

Effect of Ethanolic Root Extract of *Napoleonaea Imperialis* on Some Haematological Parameters in Albino Rats

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ABSTRACT

This study investigated the effects of *Napoleonaea imperialis* (NI) ethanolic root extracts on some haematological parameters in albino rats. Thirty six (36) apparently healthy albino rats were used, divided into 6 groups of 6 rats each. The rats were fed a commercial pelleted poultry grower's mash diet. Groups A, B, C, D and E received 10, 20, 30, 40, and 50 mg/kg body weight (bwt) of NI root extract three times a week at two-day intervals over a period of three weeks. Group F (control) received water only. 2 ml of whole blood was collected from each albino rat into EDTA container for the determination of red blood cell count [RBC], haemoglobin [Hb], packed cell volume [PCV], total white blood cell count [TWBC], platelet [PLT] count, and WBC differentials [neutrophils, lymphocytes, and mixed cells] using a three-part Biobase haematology analyzer (BK-6190). The results showed there were no statistically significant differences ($p > 0.05$) in the mean red blood cell count, packed cell volume, haemoglobin, total white blood cell count, lymphocyte count, mixed cell count and platelet count. This shows that short-term ingestion of *Napoleonaea imperialis* ethanolic root extract has no harmful effects on the haematological parameters studied.

INTRODUCTION

Herbs were used extensively in antiquity to treat, manage, and alleviate a wide range of illnesses. The quest for using plant extracts in the creation of medications in contemporary medicine was posed by this. It has been acknowledged that a number of plants contain therapeutic properties that can be used as adjuvants in the therapeutic management of diseases. As a result, numerous plant parts, including roots, barks, seeds, and leaves are used for therapeutic purposes.

Medicinal plant is any plant that has compounds that can be utilized therapeutically or that serve as building blocks for the production of effective pharmaceuticals (Sofowora *et al.*, 2013). This definition enables the distinction between plants that are considered medicinal but have not yet undergone a full scientific investigation and plants whose therapeutic capabilities and ingredients have been established scientifically (Sofowora *et al.*, 2013). In fact, a great number of plants have been studied for this purpose, while others have either not yet been studied or have not yet had their therapeutic properties completely explored (Ezeugwunne *et al.*, 2017; Ogbodo *et al.*, 2017a; Ogbodo *et al.*, 2017b; Ogbodo *et al.*, 2017c; Uduchi *et al.*, 2022c; Uduchi *et al.*, 2022d). The presence of phytochemicals, which are molecules found in plants and are responsible for the biological activities of the herbs, has been linked to the medicinal benefits of herbs (Mendoza and Silva, 2018).

Napoleonaea imperialis P. Beauv is a Nigerian folkloric medicinal plant. It is a small, evergreen tropical West African tree in the family, Lecythidaceae (Badri and Renu, 2011). *Napoleonaea imperialis* P. Beauv is one of the plants employed in ethnomedicine in Nigeria. *Napoleonaea imperialis* P. Beauv has previously been found to contain phytochemicals such as terpenoids, saponins, steroids, and tannins (Jack *et al.*, 2020). The bark and the fruit pulp are used as a cough medicine and

raw bark is chewed for this effect (Ojinnaka and Okpala, 2012). Also, Chah *et al.* (2006) demonstrated antibacterial and wound healing properties of *N. imperialis* in rats. In another experiment, Esimone *et al.* (2005), prepared a herbal ointment of the methanolic extract of *N. imperialis* and evaluated its wound healing effect by the excision wound model on guinea pigs. Furthermore in their study, Etim *et al.* (2020) suggested that *Napoleonaea imperialis* extract has strong anti-oxidative and anti-inflammatory potential and could be used to treat disorders caused by inflammation and oxidation. Although the plant is said to have several health advantages, very little research has been done on it. Therefore, the current study examined the effects of *Napoleonaea imperialis* ethanolic root extracts on certain haematological parameters in albino rats.

MATERIALS AND METHODS

Study Area

This study was carried out in Orlu, Imo State, South-Eastern Nigeria. Imo State shares boundary with Anambra State in the North, Rivers State in the South and West and Abia State in the East.

Plant Materials

Roots of *Napoleonaea imperialis* P. Beauv were purchased from Orie market, Umuna, Orlu. This was identified in the Department of Plant Science and Biotechnology, Imo State University, Owerri and voucher specimens of the plant were deposited in the herbarium of the same department.



Figure 1: Diagram of *Napoleonaea imperialis* root.

Laboratory Animals:

The laboratory *Rattus albus* Wistar strain animals used in this investigation ranged in age from 2 to 3 months and weight from 120 to 180g. The albino rats were purchased from an established pet store. The albino rats were housed in isolation for two weeks to acclimatize to the laboratory environment, and they were fed a commercial chicken grower's mash diet. Potable water was also provided at intervals.

Laboratory Animal handling

Each animal was cared for in accordance with the National Institutes of Health (NIH) Guidelines for the Care and Use of Laboratory Animals (NIH publication, 1985).

Inclusion Criteria: Apparently healthy albino rats were selected for the study.

Exclusion criteria: Albino rats with cardiac and renal disease markers when tested were excluded from the research.

Processing of Plant Materials

The roots of *Napoleonaea imperialis* were dried under the shade and finally in thermostatically controlled hot air oven at 40°C until each maintained constant weight. Each was ground into fine powder using a warren blender machine and sieved using 1mm mesh sieve. The powdered plant materials were stored in labeled screw capped bottles and stored in the fume cupboard until required for extraction.

Extraction of Active Principles of the Plant Materials

The active components of *Napoleonaea imperialis* root extract were extracted with ethanol at 78 °C using the soxhlet extraction method, according to Obiajuru and Ozumba (2009) and Harborne (1998). The extracts were gathered and stored at +8 °C in screw-capped MacCartney vials until they were needed.

Experimental Design

Thirty six (36) apparently healthy albino rats were used in the study to determine the effects of *Napoleonaea imperialis* ethanolic root extract on haematological parameters. Each albino rat received its normal daily feed and water while experimental groups in addition to their normal feed and water were treated with different doses of the *Napoleonaea imperialis* ethanolic root extracts three times a week at two days interval for a period of 3 weeks.

The experimental rats were divided into six groups, each made up of six albino rats according to:

- Group A:** received 10mg of *Napoleonaea imperialis* ethanolic root extract/Kg body weight
- Group B:** received 20mg of *Napoleonaea imperialis* ethanolic root extract/Kg body weight.
- Group C:** received 30mg of *Napoleonaea imperialis* ethanolic root extract/Kg body weight.
- Group D:** received 40mg of *Napoleonaea imperialis* ethanolic root extract/Kg body weight.
- Group E:** received 50mg of *Napoleonaea imperialis* ethanolic root extract/Kg body weight.
- Group F:** received feed and 0.5ml of water only.

The albino rats were anesthetized at the end of the third week by placing them on wire gauze and placing cotton wool soaked in diethyl ether beneath the gauze in a clear glass dessicator.

Thereafter, 2 ml of blood from each albino rat was drawn through heart puncture into Ethylene-diamine-tetra-acetic acid (EDTA) bottles for evaluation of haematological parameters.

Estimation of Haematological Parameters

The haematological parameters (red blood cell count [RBC], haemoglobin [Hb], packed cell volume [PCV], total white blood cell count [TWBC], platelet [PLT] count, and WBC differentials [neutrophils, lymphocytes, and mixed cells]) were analyzed using a three-part Biobase full blood count analyzer (BK-6190).

Statistical analysis

Using IBM SPSS Statistics version 23.0, the study's data were analyzed using analysis of variance (ANOVA) and posthoc tests. Results were presented as mean \pm standard deviation, and significance was assumed at a p-value of < 0.05.

RESULTS

When analyzed using an ANOVA table and a posthoc test, there were no statistically significant differences in the mean packed cell volume, red blood cell count, and hemoglobin levels ($p > 0.05$) respectively (**Table 1**).

Additionally, when the mean total white blood cell count (TWBC) and platelet count were compared using an ANOVA table and a posthoc test, respectively, there were no statistically significant differences ($p > 0.05$). See **table 1**.

Furthermore, there were no statistically significant differences in the neutrophil count, lymphocyte count and mixed cell count in the experimental groups when compared to the control group respectively (Table 2).

Table 1: Levels of PCV, Hb, RBC, TWBC and Platelet count in the albino rats administered with different doses of *Napoleonaea imperialis* root extracts (Mean± SD, n=36).

Groups	PCV (%)	Hb (g/dl)	RBC (pg/L)	TWBC (cells/ μ L)	Platelet (cells/ μ L)
Group A(10mg/Kg bwt; n=6)	34.50±0.71	11.50±0.24	5.75±0.12	6.95±0.35	257.00±8.49
Group B (20mg/Kg bwt; n=6)	37.25±1.71	12.42±0.57	6.21±0.28	6.73±2.07	226.75±9.98
Group C (30mg/Kg bwt; n=6)	36.67±1.21	12.22±0.40	6.11±0.20	9.28±1.68	274.83±17.24
Group D (40mg/Kg bwt; n=6)	37.67±1.37	12.56±0.46	6.28±0.23	11.33±2.11	257.67±15.65
Group E (50mg/Kg bwt; n=6)	35.50±2.12	11.83±0.71	5.92±0.35	6.85±0.35	226.00±8.49
Group F (control; n=6)	35.75±0.96	11.92±0.32	5.96±0.16	8.46±1.86	274.00±6.83
f-Value	1.518	1.518	1.518	1.311	0.284
p-Value	0.321	0.295	0.295	0.288	0.954
A Vs B	1.000	1.000	1.000	1.000	1.000
A Vs C	1.000	1.000	1.000	1.000	1.000
A Vs D	0.381	0.381	0.381	1.000	1.000
A Vs E	1.000	1.000	1.000	1.000	1.000
A Vs F	1.000	1.000	1.000	1.000	1.000
B Vs C	1.000	1.000	1.000	1.000	1.000
B Vs D	1.000	1.000	1.000	0.983	1.000
B Vs E	1.000	1.000	1.000	1.000	1.000
B Vs F	1.000	1.000	1.000	1.000	1.000
C Vs D	1.000	1.000	1.000	1.000	1.000
C Vs E	1.000	1.000	1.000	1.000	1.000
C Vs F	1.000	1.000	1.000	1.000	1.000
D Vs E	1.000	1.000	1.000	1.000	1.000
D Vs F	1.000	1.000	1.000	1.000	1.000
E Vs F	1.000	1.000	1.000	1.000	1.000

*Statistically significant at $p < 0.05$.

Table 2: Levels of Neutrophil, Lymphocyte and Mixed cell count in the albino rats administered with different doses of *Napoleonaea imperialis* root extracts (Mean± SD, n=36).

Groups	Neutrophil (%)	Lymphocyte (%)	Mixed cells (%)
Group A(10mg/Kg bwt; n=6)	37.00±8.49	63.00±1.41	0.00±0.00
Group B (20mg/Kg bwt; n=6)	41.25±4.57	58.75±4.57	0.00±0.00
Group C (30mg/Kg bwt; n=6)	37.17±2.32	62.33±2.25	1.50±0.71
Group D (40mg/Kg bwt; n=6)	33.33±4.50	65.54±2.85	2.01±0.00
Group E (50mg/Kg bwt; n=6)	38.50±2.12	60.00±2.83	1.50±0.71
Group F (control; n=6)	37.00±9.97	60.50±10.47	2.75±0.96
f-Value	1.830	1.633	0.297
p-Value	0.128	0.174	0.872

A Vs B	1.000	1.000	1.000
A Vs C	1.000	1.000	1.000
A Vs D	1.000	1.000	1.000
A Vs E	1.000	1.000	1.000
A Vs F	1.000	1.000	1.000
B Vs C	1.000	1.000	1.000
B Vs D	0.937	1.000	1.000
B Vs E	1.000	1.000	1.000
B Vs F	1.000	1.000	1.000
C Vs D	1.000	1.000	1.000
C Vs E	1.000	1.000	1.000
C Vs F	1.000	1.000	1.000
D Vs E	1.000	1.000	1.000
D Vs F	1.000	1.000	1.000
E Vs F	1.000	1.000	1.000

*Statistically significant at $p < 0.05$.

DISCUSSION

The mean packed cell volume, red blood cell count, and haemoglobin level between the groups given different dosages of *Napoleonaea imperialis* and the control group did not show any statistically significant differences in the current study. This finding shows that *Napoleonaea imperialis*, when ingested briefly, is not haematotoxic and may not have haematinic value. Furthermore, it demonstrates that there was no reduction in the number of red blood cells produced and no change in the rate of red blood cell formation (erythropoiesis) as a result of its inability to promote the production and release of erythropoietin, a substance essential for the formation of red blood cells. The non-significant effects of the *Napoleonaea imperialis* ethanol extract suggest that there were no changes in the blood's capacity to carry oxygen and the amount of oxygen given to tissues, since red blood cells and haemoglobin are essential for transporting respiratory gases (Isaac *et al.*, 2013). This is in contrast with the report of Etim *et al.* (2015) which recorded significantly higher mean packed cell volume and haemoglobin levels in rats fed with *Napoleonaea imperialis* root. These differences in result may be due to disparity in dosages administered and duration of administration.

This study found no statistically significant difference in the mean total white blood cell count in test groups compared to the control. This corroborate well with the finding of Etim *et al.* (2015) that recorded no significant effect of *Napoleonaea imperialis* root on white blood cell count based on their study that evaluated the effect of methanolic root extract of *Napoleonaea imperialis* on some haematological parameters of albino rats. The non-significant change in TWBC count caused by the ethanolic *Napoleonaea imperialis* root extract suggests that the immune system was not compromised or affected by the administration of the root.

Additionally, there were no statistically significant differences in the mean neutrophil count, lymphocyte count, or mixed cell count between the experimental groups and the control group, respectively.

This would imply that the short-term ingestion of *Napoleonaea imperialis* ethanolic root extract does not impair the body's ability to fight off infections caused by foreign bodies, particularly bacteria, virus and fungi. This is in keeping with the reports of other similar studies (Etim *et al.*, 2015).

Again, when comparing the experimental groups to the control group, this study did not find any statistically significant difference in the mean platelet count. As platelets are involved in the blood clotting process, this may indicate that *Napoleonaea imperialis* ethanolic root extract is unable to enhance thrombopoietin synthesis.

CONCLUSION

This study revealed no significant changes in the red blood cell count, packed cell volume, haemoglobin, total white blood cell count, lymphocyte count, mixed cell count, or platelet count. This shows that short-term ingestion of *Napoleonaea imperialis* ethanolic root extract has no harmful effects on haematological parameters.

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