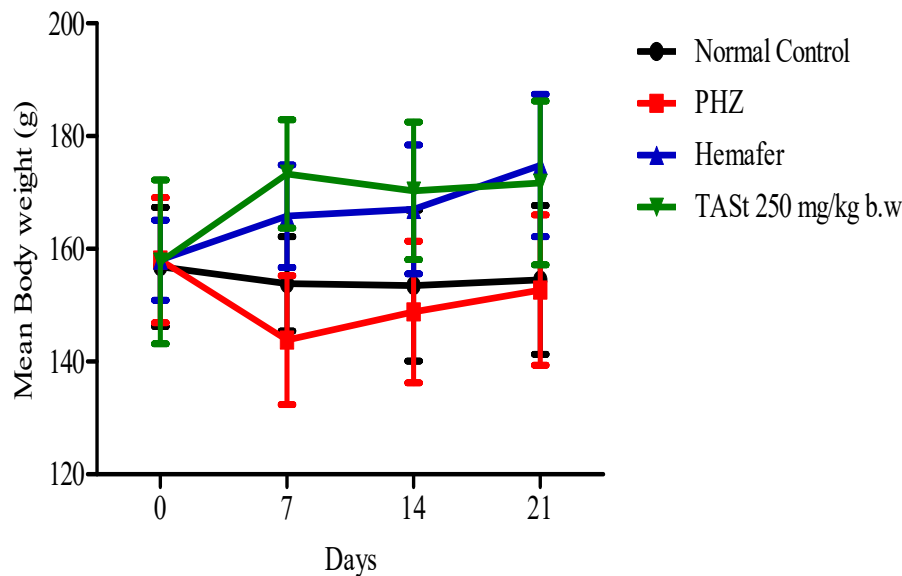


Figure 1 : Treatment effect on body weight

Values expressed as mean \pm ESM with n=6 in each group. Normal control: Distilled water; PHZ: Phenylhydrazine 40 mg/kg; Hemafer: Phenylhydrazine 40 mg/kg + Hemafer; TAST 250 mg/kg: Phenylhydrazine 40 mg/kg + TAST 250 mg / kg. TAST : Total aqueous fruit extract of *Solanum torvum*

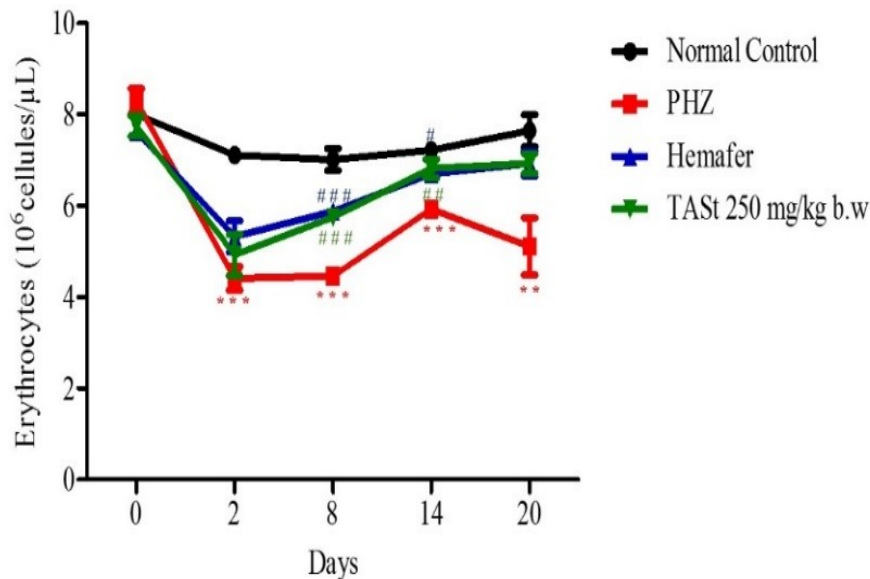


Figure 2 : Treatment effect on erythrocytes cells number

Values expressed as mean \pm ESM with n=6 in each group. **P<0.01, ***P<0.001 compared to Normal control; #P<0.05, ##P<0.01, ###P<0.001 compared to PHZ. Normal control: Distilled water; PHZ: Phenylhydrazine 40 mg/kg; Hemafer: Phenylhydrazine 40 mg/kg + Hemafer; TAST 250 mg/kg: Phenylhydrazine 40 mg/kg + TAST 250 mg / kg. TAST : Total aqueous fruit extract of *Solanum torvum*

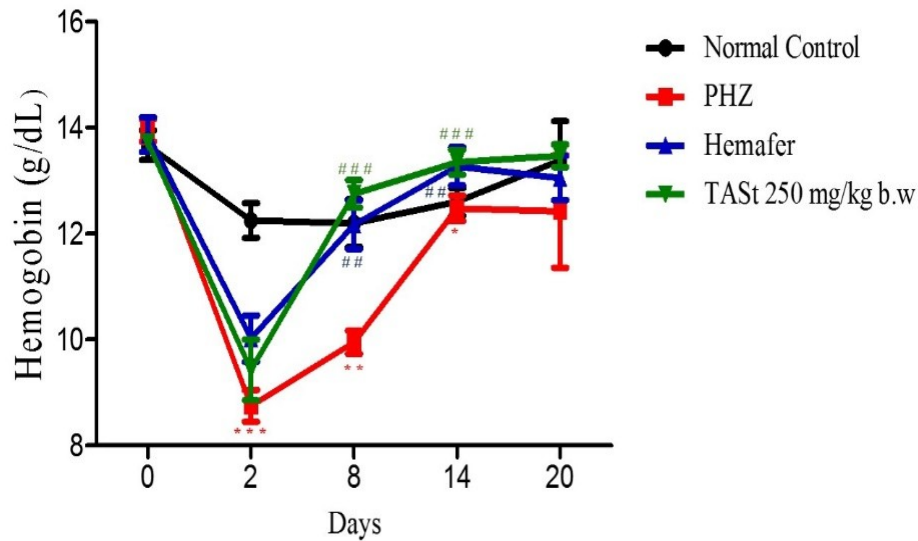


Figure 3 : Treatment effect on hemoglobin concentration

Values expressed as mean \pm ESM with n=6 in each group. **P<0.01, ***P<0.001 compared to Normal control; ##P<0.01, ###P<0.001 compared to PHZ ; Normal control: Distilled water; PHZ: Phenylhydrazine 40 mg/kg; Hemafer: Phenylhydrazine 40 mg/kg + Hemafer; TAST 250 mg/kg: Phenylhydrazine 40 mg/kg + TAST 250 mg / kg. TAST : Total aqueous fruit extract of *Solanum torvum*

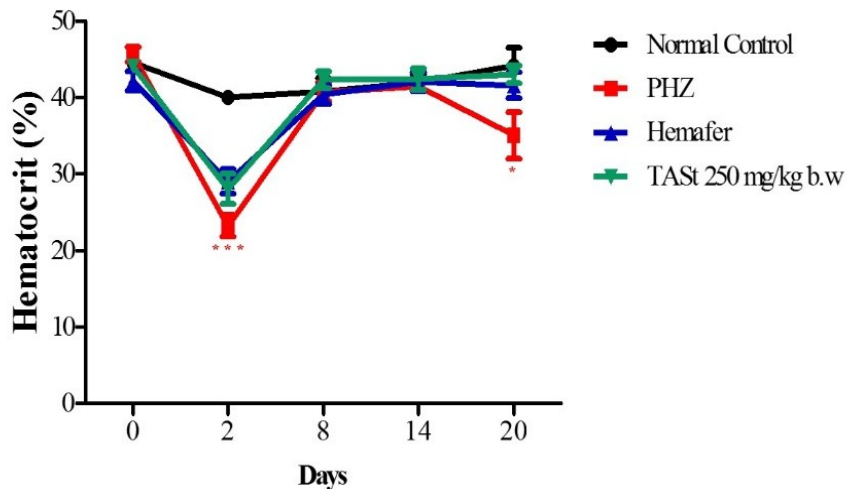


Figure 4 : Treatment effect on hematocrit.

Values expressed as mean \pm ESM with n=6 in each group. *P<0.05, ***P<0.001 compared to Normal control. Normal control: Distilled water; PHZ: Phenylhydrazine 40 mg/kg; Hemafer: Phenylhydrazine 40 mg/kg + Hemafer; TAST 250 mg/kg: Phenylhydrazine 40 mg/kg + TAST 250 mg / kg. TAST : Total aqueous fruit extract of *Solanum torvum*

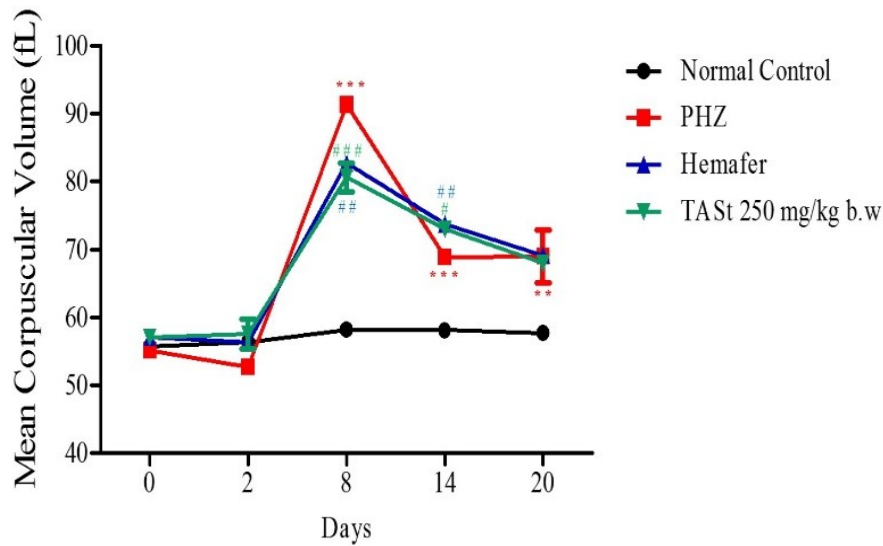


Figure 5 : Treatment effect on mean corpuscular volume

Values expressed as mean \pm ESM with n=6 in each group. **P<0.001, ***P<0.001 PHZ compared to Normal control ; #P<0.05, ##P<0.01 compared to PHZ

Normal control: Distilled water; PHZ: Phenylhydrazine 40 mg/kg; Hemafer: Phenylhydrazine 40 mg/kg + Hemafer; TAST 250 mg/kg: Phenylhydrazine 40 mg/kg + TAST 250 mg / kg. TAST : Total aqueous fruit extract of *Solanum torvum*

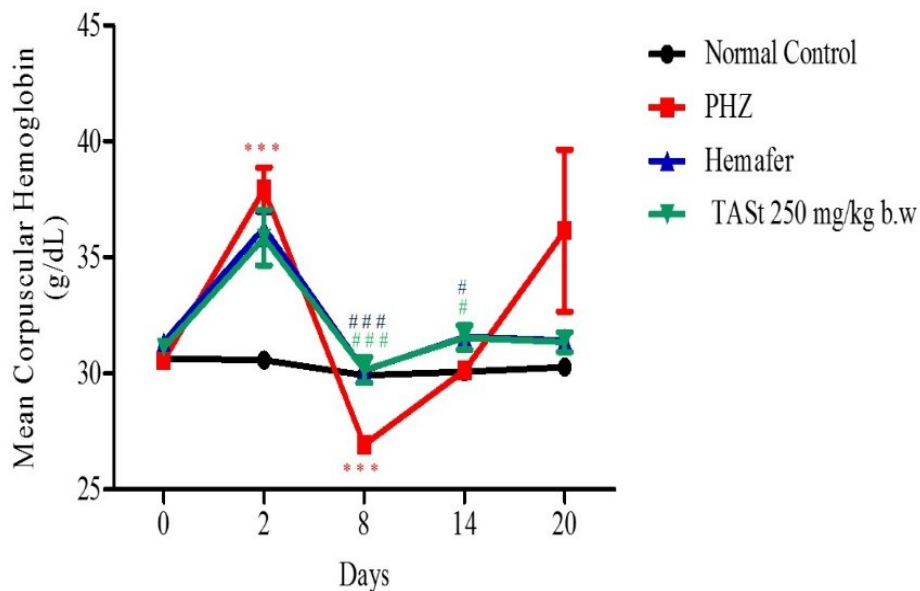


Figure 6 : Treatment effect on mean corpuscular hemoglobin

Values expressed as mean \pm ESM with n=6 in each group. ***P<0.001 compared with Normal control ; #P<0.05, ###P<0.001 compared with PHZ.

Normal control: Distilled water; PHZ: Phenylhydrazine 40 mg/kg; Hemafer: Phenylhydrazine 40 mg/kg + Hemafer; TAST 250 mg/kg: Phenylhydrazine 40 mg/kg + TAST 250 mg / kg. TAST : Total aqueous fruit extract of *Solanum torvum*

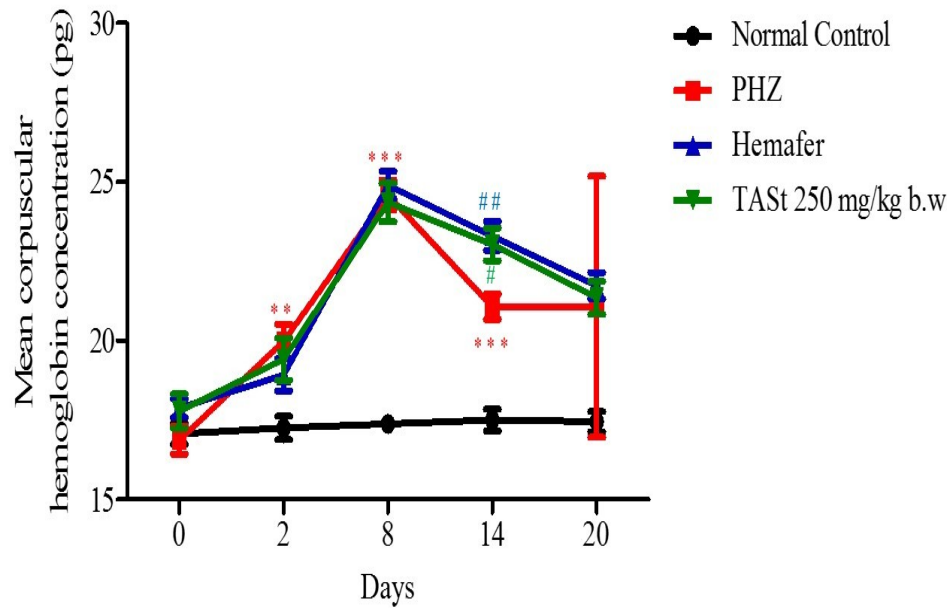


Figure 7 : Treatment effect on mean corpuscular hemoglobin concentration

Values expressed as mean \pm ESM with n=6 in each group. **P<0.001, ***P<0.001 PHZ compared to Normal control ; #P<0.05, ##P<0.01 compared with PHZ

Normal control: Distilled water; PHZ: Phenylhydrazine 40 mg/kg; Hemafer: Phenylhydrazine 40 mg/kg + Hemafer; TAST 250 mg/kg: Phenylhydrazine 40 mg/kg + TAST 250 mg / kg. TAST : Total aqueous fruit extract of *Solanum torvum*