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# Toxicological Evaluation of Dieffenbachia Seguin (Jacq.) Schott (Dumb Cane) on Wister Albino Rats

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## ABSTRACT

*The present study evaluated the toxicological effect of ethanol leaf extract of Dieffenbachia Seguin Schott. Through acute oral toxicity test on wister albino rats. The plant species commonly called Dumb cane is a well known plant in Nigeria, used extensively for ornamental purposes in houses and business areas. It is a poisonous and toxic plant, in the family Araceae. Single oral doses of the leaf extract at the concentrations of 0, 4000, 6000, and 8000mg/kg/day was administered to albino rats to determine the acute toxic effects and the median lethal dose ($LD_{50}$) in rats. For hematological study, blood samples were collected by cardiac puncture and analyzed. The kidney, liver, and heart organs were collected, sectioned and viewed under the microscope for histological study. The results revealed no significant changes in the control group but in the treatment groups, several changes such as fast and labored breathing, blurred vision and death were observed. Hematological study showed reduced blood level in the treatment group. Morphological observation of the organs including the liver showed alteration and paled coloration compared to the control group. Histopathological examination showed distorted, congested and focal necrosis of the liver and renal tubules. The results obtained suggested that the $LD_{50}$ of the ethanol leaf extract of D. Seguin is slightly higher than 8000mg/kg. It is recommended that further studies aimed at corroborating these observations be carried out.*

**Keywords**: *Dieffenbachia Seguin, Ethanol leaf extract, acute oral Toxicity, Araceae, Albino Rats*

## INTRODUCTION

Dieffenbachia seguine, commonly called “dumb cane”, is species of Dieffenbachia native to the tropical Americas from South Mexico, through Central America, to northern South America and Brazil (Resources Conservation Services, 2016).

It is also native to several Carribean islands; including Puerto Rico. The herbaceous perennial grows 3 feet (0.91m) to 10 feet (3.0m) in height and 2feet (0.61m) to 3feet (0.91m) in width (Missouri Botanic Garden, 2011).

The leaves are large and green and often with variegated white patterns. Like other Dieffenbachia, the sap is very toxic. It has showy white flowers (Missouri Botanic Garden, 2011). D. seguine is cultivated as an ornamental plant in temperate shade gardens and as a potted house plant. Cultivars emphasize different patterns of variegations. D. seguine is a perennial herbaceous plant with straight stem, simple and alternate leaves containing white spots and flecks making it attractive as indoor foliage. Species in this genus are popular as house plants because of their tolerance to shade. Its common name, “dumb cane” refers to the poisonous effects of raphides which are needle shaped crystals of calcium oxalate as the monohydrate or calcium carbonate as aragonite, found in more than 200 families of plants. Raphides tend to be blunt at one end and sharp at the other which can cause temporary inability to speak. D. seguine was named by Heinrich Wilhelm Schott, director of Botanical Gardens in Vienna to honor his head gardener Joseph Dieffenbachia (1797-1863). With a minimum temperature of $5^0$c (41F), the plant must be grown indoors in temperate areas.

They need light but filtered sunlight through a window which is usually sufficient. They also need moderately moist soil, which should be fertilized regularly with a proprietary houseplant fertilizer. Leaves will periodically roll up and

fall off to make way for new leaves. Yellowing of the leaves is generally a sign of problematic conditions such as a nutrient deficiency in the soil.

Dieffenbachia responds well to hot temperatures and dry climates. The cells of the Dieffenbachia plant contain needle-shaped calcium oxalate crystals called raphides. If a leaf is chewed, these crystals can cause a permanent burning sensation and erythema (redness of the skin or mucous membrane). In rare cases, edema (abnormal accumulation of fluid) of tissues exposed to the plant has been reported.

Mastication and indigestion generally result in only mild symptoms (Mrvos et al., 1991). With both children and pets, contact with Dieffenbachia (typically from chewing) can cause a host of unpleasant symptoms including intense numbing, oral irritation, excessive drooling, localized swelling and death.

Primarily, it is employed for medicinal and other medicinal miscellaneous uses (Bosch et al., 2002). The stem and root extracts have been described as a narcotic , a gastric and kidney irritant and historically used as arrow poisons by the West Indians in the new world (Cheney, 1931). It is reported to be highly poisonous to humans and when brought into the eye, the sap can cause injury of the cornea and all parts of the plant are very poisonous when ingested (Ardittiaad, 1982).

Dieffenbachia species has invaded most part of the Niger Delta, es[ecially the swampy areas because they flourish well and are able to displace the native species and invade the habitat, This dumb-cane plant can also be found around the First Walkway of Finima Nature Park, managed by the Nigerian Conservation Foundation in partnership with Nigeria LNG in Bonny Island Rivers State, Nigeria. Based on the use of this plant for medicinal and ornamental purposes, the present study was undertaken to evaluate the comprehensive acute toxicity in the animal model and is reported hereunder.

## MATERIALS AND METHODS

### Plant Sample Collection

The leaves of D. Seguin were collected in the month of June from Trans Amadi, off Dr Peter Odili road, Port Harcourt Rivers State, Nigeria and conveyed to the Department of Plant Science and Biotechnology, Rivers State University for proper identification and authentication.

A voucher specimen was deposited in the Department for future reference. The Plant material was then shade dried at a temperature of $30^0$C for a period of 15 days and ground to get a fine powder

### Preparation of the Ethanol Extract

Eighty-eight (88) grams of D. Seguin fine powdered sample was extracted with 1100ml 70% ethanol and allowed to stay for 24 hours before filtration. The brownish liquid obtained was filtered with muslin cloth and the filtrate was further evaporated to dryness in a water bath at 100C.

### Experimental Animals

Wister albino rats were purchased from the University of Port Harcourt, Rivers State, Nigeria. The animals were fed and kept under laboratory conditions for an acclimatization period of 7days before carrying out the experiments.

### Acute Toxicity Study

The acute toxicity of D. Seguin ethanol extract was evaluated in rats (100-120g). Four groups containing three rats each, randomly selected according to their average body weight received ethanol extract at doses of 0, 4000, 6000 and 8000mg/kg body weight, orally after a short fasting period.

The general behavior of the animals was continuously monitored during the first 24 hours (with special attention given within the first four (4) hours and daily thereafter for a total of 14days. The detailed cage-side observations were conducted including changes in eyes, skin and fur, respiratory, autonomic, nervous systems, somatic motor activity and behavior pattern.

Special attention was directed to and observation of convulsions, tremors, diarrhea, salivation, lethargy, sleep and coma. In addition, body weight and food and water intake were recorded at 2-day intervals.

Surviving animals were fasted overnight, then weighed and humanely sacrificed and blood samples collected for hematological analysis.

### Hematological study

Blood samples were collected by cardiac puncture for hematology tests. Full blood count including red blood count (RBC) , hemoglobin

concentration (HGB), white blood count (WBC) packed cell volume (PCV) were evaluated

### Histological Study

For histological study, liver, kidney and heart sections were cut and processed for paraflin sections of 5µm Thickness to be stained with Hematoxylin and Eosin (Marilyn, 2008).

## RESULTS

### Physical and Behavioral Changes

No significant changes were observed in the Wister rats in the control group prior to sacrificing. The wistar rats in this group were as agile and strong as they were at the time they were purchased from the animal farm. There was no loss of appetite and they responded approximately to external stimuli.

But within four hours of administration of the extract, the Wister albino rats in the treatment groups exhibited labored and fast breathing, tremor, restlessness, with some trying to escape through the holes in the cages, loss of appetite (maybe due to swelling of the tongue, blisters in the mouth and itching), general body weakness, dullness, slow response to external stimuli, blurred vision, sluggish movement, itching in the mouth area, loss of fur around the neck region, blisters on the mouth and tongue. Death was recorded for two (2) rats in the treatment group administered with 8000mg/kg body weight within 72 hours of administration of the plant extract, with protruded scrotum.

The degree of observable effects depended on the concentration of ethanol extract administered. The higher the concentration of the extract, the higher the observable effects, though this may be not be conclusive as one rat in group administered with 6000mg/kg exhibited a higher degree of itching than those in group administered with 8000mg/kg.

The symptoms eased in these groups after 6 days and the rats started feeding well, becoming agile and moving freely though the blisters in the mouth and tongue and itching persisted.

Based on these findings, the $LD_{50}$ value of ethanol leaf extract of D. Seguin was determined to be slightly greater than 8000mg/kg body weight (Table 1).

**Table1.** *Acute Oral Toxicity Study of the Ethanol extract of D. Seguin*

| Number of Rats | Doses of extract mg/kg | Number of Rats dead | Percentage of Rats dead |
|---|---|---|---|
| 3 | 0 | 0 | 0 |
| 3 | 4000 | 0 | 0 |
| 3 | 6000 | 0 | 0 |
| 3 | 8000 | 3 | 100 |

### Hematological Examination

The ethanol plant extracts at all doses exhibited a significant decrease in red blood cell count when compared with control (Table 2)

The extract showed reduction in hemoglobin concentration at all doses. However, the reduction was highly significant in the highest dose (8000mg/kg) body weight, followed by 6000mg/kg body weight dose and 4000mg/kg, while the control group showed no significant difference ($P > 0.05$).The control group showed significantly ($P < 0.05$) the lowest value of mean cell volume as compared with the ethanol extracts at all dose levels ($P < 0.05$).

Extracts at all doses significantly elevated mean corpuscular hemoglobin in all groups when compared with control group.

**Table2.** *Hematological data for albino rats used for the study (Dose (mg/kg/day)*

| Parameters | 8000 | 6000 | 4000 | 0 |
|---|---|---|---|---|
| RBC ($10^6$/$mm^3$) | 2.02± 0.02 | 8.08 ± 0.01 | 15.15 ± 0.02 | 28.28 ± 0.02 |
| PCV (%) | 2.00± 0.01 | 8.00± 0.02 | 15.00 ± 0.01 | 28.00 ±0.01 |
| Hemoglobin (g/dl) | 0.06± 0.03 | 0.24 ± 0.01 | 0.45 ± 0.02 | 0.84 ±0.02 |
| WBC | 0.05± 0.3 | 0.04 ± 0.02 | 0.06 ±0.01 | 15.55 ±0.01 |

The ethanol extract groups at all doses showed significant decrease ($P < 0.05$) in packed cell volume in the respective groups.

Reductions in white blood cells (WBC) were also observed in the rats treated with the extract as compared to the control group. Results of the full blood count test carried out on the rats used in the study are summarized in Table 2.

### Histopathological Examination

After sacrificing, some physical changes were observed such as abnormal size and dark coloration, congestion and focal necrosis in the liver and renal tubules, drainage of blood from the heart especially in the treatment group with the highest dose of plant extract.

Administration of *ethanol extract of D. Seguin* showed normal histological features of the kidney in the control group (Group D)

The kidneys of the rats in group A treated with 4000mg/kg body weight of *D. seguine* revealed some level of cyto-architectural distortion of the cortical structures, vascular hypertrophy, interstitial oedema, mild chronic inflammatory infiltrates and hemorrhage as compared to the control.

The kidney sections of rats in group B treated with 6000mg/kg body weight/ day of *D. Seguin* revealed marked distortion of cyto-architecture of the renal cortical structures, and degenerative and atrophic changes. There were tubular necrosis, interstitial hemorrhage, severe chronic inflammatory cell infiltrates, vascular hypertrophy, and vacuolations appearing in the stoma.

The Kidney sections of rats in group C treated with 8000mg/kg body weight/day of D. seguine revealed severe distortion of cyto-architecture of the renal cortical structures, and degenerative and atrophic changes. There were severe tubular necrosis and chronic inflammatory cell infiltrates, interstitial hemorrhage, vascular hypertrophy and vacuolations in the stroma.

The results of histopathological examination of liver section in rats treated with normal saline and ethanol extracts are shown in Figure 20. The liver in rats administered with 0.5 mL normal saline presented the normal hepatic plates and portal vein. For the rats administered with 4000 and 6000 mg/kg body weight of ethanol extract of D. Seguin, there was moderate hepatic necrosis, while 8000 mg/kg body weight of ethanol extract of D. Seguin treated rats showed severe degeneration of hepatocytes.

## DISCUSSION

Herbal products traditionally have been used by general public and traditional healers worldwide to treat various ailments and are considered non toxic. Though Herbal extracts are of natural origin, however, they are not necessarily said to be safe. Plants contain chemicals that are active ingredients that perform the same function like those found in synthetic or orthodox drugs (abayomi, 1999; Ansai and Inamadar, 2010). They may be ineffective in low doses, highly beneficial in the right doses, but may be toxic or have adverse effects in high doses administered for a prolonged period. Medicinal plants possess several biological activities in humans but very little is known regarding their potential toxicity (Rosidah et al; 2009). Toxicity is an indication of a substance being poisonous, pointing to the state of adverse effects led by the interaction between toxicants and cells (Das et al; 2015; Keddy, 2007; Lewis & Elvis-Lewis, 1997). High dosage of some plant extracts has the potential to cause serious toxic effects (D'Arcy, 1991). Therefore in this study, acute oral toxicity studies of the ethanolic extract of the leaves of Dieffenbachia sequin was carried out. These toxicity studies in albino rats are usually necessary for any drug intended for human consumption. These studies are also useful in selecting doses for chronic toxicity studies and providing preliminary identification of target organs of toxicity (Maikai et al; 2008).

In this study, observation of the physical and behavioral changes in the rats administer with various doses of the ethanol plant extract showed adverse reactions such as labored and fast breathing, tremor, restless, with some trying to escape through the holes in the cages, loss of appetite (maybe due to swelling of the tongue, blisters in the mouth and itching), general body weakness, dullness, slow response to external stimuli, blurred vision, sluggish movement, itching in the mouth area, loss of fur around the neck region, blisters on the mouth and tongue within four hours of administration of the plant extract. Death was recorded for two (2) rats in the treatment group administered with the highest dose (8000mg/kg body weight) within 72 hours of administration of the plant extract, with protruded scrotum.

The LD50 (dose of the extract that caused 50 % mortality in the animals) was showed to be 8000 mg/kg. This suggests that the extract was toxic according to a toxicity classification (Saeed et al; 2008).

The result from the study revealed that administration of *ethanol extract of D. Seguin* caused varying degree of cyto-architectural distortion and vasculogenic effect on the kidney which affected blood vessels, hemorrhagic and chronic inflammatory cells appearing in the treatment groups compared to the control group. Narrowing of the lumen also occurred with associated hypertrophic blood vessels and

hemorrhage extending into the interstitium. There were several diffuse degeneration and necrosis of the tubular epithelial cells in the kidneys of the treated animals (4000–8000 mg/Kg). The degenerative and atrophic changes where observed more in the kidneys of rats that received the higher dose (8000mg/kg).

Higher doses of *D. seguine* may have resulted in degenerative and atrophic changes observed in the renal corpuscle. This observation corresponds with the notion that antioxidant is associated with toxicities especially if taken arbitrarily (Miller, 2005). Antioxidants are important relieving oxidative stress, but indiscrete intake of alkaloids and antioxidant constituents of *D. Seguin* may present their toxic effects by inducing oxidative stress (Atici et al., 2005).

From the study, drainage of blood from the heart especially in the treatment group with the highest dose of plant extract was observed and this showed the adverse effect of the plant extract on this organ.

The results from the study showed that liver section administered with 0.5 mL normal saline presented the normal hepatic plates and portal vein. For the rats administered with 4000 and 6000 mg/kg body weight of ethanol extract of D. Seguin, there was moderate hepatic necrosis, while 8000 mg/kg body weight of ethanol extract of D. Seguin treated rats showed severe degeneration of hepatocytes

Estimation of blood parameters is crucial in evaluating the toxicity of drugs as changes in hematological system in animal studies have a high predictive value for human (Olson et al; 2000). The ethanol plant extracts at all doses exhibited a significant decrease in red blood cell count when compared with control. The extract showed reduction in hemoglobin concentration at all doses. However, the reduction was highly significant in the highest dose (8000mg/kg) body weight, followed by 6000mg/kg body weight dose and 4000mg/kg, while the control group showed no significant difference ($P > 0.05$).The control group showed significantly ($P < 0.05$) the lowest value of mean cell volume as compared with the ethanol extracts at all dose levels ($P < 0.05$). Extracts at all doses significantly elevated mean corpuscular hemoglobin in all groups when compared with control group. The ethanol extract groups at all doses showed significant decrease ($P < 0.05$) in packed cell volume in the respective groups. Reductions in white blood cells (WBC) were also observed in the rats treated with the extract as compared to the control group. Results of the full blood count test carried out on the rats used in the study.

## CONCLUSION

In conclusion, the present study suggests that the ethanolic extract of D. Seguin has high toxicity profile regarding hematological and histological parameters in albino rats. It is recommended that caution should therefore be advocated in the use of this plant for ornamental purpose in homes to avoid accidental intake of this product and further studies be carried out to examine these findings.

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