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Acute Oral Toxicity Study Of The Total Aqueous Extract Of The Dry Bark Of The Trunk Of Albizia Ferruginea (Mimosaceae) (Guill. & Perr.) Benth In Wistar Albino Rats

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Abstract

Objective: The study conducted aimed at contributing to the assessment of the acute oral toxicity of the total aqueous extract of the dry bark of the trunk of Albizia ferruginea (Mimosaceae) (Guill. & Perr.) Benth in Wistar albino rats.

Material and methods: The acute oral toxicity of the total aqueous extract of the dry bark of the trunk of Albizia ferruginea was assessed according to OECD Guideline 420 on 15 male Wistar albino rats. After 12 h of fasting, they were divided into 3 groups of 5 rats each as follows: a control group receiving distilled water (10 mL/kg); two test groups each receiving a single dose of the total aqueous extract of the dry bark of the trunk of Albizia ferruginea (2000 mg/kg and 5000 mg/kg respectively). The behavioral reactions were observed for 4 hours and the rats were left under observation for 14 days to determine possible cases of death, weight development, water and food intake and the relative weight of various vital organs.

Results: Oral administration of a single dose of 2000 mg/kg or 5000 mg/kg of the total aqueous extract of the dry bark of the trunk of Albizia ferruginea to rats did not cause any significant change in rat behavior or death. The LD_{50} was therefore estimated to be greater than 5000 mg/kg. At each of the above doses, there was no change in weight development, water and food intake and the relative weight of the different organs (liver, lungs and kidneys) during the 14 days of the experiment.

Conclusion: The study showed that the total aqueous extract of the dry bark of the trunk of Albizia ferruginea has an LD_{50} greater than 5000 mg/kg body weight and may be devoid of toxic effects. It would therefore be favorable for the production of an improved traditional drug after preclinical and clinical trials.

Keywords – Albizia ferruginea, acute toxicity, traditional drug, dry bark, LD50, rats.

I. INTRODUCTION

Toxicity is the set of harmful effects caused by a substance introduced into a living organism at a single relatively high dose or at long-repeated small doses [**1**]. Its study is the set of pharmacological tests, which determine the degree or the harmfulness of the latter in order to regulate its use. The action of a toxic substance is evaluated according to several parameters including its mode of administration (oral, intravenous, intraperitoneal), the dose administered, the observed mortality rate, weight change, histology of certain organs etc. [**2**]. Plants are a valuable source of natural products for therapeutic purposes [**3**]. Their use grew

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rapidly and has become very popular. Indeed, the World Health Organization (WHO) has found that about 80% of the developing countries' populations are handicapped to affording pharmaceutical drugs; rely on traditional medicines, mainly from plants, to sustain their primary health care needs ([4**],[5**]). Traditional systems of medicine are popular in developing countries with up to 80% of the population relying on traditional medicines or folk remedies for their primary health care needs [**6**]. Medicinal plants can therefore constitute important resources for new substances with therapeutic potential and at lower costs. The traditional use of any plant for medicinal purposes requires safety guarantees for the treatment of that plant.

A. ferruginea is a species of plant in the Mimosaceae family [7], found in Angola, Benin, Cameroon, Republic of Congo, Nigeria, Senegal, Togo, Uganda among others [8]. This species threatened by deforestation is widespread in west and central Africa [9]. It is called 'Evouvous' by the Ewondo tribe in the Central region of Cameroon. It is also called ''Ugeehu'' in Abakaliki dialect of Ebonyi State, Nigeria [**10**]. Traditionally, in the Centre, Littoral and Southern regions of Cameroon, the stem bark of *A. ferruginea* are used to treat diarrhea, rheumatism, abdominal and dental pain, headache, bronchitis, dysentery, hemorrhoids and to relieve inflammatory pain due to fever [**11**]. In Central Africa, a juice made from the leaves of *A. ferruginea* is used as an emollient to soothe rashes, swelling, boils and carbuncles, the leaves are used to treat malaria **[12**]. A leaf decoction is used as a lotion or as a vapor inhalation against fever, headaches and toothaches [**13**]. Previous pharmacological studies carried out on the plant have shown that the ethanolic extract of the leaves of *A. ferruginea* corrects anemia [**14]** and the aqueous extract of the dry bark of the trunk of *A. ferruginea* was able to reduce inflammation, pain and pyrexia [**15**]. There is almost no information on the harmlessness of this plant. The main objective of this study was to assess the acute toxicity of the total aqueous extract of the dry bark of the trunk of *A. ferruginea*.

II. Materials And Methods

2.1 Collection and Extraction of Plant Materials

The stem barks of *A. ferruginea* were harvested from Angallé village in the South Region of Cameroon. The plant materials were identified by Dr Barthélémy TCHIENGUE of the National Herbarium of Cameroon, where a voucher specimen of the plant was deposited under the number 49871. Fresh stem barks were air-dried and reduced to a fine powder. The powder (500 g) was macerated with 2.5 L of distilled water for 24 hours. The mixture was filtered with Whatman N°3 filter paper, concentrated under reduced pressure and lyophilized at 50°C for 48 hours. A dark brown solid (84 g) representing the stem barks aqueous extract of *A. ferruginea* was obtained (yield of 16.8%).

2.2 Qualitative and quantitative phytochemical analysis

The qualitative phytochemical investigations of the stem bark aqueous extract of *A. ferruginea* were performed for alkaloids, flavonoids, saponins, phenols, steroids, glycosides and tannins, by our research team (**[16],[17]**) using standard methods previously described (**[18], [19]**).

2.3 Experimental Animals

Male albino Wistar rats (200-250 g) were obtained from the animal house unit of the Faculty of Science of the University of Yaounde I, Cameroon. They were maintained under standard environmental conditions with a dark and light circle of 12/12h. They were fed with standard commercial diet and water was provided ad libitum. The experimental protocol was in conformity with guidelines of the Cameroon National Ethical Committee on the use of laboratory animals for scientific research (CEEC Council 86/609).

2.4 Acute oral toxicity test of the total aqueous extract of the dry bark of Albizia ferruginea

The acute oral toxicity test of the total aqueous extract of the dry bark of *A. ferruginea* was evaluated according to OECD Guideline 420 on 15 male Wistar albino rats over a period of 14 days. After 12 h of fasting, they were divided into 3 groups of 5 rats as follows: a control group receiving distilled water (10 mL/kg); two test groups each receiving a single dose of the total aqueous extract of the dry bark of *A. ferruginea* (2000 mg/kg and 5000 mg/kg respectively). Behavioral reactions were observed for 4 h and the rats were left under observation for 14 days to determine possible cases of death. Observations focused on symptomatic disturbances seen with the naked eye, including changes in the skin, hair, eyes and mucous membranes. Attention was also paid to manifestations of tremor, convulsion, salivation, diarrhea, lethargy, sleep and coma **[20].** The mortality rate was

determined after 24 hours and then during the 14 days of observations. The weight variations compared to the 1st day were expressed as a percentages (%) according to the following formula:

$$\% P = \frac{\overline{Pj} - \overline{Pj_0}}{\overline{Pj_0}} \times 100$$

% P = weight percentage

Pd0 = body weight on the 1st day

Pd = body weight on day d

During the experimentation period (14 days), parameters such as water and food consumption, and weight change were noted every two days. On day 14, the animals were fasted again. On day 15, the animals were sacrificed by decapitation and the gross appearance of the liver, heart, kidneys, lungs, spleen and digestive tract was assessed. The relative weight of the organs removed was evaluated.

2.5. Statistical Analysis

All the results were expressed as Mean ± SEM. The data were statistically analysed by one-way ANOVA, followed by Dunnett's test using Graph pad prism (5.03) software. P values less than 0.05 were considered statistically significant.

III. RESULTS

3.1. Effects of total aqueous extract of dry bark of Albizia ferruginea on the behavioral reactions and morbidity rate

Oral administration of a single 2000 mg/kg dose of the total aqueous extract of the dry bark of *A. ferruginea* to rats did not cause significant changes in behavior in treated rats compared to control rats. At a dose of 5000 g/kg, a decrease in locomotion was observed (Table 1). No deaths were observed at doses of 2000 and 5000 mg/kg. The LD_{50} of the aqueous extract of the dry bark was therefore estimated to be greater than 5000 mg/kg according to the general and harmonized system (GHS) of the OECD.

Table 1: Effects of the total aqueous extract of the dry bark of *Albizia ferruginea* on the behavioral reactions of rats during acute toxicity

Observations	Rats		
	Controls	2000 mg/kg	5000 mg/kg
Salivation	A	A	A
Appearance of faeces	G	G	G
Coat appearance	N	N	N
Locomotion	N	N	D
Sleep	A	A	A
Coma	A	A	A

A= Absent; D=Diminished; G: Granular; N=Normal. n= 5.

3.2 Effects of the total aqueous extract of the dry bark of *Albizia ferruginea* on food and water consumption

The total aqueous extract of the dry bark of *A. ferruginea* at doses of 2000 mg/kg or 5000 mg/kg did not induce a significant change in food and water consumption in the treated rats compared to the control rats (Table 2).

Table 2: Effects of the total aqueous extract of the dry bark of *Albizia ferruginea* on food and water consumption during acute toxicity

	Temps (Week)	Control	2000 mg/kg	5000 mg/kg
Water intake (mL/rat/day)	1	24.4±1.5	27.5±2.1	28.6±1.1
	2	31.4±2.0	33.9±2.0	39.1±2.8
Foot intake (g/rat /day)	1	34.7±3.3	32.6±3.9	36.7±2.9
	2	40.6±1.6	39.7±1.6	38.3±1.6

Values represent means ± ESM

3.3 Effects of the total aqueous extract of the dry bark of *Albizia ferruginea* on the weight development of rats

No significant modification was observed in the evolution of weight during the 14 years of the experiment following the administration of the single dose of 2000 mg/kg or 5000 mg/kg of the total aqueous extract of the dry bark of *A. ferruginea* in treated rats compared to controls rats.

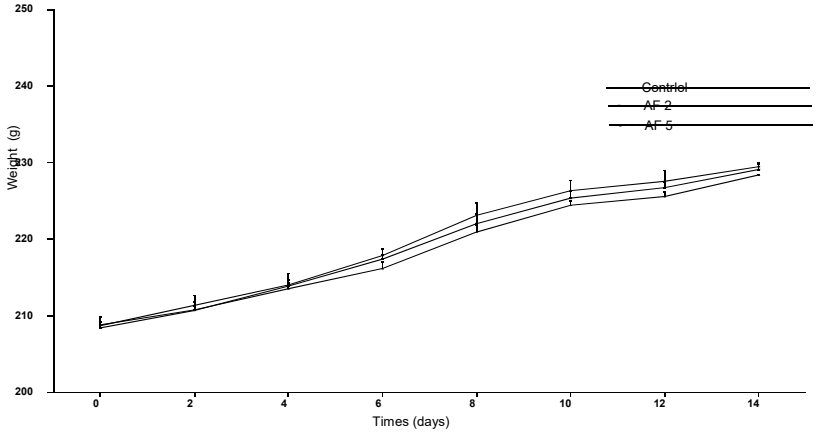

Figure 1: Effects of the total aqueous extract of the dry bark of *Albizia ferruginea* on the weight development of rats during acute toxicity.

Each point represents *the mean weight ± ESM, n = 5; Each bar represents the standard deviation of the mean of 5 animals per group. AF = aqueous extract of Albizia ferruginea, AF 2 = dose of 2000 mg/kg and AF 5 = dose of 5000 mg/kg.*

3.4 Effects of total aqueous extract of *Albizia ferruginea* on the relative weight of detoxification organs

At the end of the experimental period (14 days), the relative weight of the different organs (liver, lungs and kidneys) did not vary significantly following the administration of the single dose of 2000 mg/kg or 5000 mg/kg of the total aqueous extract of the dry bark of A. ferruginea in the treated rats compared with the control rats (Figure 2).

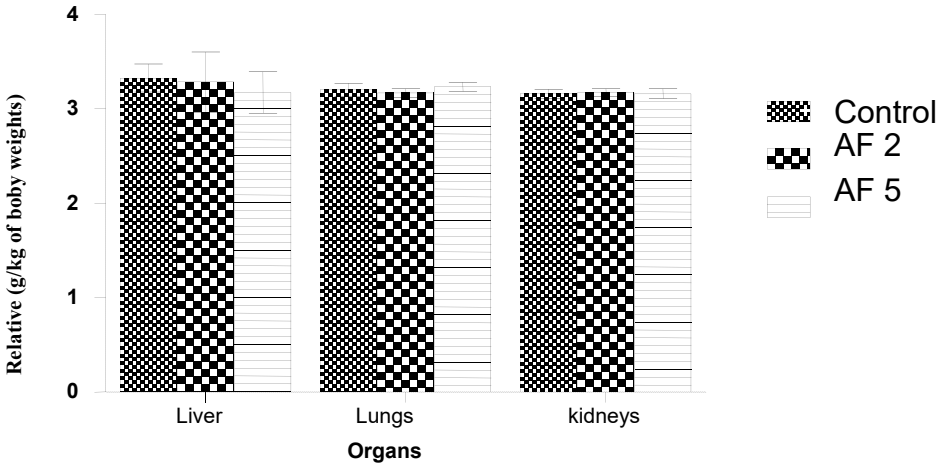

Figure 2: Effects of total aqueous extract of *Albizia ferruginea* on the relative weight of detoxification organs during acute toxicity.

Each column represents the mean weight ± ESM, n = 5; Each bar represents the standard deviation of the mean of 5 animals per group. AF = total aqueous extract of Albizia ferruginea, AF 2 = dose of 2000 mg/kg and AF 5 = dose of 5000 mg/kg.

IV. Discussion

The acute toxicity test evaluates the adverse effects that occur within a short period of time after administration of a single dose of a test substance. It is performed primarily on rodents and usually early in the development of a new product to provide information on its potential toxicity [**21**]. The acute oral toxicity study of the total aqueous extract of the dry bark of *A. ferruginea* in rats at a single dose of 2000 mg/kg or 5000 mg/kg showed an absence of mortality in rats after the 14 days of observation. This implies estimating that the LD_{50} is greater than 5000 mg/kg of body weight. These results corroborate those of Sarkiyayi et al. who estimated the LD_{50} of the ethanoic extract of the leaves of *A. ferruginea* to be greater than 5000 mg/kg [**22**]. According to the globally harmonized classification system of the OECD [**20**], the total aqueous extract of the dry bark of *A. ferruginea* can be classified in category 5 and considered as an oral non-toxic substance. This same method was used in 2007 by Adeneye and Agbaje, who showed that the LD_{50} of the aqueous extract of *Cymbopogon citratus* (Poaceae) is greater than 5000 mg/kg body weight [**23**]. Koné and collaborators in 2009 by the same method, showed that the LD_{50} of the total aqueous extract of *Sacoglottis gabonensis* is greater than 5000 mg/kg body weight [**24**]; as well as Lebri and colleagues, who in 2015 showed that the LD_{50} of the total aqueous extract of the leaves of *Abrus precatorius* Linn (Fabaceae) taken orally in rats is greater than 5000 mg/kg body weight. The change in body weight is an important index for the assessment of toxicity [**25**]. The administration of a single dose of the total aqueous extract of the dry bark of *A. ferruginea* (2000 mg/kg or 5000 mg/kg) did not induce a noticeable change in behavior, neither did it cause significant changes in food consumption and water and weight changes in rats at the end of the experimental period (14 days). Macroscopic pathological examinations of the liver, lungs and kidneys of the treated groups showed no major visual differences in terms of size, shape, color and texture compared to the control group.

V. Conclusion

This study showed that the total aqueous extract of the dry bark of *Albizia ferruginea* is non-toxic at doses tested orally in Wistar rats. The LD_{50} is estimated to be greater than 5000 mg/kg body weight. These results would be in favor of its safety by the oral route in the traditional treatment of certain diseases. However, other work such as the in-depth research of the effect of this extract on certain target organs (liver, kidney, heart), by assaying serum biochemical parameters in subacute or chronic oral toxicity deserves attention and need to be conducted in order to conclude on its non-toxic nature. Scientific evaluation of

traditional plants and their method of use in disease management can enable their integration into the formal health system in Africa and other developing countries.

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ETHICAL APPROVAL

As per international standard or university standard, written ethical approval has been collected and preserved by the authors.

COMPETING INTERESTS

Authors declare that no competing interests exist.

ABBREVIATIONS

A. ferruginea: Albizia ferruginea

LD_{50}: Dose lethal to 50% of animals

OECD: Organisation for Economic Cooperation and Development

EEC: European Economic Community

REFERENCES

[1] Etame LG, Yinyang J, Okalla EC, Makondo BV, Ngaba GP, Mpondo ME, Dibong SD. (2017). Étude de la toxicité aiguë et subaigüe de l'extrait au vin des graines de Carica papaya Linn. Journal of Applied Biosciences **120** : 12077-12085.

[2] Serrano JJ .(1990). Toxico–pharmacologie expérimentale des plantes médicinales. Actes du 1er colloque européen d'ethnopharmacologie. Office de la recherche scientifique d'outre-mer (ORSTOM). Pp. 210-218

[3] Offor CE. (2015). The effects of ethanol leaf-extract of Mucuna pruriens on haemoglobin level and packed cell volume in albino rats. Journal of Research in Pharmaceutical Science, **2**(6): 3.

[4] Rudy AN, Retno A, Hetty M, Rudianto R, Widha P, Amelia J, Abdul KA, Nur AOP et Agnesia L .(2020). Acute and Subchronic Toxicity Study of the Ethanol Extracts from Ficus deltoidea Leaves in Male Mice. (2020). Macedonian Journal of Medical Sciences, **8**(A):76-83.

[5] Goyal BR, Goyal RK et Mehta AA .(2008). Phyto-pharmacognosy of Archyranthes aspera: a review. Pharmacognosy Reviews, **1**(1): 143-150.

[6] Edgar JD, Elias B et Adnan B. (2002). Biotechnology and the developing world. Electronic Journal of Biotechnology, **5**(1): 543-548.

[7] Lakouéténé DPB, Ndolngar G, Berké B, Moyen JM, Kosh Komba E, Zinga I, Silla S, Millogo-Rasolodimby J, Vincendeau P, Syssa-Magalé JL, Nacoulma-Ouedraogo OG, Laganier R, Badoc A, Chèze C. (2009). Enquête ethnobotanique des plantes utilisées dans le traitement du paludisme à Bangui. Bulletin de la Société de Pharmacie Bordeaux, **148** : 123-138.

[8] Ukpabi SEN et Offor CE. (2018). Phytochemical, vitamin and mineral compositions of ethanol leaf-extract of Albizia ferruginea and its proximate composition. Caribbean Journal of Science and Technology, **6**(1): 029-035.

[9] Kareru PG, Kenji GM, Gachanja AN, Keriko JM et Mungai G. (2007). Traditional medicines among the Embu and Mbeere peoples of Kenya. African Journal of Traditional, Complementary and Alternative Medicines, **4**: 75-86.

[10] Noumi E, Eboule AF et Nanfa R. (2011). Traditional health care of male infertility in Bansoa, West Cameroon. Journal of Pharmaceutical and Biomedical Sciences, **2**: 42-50.

[11] Jiofack T, Ayissi I, Fokunang C, Guedje N et Kemeuze C. (2009). Ethnobotany and phytomedicine of the upper Nyong valley forest in Cameroon. African Journal of Pharmacy and Pharmacology, **3**: 144-150.

[12] Burkill HM. (1995). The useful plants of West Tropical Africa. 2nd Edition.Volume 3, Families J-L. Royal Botanic Gardens, Kew, Richmond, United Kingdom, 857 P.

[13] Adjanohoun EJ, Adjakidjè V, Ahyi MRA, Aké AL, Akoègninou A, d'Almeida J, Apovo F, Boukef K, Chadare M, Cusset G, Dramane K, Eyme J, Gassita JN, Gbaguidi N, Goudote E, Guinko S, P. Houngnon, Lo I, Keita A, Kiniffo HV, Kone-Bamba D, Musampa NA, Saadou M, Sodogandji T, De Souza S, Tchabi A, Zinsou CD et Zohoun T. (1989). Contribution aux études ethnobotaniques et floristiques en République populaire du Bénin. Agence de Coopération Culturelle et Technique, Paris, France ; 1989.

[14] Ukpabi SEN, Offor CE, Udeozor PA, Obiudu IK. (2018). Effects of Ethanol Leaf-Extract of *Albizia ferruginea* on Selected Haematological Indices in Wistar Albino Rats. *Journal of Science and Technology*, **3** (1):74-81.

[15] Minoue Kumm MG, Fotio Lambou A, Atsang A. Kiki G, Bella Ndzana MT, Keugni BA, Moukette B, Mezui C, Dzeufiet Djomeni PD et Dimo T. (2018a). Evaluation of Anti-inflammatory, Analgesic and Antipyretic Potential of the Stem Barks Aqueous Extract of Albizia ferruginea (Guill. & Perr.) Benth. (Mimosaceae) in Rats and Mice. Journal of Advances in Biology & Biotechnology, **20**(3): 1-15.

[16] Minoue Kumm MG, Temdie GRJ, Bella Ndzana MT, Tchadji JC, Lissom A, et Dimo T. (2018b). Albizia Ferruginea (Mimosaceae) on Chronic Inflammation Induced in Rats. International Journal of Innovative Research in Medical Science (IJIRMS), **3** (9): 2183-2195.

[17] Minoue Kumm MG, Tchadji JC, Atsang A. Kiki G, Temdie GRJ et Dimo T. (2021). Evaluation of the anti-inflammatory effects of the aqueous extract of the bark of Barteria fistulosa. World Journal of Pharmacy and Pharmaceutical Sciences, **10** (10):1-18.

[18] Odebeyi OO et Sofowora EA. (2006). Phytochemical screening of Nigeria plants 2. *LLoydia*, **41**: 234-238.

[19] Tanko Y, Kamba B, Saleh M, Musa KY et Mohammed A. (2008). Anti-nociceptive and anti-inflammatory activities of ethanolic flower extract of *Newbouldia laevis* in mice and rats. *International Journal of Applied Research in Natural Products*, **1**: 13-19.

[20] OCDE. (2001). Toxicité orale aiguë-Méthode de la dose prédéterminée. In Lignes directrices de l'OCDE pour les essais de produits chimiques n°420. OCDE, Paris, **1** (4):1-15.

[21] Million L, Abay M, Solomon MA, Wondwossen E et Bekesho G (2019). Acute and Subacute Toxicity of Methanol Extract of Syzygium guineense Leaves on the Histology of the Liver and Kidney and Biochemical Compositions of Blood in Rats. Evidence-Based Complementary and Alternative Medicine, 1-16.

[22] Sarkiyayi S, Karago J et Hassan M. (2011). Studies on anti-typhoid properties of aqueous methanol leaves extract of Albizia ferruginea (Musase). International Journal of Biochemistry Research and Review, **1** (1):24-30.

[23] Adeneye AA et Agbaje EO (2007). Hypoglycemic and hypolipidemic effects of fresh leaf aqueous extract of Cymbopogon citrates Stapf. in rats. Journal of Ethnopharmacology , **112**(3): 440-444.

[24] Koné M, Bleyere NM, Yapo AP, Vangah MO et Ehilé EE. (2009). Evaluation de la toxicité d'un extrait aqueux de Sacoglottis gabonensis (Baille) Urban (Humiriaceae) chez les rongeurs, une plante utilisée dans le traitement de l'ulcère de Buruli en Côte d'Ivoire. International Journal of Biological and Chemical Sciences, **3**(6): 1286-1296.

[25] Lebri M, Bahi C, Fofie NBY, Gnahoue G, Lagou SM, Achibat H, Yapi A, Zirihi GN, Coulibaly A, Hafid A et Khouili M. (2015). Analyse phytochimique et évaluation de la toxicité aiguë par voie orale chez des rats de l'extrait total aqueux des feuilles de Abrus precatorius Linn (Fabaceae). International Journal of Biological and Chemical Sciences, **9**(3) : 1470-1476.