

Effect of Ethanolic Root Extract of *Sphenocentrum jollyanum*, *Baphia nitida*, *Pinus koraiensis*, and Sildenafil on Some Haematological Test Parameters in Albino Wistar Rats

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Abstract- *Sphenocentrum jollyanum*, *Baphia nitida* and *Pinus koraiensis* are evergreen shrub plants usually used as an aphrodisiac and have been used in traditional medicine practice for centuries with its leaves, roots and latex all in use. Sildenafil (Viagra) are drugs used to treat erectile dysfunction in men. This study is aimed at investigating the effect of ethanolic root extract of *Sphenocentrum jollyanum*, *Baphia nitida*, *Pinus koraiensis* and Sildenafil (Viagra) tablets on some haematological test parameters of albino rats. The experimental animals were grouped into five different groups with 5 animals each. Group 1 (control) were fed with vital pellet feed and tap water only, Group 2 received 80mg/kg of grinded tablets of sildenafil, Group 3 received 200mg/kg *Baphia nitida*, Group 4 received 200mg/kg of *Sphenocentrum jollyanum*, while Group 5 received 600mg/kg of *Pinus koraiensis* (Korean pine nuts). Haemoglobin, Packed cell volume, White blood cell, Red blood cell, and Platelet concentration were determined using automated haematological analyzer systemex KX-21 (Japan). The result shows a significant difference ($p < 0.05$) in PCV level of Group 5 (32.4 ± 3.2 %) compared to the control rats (41.7 ± 1.2 %) and Group 2 (39.6 ± 2.9 %), as well as Group 3 (38.8 ± 1.2 %) and Group 4 (35.7 ± 8.2 %). Platelet counts also showed statistically significant difference ($p < 0.05$) in Group 4 ($307.6 \pm 9.18 \times 10^9/L$) compared to Group 2 ($428.5 \pm 14.1 \times 10^9/L$), as well as Group 3 and Group 5. There was no significant difference ($p > 0.05$) in the haemoglobin levels between the control rats and Group 2, 3 and Group 4. White blood cell counts showed higher mean values in Group 2 ($13.6 \pm 4.1 \times 10^9/L$), compared to the control rats ($9.9 \pm 1.4 \times 10^9/L$) and other groups, but the mean difference were not statistically significant ($p > 0.05$). While the red blood cell counts were not statistically significant. This result therefore suggests that ethanolic extract of the aphrodisiac plant *Sphenocentrum jollyanum* (Ikeagwu), *Baphia nitida* (Abosi) and *Pinus koraiensis* as well as Sildenafil tablets at certain concentration could be desirous or deleterious so caution should be applied during oral ingestion.

Keywords: *Sphenocentrum jollyanum*, *Baphia nitida* and *Pinus koraiensis*, Red blood cell, Platelets

I. INTRODUCTION

Medicinal plants are plants that possess therapeutic properties and despite the widespread use of modern medicine, herbal products are still in use in most developing countries of Africa and Asia for the management of ailments. A considerable percentage of medicinal plants identified over the World are from tropical Africa [1]. Herbal medicine is generally believed to be more effective and having fewer side effects compared to synthetic medicines. More often, this has led to indiscriminate use without appropriate dose resulting in abuse. The incidence of adverse effects of these herbal remedies and sometimes life-threatening conditions has been reported among various ethnic groups [2]. This has made it imperative to ascertain their effects on blood parameters even though they have been used for ages to enable for scientific documentation on their safety/risk potentials [3].

Sphenocentrum jollyanum is an evergreen shrub growing around 1.5 metres tall and has been in traditional medicine practice for centuries with its leaves, roots and latex all in use [4].

Baphia nitida (camwood, also barwood), also known as African sandalwood, is a shrubby, leguminous, hardwooded tree from central West Africa. This wood is of a very fine colour, and is used in woodturning for making knife handles and similar articles. The tree's bark and heartwood are commonly used to make a brilliant but non-permanent red dye, which is soluble in alkali [5].

Pinus koraiensis (Korean pine) is a member of the white pine group. It grows to 30-50' tall in cultivation, but may reach 100' or more in its native habitat. When young, this tree typically grows in a narrow pyramidal form with ascending branching. With age, it relaxes into a loose pyramidal shape with a rounded crown and branching that is almost horizontal [6].

Sildenafil (Viagra) are drugs used to treat erectile dysfunction (impotence, inability to sustain a satisfactory erection to complete intercourse) in men, by increasing blood flow to the penis during sexual stimulation [7]. This increased blood flow can cause an erection. Sildenafil acts by inhibiting cGMP-specific phosphodiesterase type 5 (PDE5), an enzyme that promotes degradation of cGMP, which regulates blood flow in the penis. It also used to improve the ability to exercise in adults with pulmonary arterial hypertension [8].

Research has shown that oral ingestion of medicinal compounds or drugs can alter the normal range of some haematological parameters. These alterations could either be positive or negative [9]. It has therefore become necessary to investigate the effect of ethanolic extract of *Sphenocentrum jollyanum*, *Baphia nitida*, the seed of *Pinus koraiensis* and sildenafil tablets on blood parameters.

Rationale

In the world today, *Sphenocentrum jollyanum*, *Pinus koraiensis*, *Baphia nitida*, and Sildenafil is widely known and used in increasing libido activity when consumed. As a result, it has therefore become necessary to evaluate its effects on some blood parameters. Since much work has not been done in this field, as a result individuals ignorantly take these plants [10].

II. MATERIALS AND METHODS

Materials

The roots of *Sphenocentrum jollyanum* and *Baphia nitida* were purchased from Aba, Abia state. The seeds of *Pinus koraiensis* and 100mg tablets of sildenafil were purchased from Port Harcourt, Rivers state. All plant materials (*Sphenocentrum jollyanum* and *Baphia nitida* roots) were identified at the herbarium, University of Nigeria, Nsukka. It was pilled, chopped, air-dried and then grounded and sieved to fine powder.

Animals

A total of 25 healthy red eyed albino rats were collected from the animal house of the Federal University of Technology, Owerri. Using movable plastic baskets covered with wire gauge which the albino rats were weighed between (200-240g, initial weight). They were kept in the animal house of Madonna University, Elele Campus, where they were fed with standard pellet die (vital feed) and tap water and were allowed to acclimatize for two weeks before administration of any substance.

Proximate Composition of the Rats' Diet

Before the experiment, the rats were fed with palletized fowl grower's mash. The proximal composition of rat' diet was;

Phosphorus	0.33%
Calcium	0.8%

Crude ash	0.8%
Crude fat	4.8%
Crude fibre	7.2%
Lysine	0.6%
Manganese	30mg
Methionine	0.29%
Sodium	0.15%
Vitamin A	8000 I.U
Vitamin C	50mg
Vitamin D	2400 I.U
Vitamin E	15mg
Zinc	30mg

2.1.4 Apparatus Used

Spectrophotometer	Jenway, England
Dissecting kits	Hawsley, England
Centrifuge	Labman, U.K
Water bath	Kutterman, U.K
Electronic Weighing balance	Arman, England
Automated haem analyzer	KX-21 (Japan)

Protein Evaporator

Chemicals Used

Ethanol
Distilled Water

Methods

Extraction of Plant Materials

The ethanolic extract of *Sphenocentrum jollyanum* and *Baphia nitida* was obtained by soaking 1000g of the fine powder of *Sphenocentrum jollyanum* and 500g of the fine powder of *Baphia nitida* in 99/100% of ethanol and the mixture was kept for 72 hours with occasional shaking. The macerated mixture was filtered and the ethanolic filtrate was concentrated using a protein evaporator to obtain a crude extract which was then air-dried in a carefully regulated water bath (maintained at a temperature of 24°C) for 36 hours to yield 50.2g of dark solid extract.

Preparation of Extracts

The roots of *Sphenocentrum jollyanum* and *Baphia nitida* were pilled, cut into pieces and dried. The dried pieces were grounded into fine powder using an electric blender. The powdered samples (1000g of *Sphenocentrum jollyanum* and 500g of *Baphia nitida*) were extracted in 6000ml of ethanol for 72 hours. The extracts were filtered and concentrated by

removing the solvent completely using water bath. The volumes of extract suspension were given according to various weights of the rats. *Pinus koraiensis* and tablets of sildenafil were grinded with a mortar and pestle.

Experimental Design

The experiment lasted for 30 days. Before the commencement of the project, the 25 albino animals (rats) were fed with standard pellet die (vital feed) and tap water and were allowed to acclimatize for two weeks. The rats were weighed and then divided into five (5) different groups which contained 5 rats each and then treated accordingly.

Administration of Extract

The plant extracts were administered to the different groups except Group 1 (control group) which was only fed with standard pellet die (vital feed) and tap water. Group 2 were administered 80mg/kg body weight of the grinded tablets of sildenafil for 30 days. Group 3 were administered 200mg/kg body weight of the extract of *baphia nitida* for 30 days, Group 4 were administered 200mg/kg body weight of the extract of *Sphenocentrum jollyanum* for 30 days while Group 5 was administered 600mg/kg body weight of the grinded seeds of *Pinus koraiensis* (korean pine nuts) respectively for 30 days.

All animals were reweighed before they were sacrificed on day 30. They were anaesthetized with chloroform and were sacrificed. Blood samples were obtained by ocular puncture into EDTA sample bottles for haematological analysis within 12 hours of collection.

Determination of Haematological Parameters

The haematological parameters were evaluated with an automated haematological analyzer systemex KX-21 (Japan)

Statistical Analysis

Data obtained from this study were analyzed using the statistical package for social sciences (SPSS) version 20.0 for windows. Analysis of variance (ANOVA) were used to compare means, and values were considered significant at $p < 0.05$. Post hoc multiple comparisons for the ANOVA were done using least significant difference (LSD).

III. RESULTS

Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Packed Cell Volume (PCV) in Albino Rats

Fig 1 below shows that the PCV levels were significantly lower ($p < 0.05$) in the albino rats administered aphrodisiac plant *Pinus koraiensis* ($32.4 \pm 3.2 \%$) compared to the control rats ($41.7 \pm 1.2 \%$) and the group administered Sildenafil ($39.6 \pm 2.9 \%$), as well as those administered the aphrodisiac plants *Baphia nitida* (Abosi) ($38.8 \pm 1.2 \%$) and *Sphenocentrum jollyanum* (Ikeagwu) ($35.7 \pm 8.2 \%$).

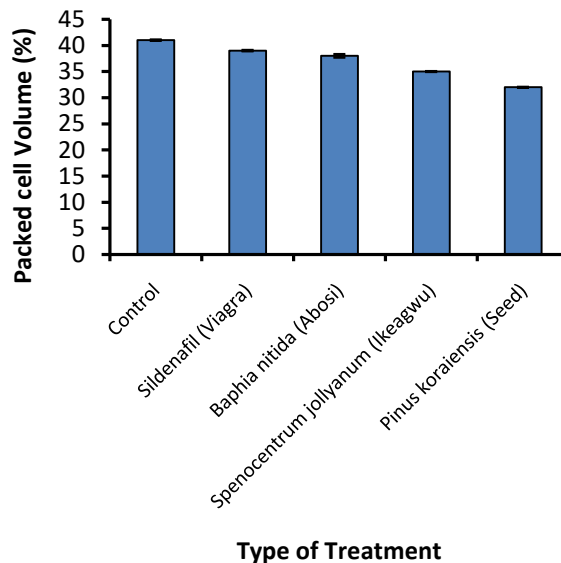


Fig 1: Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Packed Cell Volume (PCV) in Albino Rats

Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Platelet Counts in Albino Rats

As shown in Fig 2 below, the platelet counts were significantly lower ($p < 0.05$) in the albino rats administered aphrodisiac plant *Sphenocentrum jollyanum* (Ikeagwu) ($307.6 \pm 9.18 \times 10^9/L$) compared to the group administered Sildenafil ($428.5 \pm 14.1 \times 10^9/L$), as well as those administered the aphrodisiac plants *Baphia nitida* (Abosi) and *Pinus koraiensis*.

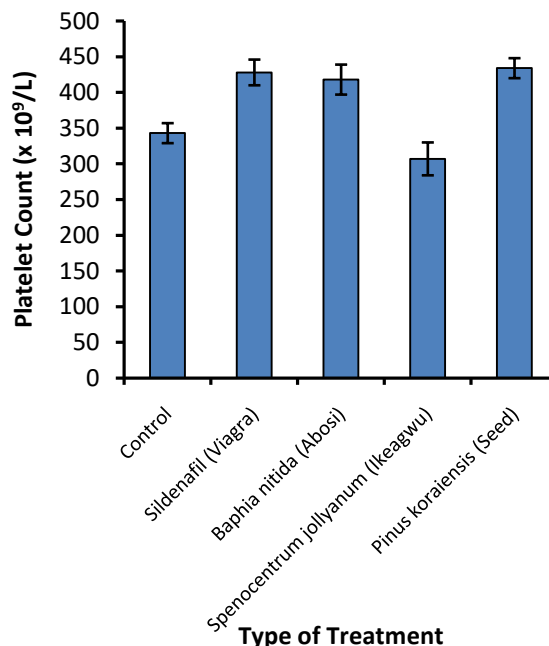


Fig 2: Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Platelet Counts in Albino Rats

Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Haemoglobin Levels in Albino Rats

Fig 3 below shows that there was no significant difference ($p > 0.05$) in the haemoglobin levels between the control rats and those administered sildenafil and the aphrodisiac plant *Sphenocentrum jollyanum* (Ikeagwu) and *Baphia nitida* (Abosi). Rats administered *Pinus koraiensis* however, had significantly lower ($p < 0.05$) haemoglobin levels compared to all other groups.

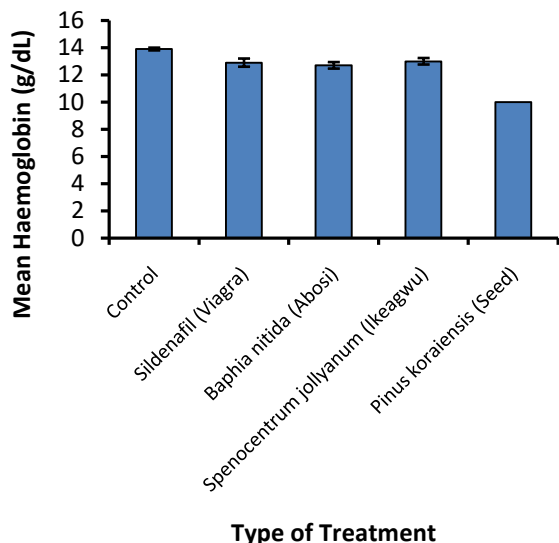


Fig 3: Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Haemoglobin Levels in Albino Rats

Effect of Sildenafil and Aphrodisiac Plant Extract Administration on White Blood Cell Count in Albino Rats

As shown in Fig 4 below, the white blood cell counts were higher in the albino rats administered Sildenafil ($13.6 \pm 4.1 \times 10^9/L$), compared to the control rats ($9.9 \pm 1.4 \times 10^9/L$) and those administered the aphrodisiac plant *Sphenocentrum jollyanum* (Ikeagwu), *Baphia nitida* (Abosi) and *Pinus koraiensis*, although the mean difference were not statistically significant ($p > 0.05$).

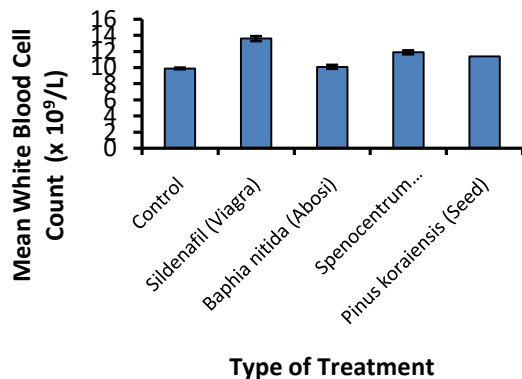


Fig 4: Effect of Sildenafil and Aphrodisiac Plant Extract Administration on White Blood Cell Count in Albino Rats

Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Red Blood Cell Counts in Albino Rats

Fig 5 below shows that there was no significant difference ($p > 0.05$) in the red blood cell count the control albino rats ($7.0 \pm 0.4 \times 10^{12}/L$) compared to those administered Sildenafil ($6.0 \pm 0.1 \times 10^{12}/L$), as well as those administered the aphrodisiac plants *Sphenocentrum jollyanum* (Ikeagwu), *Baphia nitida* (Abosi) and *Pinus koraiensis*.

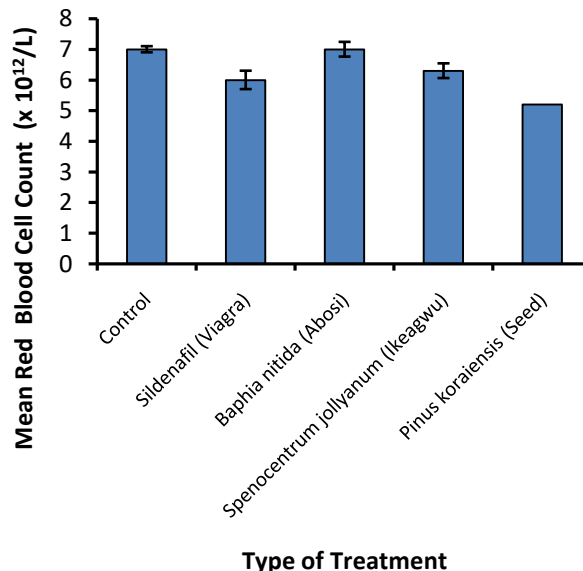


Fig 5: Effect of Sildenafil and Aphrodisiac Plant Extract Administration on Red Blood Cell Counts in Albino Rats

IV. DISCUSSION AND CONCLUSION

Discussion

Sphenocentrum jollyanum, *Baphia nitida*, and *Pinus koraiensis* are plants shown to have some medicinal functions [11]. The root of *Sphenocentrum jollyanum* has been shown to be effective as an aphrodisiac [12]. Sildenafil citrate is a drug widely known for its positive effects on male erectile dysfunction [13].

These plant extracts and sildenafil were assayed to investigate their effects on the haematological parameters of albino wistar rats.

The result shows that there was significant reduction ($p < 0.05$) in PCV, haemoglobin and Red blood cell level of the rat groups treated with *Sphenocentrum jollyanum*, *Baphia nitida* and *Pinus koraiensis* as compared with the groups treated with the known drug sildenafil and the control group. This result was contrary to the work of [14] who reported an increase in the PCV, haemoglobin and Red blood cell levels of rats exposed to *Sphenocentrum jollyanum*. Another work by reference [15] however showed an inverse relationship. It was shown that the higher the dose, the lower the PCV, haemoglobin and Red blood cell results. This could be due to

a number of reasons like cell destruction, blood loss which may be linked to higher doses of the plant extracts.

For the platelet and White blood cell result, it was observed that all the treatment groups were higher than the control group save for the group treated with *Sphenocentrum jollyanum* in the platelet results. Also the difference was statistically significant for the platelet result but not statistically significant for the White blood cell result. This result also agrees with the work of [14] who reported an increase in the platelet level but a decrease that was not significant in the white blood cell analysis. This significant increase in platelet after oral administration of the extracts show that the extracts have the potential to activate thrombopoietin production and can thus be used to manage hemostatic capacity of blood since platelets in blood mediate clotting mechanisms [16].

The rise in white blood cell levels in the test animals show that the ethanolic extracts of *Sphenocentrum jollyanum*, *Baphia nitida*, and *Pinus koraiensis* may possess immune boosting properties that are similar to those reported for seed extracts of Citrus paradise Macfad [17]. A report by reference [18] showed that granulocyte-macrophage colony stimulating factor, macrophage colony stimulating factor, interleukins IL - 2 IL-4 and IL-5 regulate the proliferation, differentiation and maturation of committed stem cells that are involved in white blood cell production. It could be argued that since *Sphenocentrum jollyanum*, *Baphia nitida*, and *Pinus koraiensis* led to an increase in the levels of the white blood cells, it is possible that the phytochemicals in the extracts stimulated the production of these regulatory factors of the committed stem cells, responsible for the generation of white blood cells [16].

Conclusion

The study has shown that at higher doses of *Sphenocentrum jollyanum*, *Baphia nitida*, and *Pinus koraiensis*, there could be both desirous and deleterious effects, hence caution should be applied in the use of these crude extracts for medicinal purposes.

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