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The Effects of Aqueous Leaf Extract of *Symphytum Officinale* (Comfrey) on the Spleen of Adult Wistar Rats.

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Abstract

Symphytum officinale (comfrey) root and leaf have been used since Roman times, dating back thousands of years. This herb has been utilized in folk medicine throughout Europe and North America and has been widely cultivated as a garden medicinal specifically for its reputation for healing various internal and external wounds. This study is aimed at investigate the effects of aqueous leaf extract of *Symphytum officinale* (comfrey) on the spleen of an adult wistar rats. Twenty wistar rats weighing between 180-210g were used for the study and were allocated into four (4) groups of five (5) animals each. Group A served as the experimental control and were

orally administered 0.3ml of distilled water; while groups B, C & D orally administered 0.2ml, 0.4ml and 0.6ml of aqueous leaf extract of *Symphytum officinale* respectively for twenty eight days. Twenty four hours after the last administration, the animals were weighed and weight were recorded, anaesthetized using chloroform inhalation method and dissected. Organ (spleen) tissues were harvested, weighed and trimmed down to a size of 3mm× 3mm thick and fixed in 10% formalin for histological studies. The final body weight of groups C and D decreased significantly ($P<0.005$) when compare with the control group A. the relative organ weight of groups C and D animals increased significantly ($P<0.005$) when compare with the control group A while group B were statistically similar with the control group A. Histological findings revealed distortion of the spleen cells of the experimental groups C and D.

Keywords: *Symphytum officinale*, Wistar Rats, Distilled Water, Organ Weight, Spleen.

Introduction

Plant-based treatment in Ayurveda may be derived from roots, leaves, fruits, bark, or seeds such as Cardamom and Cinnamon. In the 19th Century, William Dymock and co-authors summarized hundred of plants-derived medicines along with the uses, microscopic structure, chemical composition, toxicology, prevalent myths and stories and relation to commerce in British India [1]. There are roughly 13,000 medicinal used in China and over 100,000 medicinal recipes recorded in the ancient literature [2], plant elements and extracts are by far the most common elements used [3]. According to cancer Research Uk, “there is currently no strong evidence from studies in people that herbal remedies can treat, prevent or cure cancer” [4]. Pharmaceuticals are prohibitively expensive for most of the world’s population, half of whom lived on less than 2 U.S per day in 2002 [5].

At least 7,000 medical compounds in the modern pharmacopoeia are derived from plants [6]. Among the 120 active compounds currently isolated from the higher plants and widely used in modern medicine today, 80% show a positive correlation between their modern therapeutic use and the traditional use of the plants from which they are derived [7].

Dangerously low blood pressure may result from the combination of an herbal remedy that

lowers blood pressure together with prescription medicine that has the same effect. Some herbs may amplify the effects of anticoagulants [8]. Certain herbs as well as common fruit interfere with cytochrome P450, an enzyme critical to much drug metabolism [9]. Comfrey has large, rough, hairy and lance shaped leaves with whitish, pink, or purple flower spikes which have a slight heliotrope like curl typical of this family. A member of the Borage or *Boraginaceae* family, comfrey's relatives include both borage (*Borago* Sp) and heliotrope (*Heliotropium* Sp.) [10, 11]. It is a yin tonic that has been utilized for wounds, however when there is concern about the pyrrolizidine alkaloid contained in the root, often *Relbunium glutinosum* is substituted as it has similar energetic. The root is considered nutritive, cooling and moist in traditional Chinese medicine [12]. Therefore, this study is aimed at investigating the effects of aqueous leaf extract of *Symphytum officinale* on the spleen of an adult wistar rats.

Materials and Methods

Breeding of Animals

Twenty (20) wistar rats weighing between 180-205g were used for the study and were obtained from the animal farm house, Department of Anatomy, Nnamdi Azikiwe University, Nnewi Campus. They were maintained under standard housing conditions and fed with standard rat chow (Growers mash) and provided with water ad libitum during the experiment. They were acclimatized for two weeks before the experiment.

Drug Preparation

Common comfrey (*Symphytum officinale*) leaves were plucked from Okitipupa in Ondo State. It was identified at herbarium unit, Botany Department, Nnamdi Azikiwe University, Anambra State. It was sun-dried and then milled to a powder. 300mg/kg body weight was dissolved in 10mls of distilled water and administered to the animals.

Experimental Protocols

Twenty (20) animals were weighed and allocated into four groups (a, B, C and D) of five (5) animals each. Group A served as the experimental control and were orally administered 0.3ml of distilled water; the experimental groups B, C and D were orally administered 0.2ml, 0.4ml and 0.6ml of aqueous leaf extract of *Symphytum officinale* respectively for twenty eight

days. Twenty four hours after the last administration, the animals were weighed and weights were recorded. They were anaesthetized using chloroform vapours inhalation method and dissected. Spleen tissues were harvested and fixed in zenkers fluid for histological studies.

Tissues Processing

For easy study of sections under microscope, the tissues passed through several processes of fixation, dehydration, clearing, infiltration, embedding, sectioning and staining. The fixed tissues were kept in zenkers fluid for four hours. After fixation, the tissues were washed over night under a stream tap water. Dehydration of the fixed tissues was carried out in different percentages of 50%, 70% and 90% absolute. the tissues were then clear in xylene and embedded in paraffin wax. Serial sections of 5micro thick are obtained using a rotator microtome. The tissue sections were deparaffined hydrated and stained using the routine haematoxylin and eosin method. The stained sections were then examined under the light microscope.

Results

Morphometric Analysis of Body Weight.

Table 1: The result obtained from calculation of initial, final body weight and weight changes in all the groups (A, B, C & D).

(Mean \pm SEM given for each measurement)

Groups	Initial Body Weight	Final Body Weight	Weight Change
Group A	184.20 \pm 3.60	196.40 \pm 4.70	12.20 \pm 1.10
Group B	186.40 \pm 1.80	193.60 \pm 2.10	7.20 \pm 0.30
Group C	190.50 \pm 2.50	182.20 \pm 1.40	-8.30 \pm 1.10
Group D	193.70 \pm 4.30	178.40 \pm 3.20	-15.30 \pm 1.10
F-Ratio	59.160	33.410	6.470
Prob of Sig	<0.005	<0.005	<0.005

Discussion

Comfrey root and leaf have been used since roman times, dating back thousands of years. This herb has been utilized in folk medicine throughout Europe and North America and has been widely cultivated as a garden medicinal specifically for its reputation for healing various internal

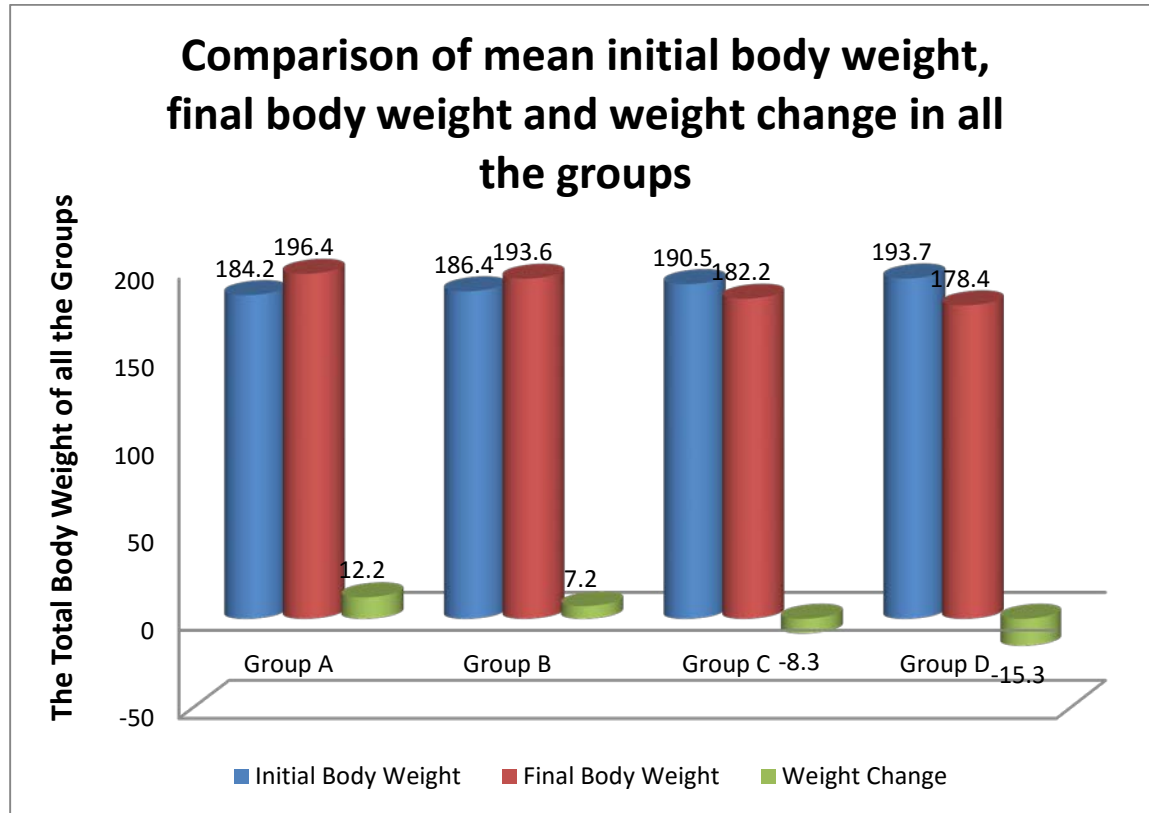


Figure 1: Bar chart showing the comparison of mean initial body weight, final body weight and weight change in all the groups.

Morphometric Analysis of Spleen Weight

Table 2: Comparison of the mean relative spleen weights of all the groups (A, B, C & D).

(Mean \pm SEM given for each measurement)

Groups	Spleen Weight
Group A	3.10 \pm 0.130
Group B	3.16 \pm 0.210
Group C	3.50 \pm 0.400
Group D	3.87 \pm 0.370
F-Ratio	26.90
Prob of Sig	<0.005

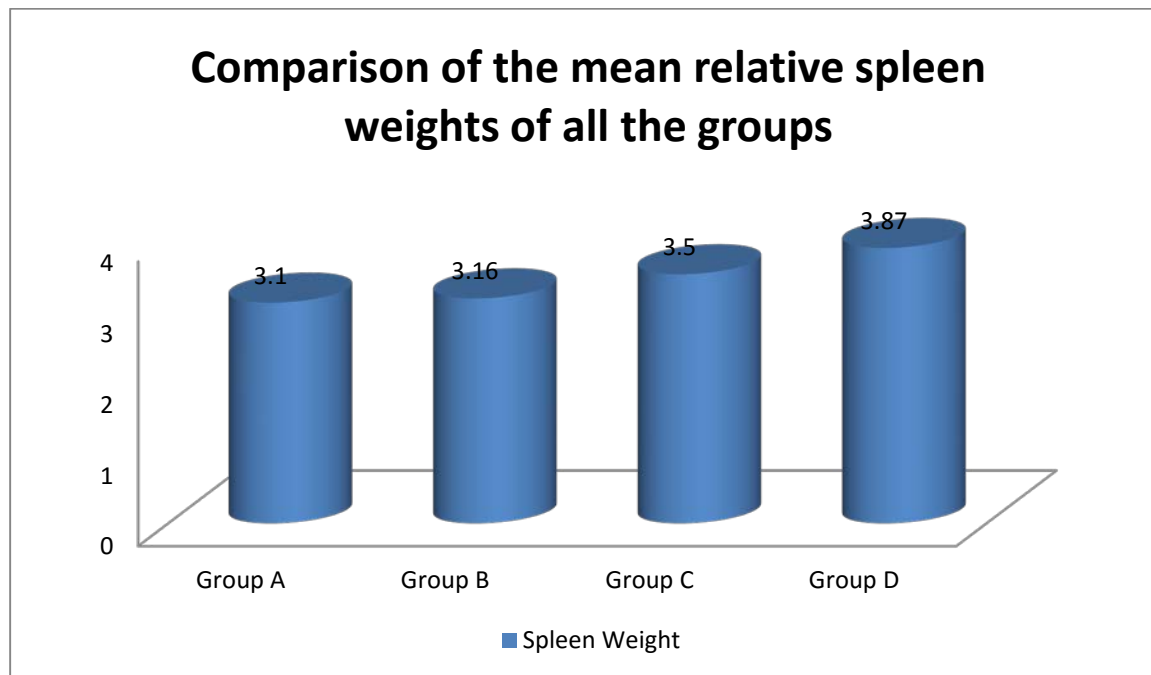
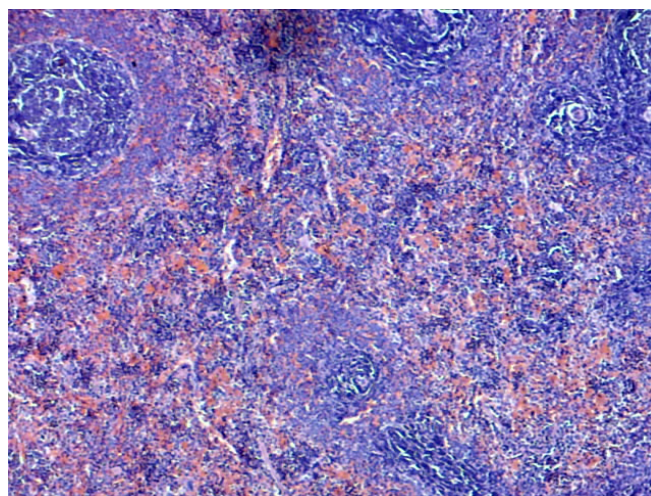
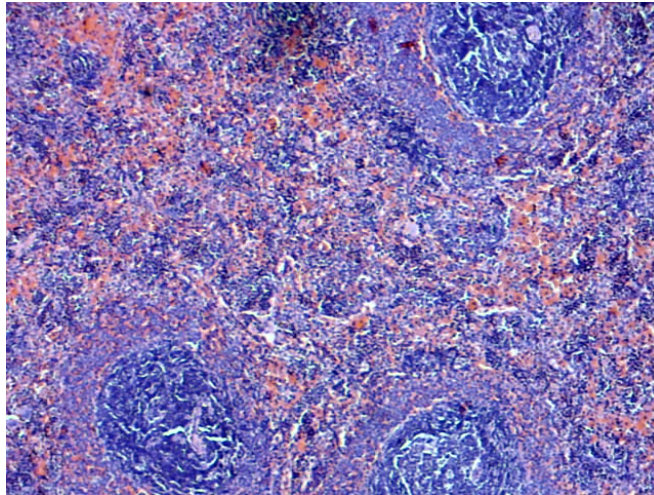


Figure 2: Bar chart showing the relative organ weights of all the groups.

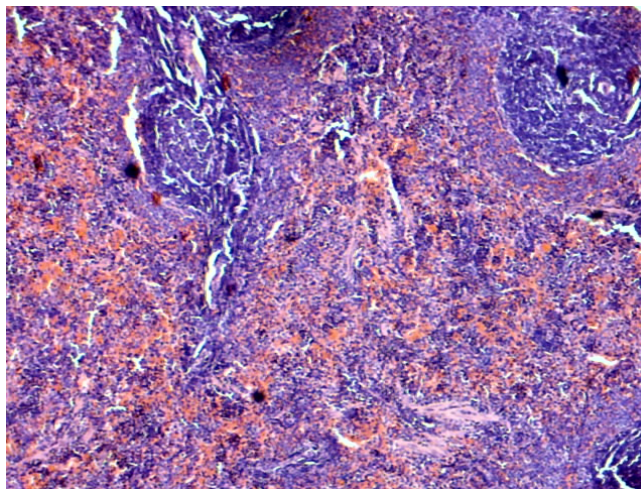
Histological Findings



Photomicrograph 1 (Group A control) showing normal architectural structure of the spleen.

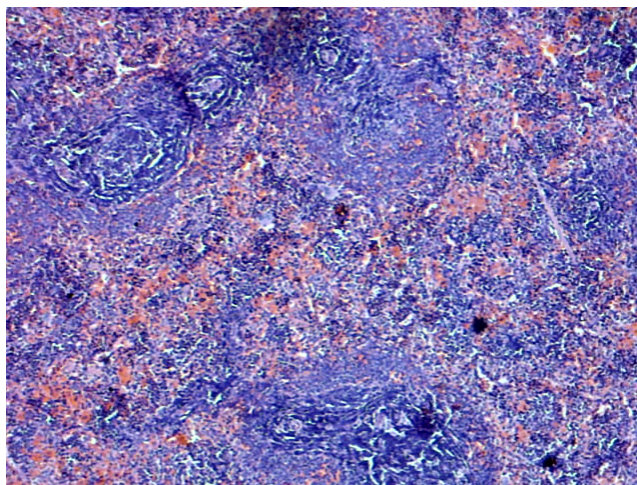


Photomicrograph 2 (Group B treated with 0.2ml of aqueous leaf extract of *Symphytum officinale*) showing normal architectural structure of the spleen.



Photomicrograph 3 (Group C treated with 0.4ml of aqueous leaf extract of *Symphytum officinale*) showing mild degeneration of the spleen cells.

and external wounds. Much debate surrounds the safety of comfrey due to various parts and preparations containing potentially toxic alkaloids. It is important to understand that the part used, species, and time of harvest all come into play when determining the safety of this herb. A large body of traditional use supports its safety and efficacy if used intelligently and cautiously [10, 13, 14, 15, 16].



Photomicrograph 4 (Group D treated with 0.6ml of aqueous leaf extract of *Symphytum officinale*) showing mild degeneration of the spleen cells.

It has been reported by some experts that comfrey should be restricted to topical use, and should never be ingested, as it contains dangerous amounts of hepatotoxic pyrrolizidine alkaloids (PAs) [17].

In this study, the final body weight of groups C and D animals decreased significantly ($P < 0.005$) when compared with the experimental control group A. The final body weight of group B animals increased significantly with the control group A.

The comparison of the mean relative organ weight of groups C and D increased significantly ($P < 0.005$) when compared with the control, while group B mean relative organ weight was statistically similar with the experimental control group A.

Histological findings revealed histological lesions in groups C and D treated with high doses of aqueous leaf extract of *Symphytum officinale*.

Conclusion

The result of this study revealed that consumption of leaf extract of *Symphytum officinale* at high doses could cause alteration in the cytoarchitecture of the spleen.

References

[1] William Dymock (1890). A history of principal drugs of vegetable origin in British India Pharmacographia Indica. Volume I.

- [2] Chen, K and Yu, B (1999) Certain progress of clinical research on Chinese integrative medicine. Chinese Medical Journal. 112 910), 934-7.
- [3] Foster and Yue (1992) P. 11 Herbal medicine. Cancer Research UK. Retrieved August 2013.
- [4] Edgard j, Dasilva, Elias baydoun, Adnan Badran (2002) Biotechnology and the developing world. Electronic Journal of Biotechnology. 5(1).123-127.
- [5] Interactive European Network for industrial crops and Application (2000-2005). Summary Report for the European Union”.
- [6] Fabricant DS and Farnsworth NR (2001) The value of plants used in traditional medicine for drug discovery. Environ. Health perspect. 109 suppl (Suppl 1), 69-75.
- [7] Spolarich, AE and Andrews, L (2007) An examination of the bleeding complications associated with herbal supplements, antiplatelet and anti-coagulant medications. I Dent Hyg. 81(3), 67.
- [8] Nakvinova, J and Anzenbacher, P (2007) Interactions of food and dietary supplements with drug metabolizing cytochrome P450 enzymes. Ceska Slov Farm.. 56(4), 165-73.
- [9] Foste, S (1984) Herbal, Renaissance. Utah: peregrine smith Books
- [10] Elpel, TJ (2004) Botany in a day.
- [11] Therra M (2014) Integrating the traditional Chinese understanding of the kidneys into Western Herbalism Gateway to Chinese Medicine, health and wellnss. Accessed at <http://www.acupuncture.com/herbs/tcmkidney.htm> on September 17th.
- [12] Grieve M.A (2014) Modern Herbal. New York Dover publications, inc. Accessed at http://botanical.com/botanical_19th September.
- [13] Gladstar, R (1993) Herbal Healing for woman. New York: fireside.
- [14] Green, J (2011) The Herbal Medicine, Maker’s Handbook: A Home Manual, Random House LLC.
- [15] Mills S, Bone K (2000) Principles and practices of phytotherapy: Modern Herbal Medicine, Edinburgh: Churchill Livingstone.
- [16] Miskelly, FG an Goodyer, LI (1992) Hepatic and pulmonary complications of herbal medicine. Med J. 68(805), 935-936.

[17] Miller, LG (1998) Herbal medicinal: selected clinical considerations focusing on known or potential drug-herb interaction? Arch Inter Med. 158 (20), 2200-2211.

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