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(RESEARCH ARTICLE)

Hematological evaluation of aqueous and methanolic leaf extracts of *Thaumatococcus daniellii* and *Alchornea cordifolia* in Wistar rats

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Abstract

The study was conducted to evaluate the hematological indices of wistar rats administered with aqueous and methanolic leaf extracts of *T. daniellii* and A. *cordifolia*. The work was separated into two portions categorized as Study A and B. A total of twenty five (25) adult male wistar rats were used for both studies. Meanwhile, each study consisted of two distinct groups of five rats each. A common control (Group I) of five rats which was administered with 2ml/kg distilled water orally was established for both studies. Groups II and III of Study A were administered with 200mg/kg b.w aqueous and methanolic leaf extracts of *T. daniellii* orally respectively, while Groups II and III of Study B were administered with 200mg/kg b.w aqueous and methanolic leaf extracts of *A. cordifolia* orally respectively. Animals were sacrificed after 14days of treatment and blood samples collected in EDTA containers were subjected to hematological analysis in accordance with standard procedures. Observations on Study A, showed that there was a significant reduction in the Hemoglobin concentration (Hb) (13.7±0.01g/l), Red Blood Cell (RBC) (62.20±0.27×106/µL) and packed cell volume (PCV) (38.75±0.02%) in group III of study A administered with 200mg/kg b.w methanolic leaf extract of *T. daniellii* compared to the control Hb (17.13±0.01g/dl), RBC (69.83×106/µL) and PCV (48.75±0.02%) respectively. However, the White Blood Cell was significantly high (10.13±0.02×103/µL) in group III compared to the control (7.13±0.02×103/µL). For Study B, administration of 200mg/kg b.w aqueous and methanolic leaf extracts of *A. cordifolia* caused a significant reduction in Hb (12.5±0.02g/dl) and (12.09±0.02), RBC (61.92±0.06×106/µL) and (61.42±0.31×106/µL) and PCV (41.15±0.02%), (42.33±0.15%) respectively compared to the control Hb (17.13±0.01g/dl), RBC (69.83±0.01×106/µL) and PCV (48.75±0.02%). On the contrary, WBC was significantly high in groups II and III (9.54±0.02×103/µL) and (9.07±0.06×103/µL) respectively compared to the control (7.13±0.02×103/µL). In conclusion, preliminary drug discovery approach on *T. daniellii* leaf should prefer aqueous extraction so as to avoid components with possible anemia inducing capacity. However, the usage *A. cordifolia* extract as therapy should be complemented with an anti-anemic medication.

Keywords: *Thaumatococcus daniellii*; *Alchornea cordifolia;* Anemia

1. Introduction

The persistent rise in the dependence on plant derived therapies as a source of primary health care for the ever increasing population of the world has been acknowledged and reported by World Health Organization [1]. *Alchornea cordifolia* a perennial evergreen shrub measuring up to 4-8cm high with erected shoots belongs to the family

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Euphorbiacea and commonly known as the Christmas Bush. It is mostly found in the marshy areas along the coastal regions of West Africa. *A. cordifolia* has been extensively used in tackling numerous human diseases in the African folk traditional medicine. These include diarrhea, bacterial as well as fungal infections etc [2].

Thaumatococcus daniellii also known as the sweet prayers plant or katemfe belonging to the family Marantaceae is a rhizomatous, perennial monocotyledonous herb which grows and thrives across the hot, humid, tropical rain forest and coastal zones of West Africa [3]. The height of a mature *T. daniellii* plant is about 3-4cm, with some large papery leaves of about 46cm long bearing pale purple flowers as well as soft fruits containing a number of shiny black seeds. In addition, the leaf contains phytochemicals such as hydroxybenzoic acid and ellagic acid (Alizarine yellow).The use of *T. daniellii* in the African folk medicine is evident by the fact that its leaf sap is used as an anti-dote against venoms, stings and bites, while the leaf and root sap are used as sedative and has been instrumental in the management of mental conditions [3].

Hematological parameters are useful indices that can be relied upon to ascertain the toxic potential of plant extracts or herbal medicine in a living system [4]. They can be used to measure the influence of certain plant derived compounds on blood. Owing to their high sensitivity and reliability they can be considered the strength of ethical and rational research on disease diagnosis, prevention as well as treatment [5].

Although, scientific reports abound on the therapeutic potentials of *T. dainiellii* and *A. cordifolia,* information deficit on the hematological implications of their usage as therapies probably owing to the irrational belief that naturally sourced medications are safe necessitates the need for this important research to either consolidate or discard this wildly held belief of the local users.

2. Material and methods

2.1. Collection and preliminary processing of plant material

Leaves of *T. daniellii* and *A*. *cordifolia* were collected from a farmland within Uturu community in Isiukwuato Local Government Area of Abia State, Nigeria. The plants were subsequently taken to the herbarium unit of the Department of Forestry, Micheal Okpara University of Agriculture Umudike, Abia State Nigeria. The leaves were separately and thoroughly washed using clean water before being dried at room temperature. Dried leaves were ground separately to powder and sieved with a suitable wire mesh to obtain a fine powder.

2.2. Extraction of plant materials

Fifty gram (50 g) each of the powdered plant samples was placed separately in 500 ml of methanol and water before being macerated at room temperature for four days. The mixture was agitated three times daily and was subsequently strained after which the damp solid material was, pressed and the combined liquids, filtered with the aid of a cheese cloth and Whatman No. 1 filter paper. The filtrate was extracted using soxhlet apparatus for 5-6hrs and subsequently concentrated under pressure to dryness in rotary evaporator at 25-30 ^{o}C [6]. The extracts were placed in the refrigerator and maintained at 5-10 ^{o}C.

2.3. Median lethal dose 50% test (LD50)

Acute toxicity test was separately carried out on *T. daniellii* and *A. cordifolia* leaf extracts. Three groups of three rats each were administered with 10 mg, 100 mg, and 1000 mg/kg of the various extracts orally before being observed for 24hrs. In the absence of mortality in any of the groups, another three groups of one rat each were administered with 1600, 2900 and 5000mg/kg of plants extract orally after which the animals were observed for 48 hrs [7].

2.4. Animal

Twenty five (25) adult male wistar rats (116-122 g) were purchased from the Animal House of the Department of Pharmacology, University of Nigeria Nsukka. The rats were housed in plastic cages in a well-ventilated room with a 12/12hr light/dark cycle and ambient temperature for three weeks to acclimatize.

2.5. Experimental design

The research was divided into two parts identified as Study A and B. A total of twenty five (25) wistar rats were used for both studies. Meanwhile, each study consisted of two distinct groups of five rats each. A common control (Group I) of five rats which was administered with 2ml/kg distilled water orally was established for both studies thus;

Study A

Group II – 200 mg/kg aqueous extract of T.d (p.o)

Group III – 200 mg/kg methanolic extract of T.d (p.o)

Study B

Group II – 200 mg/kg aqueous extract A.c (p.o)

GroupIII – 200 mg/kg methanolic extract A.c (p.o)

2.6. Determination of haematological parameters

Blood samples collected from experimental animals were introduced into appropriately labeled EDTA containers. Hematological evaluation was conducted on the samples in accordance with the method described by Malamo *et al* [8]. RBC and WBC were counted with the aid of the Neubaur's Chamber. Packed Cell Volume (PCV) was determined using Wintrobe hematocrit tube, while hemoglobin concentration (Hb) was determined using the Coulter Counter System (Beckman Coulter®, ThrmoFisher, UK).

2.7. Statistical analysis

Data generated from the study was analyzed with the aid of the Analysis of Variance (ANOVA) and were expressed as mean ± standard error of mean (SEM) of three determinations. Differences in mean were compared using Duncan multiple test range, $P<0.05$ was considered statistically significant.

3. Results and discussion

Table 1: Hematological indices of wistar rats administered with aqueous and methanol extracts of *T. Daniellii*

Group	Hb (g/dl)	RBC(×10⁶/µL)	WBC(×10³/µL)	PVC%
Group I(ctrl)	17.13±0.01^{a}	69.83±0.01^{a}	7.13±0.02^{a}	48.75±0.02^{a}
Group II(200 mg/kg aq. T.D)	17.02±0.01^{a}	68.64±0.49^{ab}	7.83±0.02^{a}	45.44±0.02^{ab}
Group III (200 mg/kg met. T.D)	13.7±0.01^{b}	62.20±0.27^{c}	10.13±0.02^{b}	38.93±0.02^{c}

Values are mean ± standard error of mean (SEM) from three determinations. Values with different superscripts in a column are significantly different (P<0.05)

Table 2: Hematological parameters of wistar rats administered with aqueous and methanol Extracts of *A. cordifolia*

Groups	Hb (g/dl)	RBC(×10⁶/µL)	WBC(×10³/µL)	PCV%
Group I(ctrl)	17.13±0.01a	69.83±0.01^{a}	7.13±0.02^{a}	48.75±0.02^{a}
Group IV(200 mg/kg aq. A.c)	12.5±0.02b	61.92±0.56^{b}	9.54±0.02^{b}	41.15±0.02^{b}
Group V (200 mg/kg met. Ac)	12.19±0.02b	61.42±0.31^{b}	9.07±0.06^{b}	42.33±0.15^{b}

Values are mean ± standard deviation from three determinations. Values with different superscripts in a column are significantly different (P<0.05)

Blood is an ideal indicator universally employed in the determination of an organism's health status [9]. It is said to be a mirror with which pathological conditions can be traced, identified and managed. Cellular components of blood are valuable tools in evaluating the toxic potentials of therapeutic compounds. Owing to these interesting qualities of blood, haematological parameters which beckon wholly on blood are essential in evaluating and establishing the body's functional status following exposure to toxicants [10].

Table 1 shows the hematological indices of wistar rats administered with aqueous and methanolic leaf extracts of *T. daniellii.* There was a significant decrease in the Hemoglobin concentration (Hb), Red Blood Cell (RBC) and Packed Cell Volume (PCV) in Group III administered with 200 mg/kg methanolic extract of *T. daniellii* compared to Group I administered with 2ml/kg distilled water orally. However, there was no significant difference in the values obtained for Groups II and I administered with 200 mg/kg aqueous leaf extract of *T. daniellii* and 2ml/kg distilled water orally

respectively. Significant increase in White Blood Cell (WBC) was observed following the administration of 200mg/kg methanolic leaf extract of *T. daniellii* compared to the control group.

Table 2 shows the hematological indices of wistar rats administered with aqueous and methanolic leaf extracts of *A. cordifolia*. A significant reduction in hemoglobin concentration (Hb), Red Blood Cell (RBC), White Blood Cell count (WBC) and Packed Cell Volume (PCV) was observed in Groups II and III administered with 200 mg/kg aqueous and methanolic leaf extracts of *A. cordifolia* respectively. The decrease in Hb, RBC and PCV observed following administration of the various leaf extracts could be as a result of the presence of some bioactive compounds with the capacity to suppress normal erythropoiesis promoting mechanisms and or deficiency of some factors required for the maturation of red blood cells [9]. Lysis of erythrocytes has been implicated in some instances [11].

The white blood cells in human, like every other higher animal respond to various sensors including infections and chemical irritants [12]. Thus, increase in the number of white blood cells is a normal reaction to exposure to toxicants [11]. The observable increase in the WBC (leukocytosis) may be as result of the excitation of the defense mechanism to counter the effects of the toxicants [13].

These results are consistent with the findings of Yaya and Ajay [9] which showed that taraxel and Apigenin bioactive compounds isolated from *Jatropha gossypifolia* belonging to the family Euphorbiacea to which *A. cordifolia* belongs could decrease blood parameters such as Hb, PCV, RBC and increased WBC.

4. Conclusion

In conclusion, diverse solvents are available for the isolation of bioactive compounds from varying plant parts. Accurate knowledge of their capacities is important in the preliminary drug discovery approach. From these studies, it can be deduced that drug discovery approach on *T. daniellii* leaf should preferably involve an aqueous medium. This is to avoid inclusion of potential anemia inducing compounds present in candidate plant. However, the usage of the aqueous leaf extract of *A. cordifolia* for therapeutic reasons should be complemented with an anti-anemic drug or preparations.

Compliance with ethical standards

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Disclosure of conflict of interest

We declare that no conflict of interest exist on this work.

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