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(RESEARCH ARTICLE)

Acute toxicity of the methanol extract of *Ficus stuhlmannii* Warb. (Moraceae) leaves

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Abstract

Ficus stuhlmannii Warb. is a medicinal plant used in Africa to treat sickle cell anaemia and various other conditions. The aim of this study was to evaluate the acute toxicity of the methanolic extract of *Ficus stuhlmannii* Warb. leaves in mice, to contribute to the safety of human consumers. The methanolic extract was obtained after extraction with solvents of increasing polarity and then administered to mice. Its effects on the behaviour, general condition and mortality of the mice were observed. Biochemical and hematological parameters were measured by spectrophotometry. No signs of toxicity were noted after administration of 5000 mg/kg body weight of the methanolic extract of *Ficus stuhlmannii* Warb. leaves. All animals survived after 14 days of observation, indicating that the LD_{50} is greater than 5000 mg/kg body weight. Variation in biochemical and haematological parameters is an indicator of acute toxicity. There were no significant differences ($p>0.05$) in the biochemical parameters between the negative control and the treated groups: dose at 1000 to 500mg/kg, all values found for these parameters were within the normal range. Thus, no significant difference ($p>0.05$) was observed in the haematological parameters except for white blood cells, where there was a significant difference ($p <0.0001$) between the control and the groups treated with doses of 2000 to 5000 mg/kg. The results obtained show that the methanolic extract of *Ficus stuhlmannii* Warb. leaves are not toxic to treated mice, even at a concentration of 5000 mg/kg.

Keywords: Acute Toxicity; *Ficus stuhlmannii* Warb; Traditional Medicine; *Mus Musculus*

1. Introduction

The use of medicinal plants in the treatment of diseases dates back several millennia. Since ancient times, humans have used plants from their environment to treat various diseases [1,2]. However, the belief that natural remedies are necessarily beneficial and that certain diseases such as AIDS, hepatitis, and cancer currently benefit from treatments that are not entirely effective. These treatments are accompanied by significant side effects and are increasingly pushing people towards traditional medicine [3].

However, recent years have been revealed that herbal medicine is not without risk. Toxic effects affect most organs, including plant-induced renal failure, cardiotoxicity linked to aconite, and pneumonia induced by certain mints [4,5]. We now know that a plant has medicinal properties thanks to its active ingredients, which can treat a specific condition.

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In fact, any biologically active substance is likely to produce undesirable or even harmful effects when taken in high or low doses over a prolonged period. Safety of use must be the primary criterion when choosing medicinal plants. For these reasons, ethnopharmacological studies always place great importance on knowledge of plants that are of interest to human health due to their therapeutic or toxic properties. It is in this context that we wanted to contribute to the study of the leaves of *Ficus stuhlmannii* Warb., which belongs to the Moraceae family [6]. This species has been shown to be effective in straightening the sickle-shaped red blood cells that cause sickle cell anemia and numerous crises in sickle cell patients [7,8]. The specific objective of this study is to evaluate, in vivo, the acute toxicity of *Ficus stuhlmannii* Warb. leaves in *Mus musculus*

2. Materials and Methods

2.1. Plant material

The plant material used in this study consisted of leaves from *Ficus stuhlmannii* Warb, collected on March 12, 2023, in the sparse forest along the Kasenga road, 20 km from the city of Lubumbashi. A specimen was prepared in the form of an herbarium and deposited at the herbarium of the National Institute for Agronomic Study and Research (NIASR/Kipopo), where its identity was confirmed by comparison with reference herbariums. This specimen was registered under the number KIP390223364. The leaves of *Ficus stuhlmannii* warb were dried away from sunlight in a ventilated room at room temperature; after drying, the plant material was pulverized using an electric grinder.

2.2. Animal material

Swiss albino mice (*Mus musculus*) were used as an animal model for the evaluation of subacute toxicity in vivo. These healthy animals weighed between 50 and 245 grams and were between 6 and 10 weeks old. They were purchased from local breeders located on the outskirts of the city of Lubumbashi. After transport to the experimental laboratory, the mice were housed in appropriate cages with free access to standard feed and tap water. They were kept under control of environmental conditions: a 12-hour light/dark cycle and an ambient temperature of 26 ± 1 °C.

2.3. Harvesting and identification of the plant

The plant material consists of leaves from *Ficus stuhlmannii* Warb., which were harvested in Mikembo Sanctuary Park as shown in Figure 1 below.

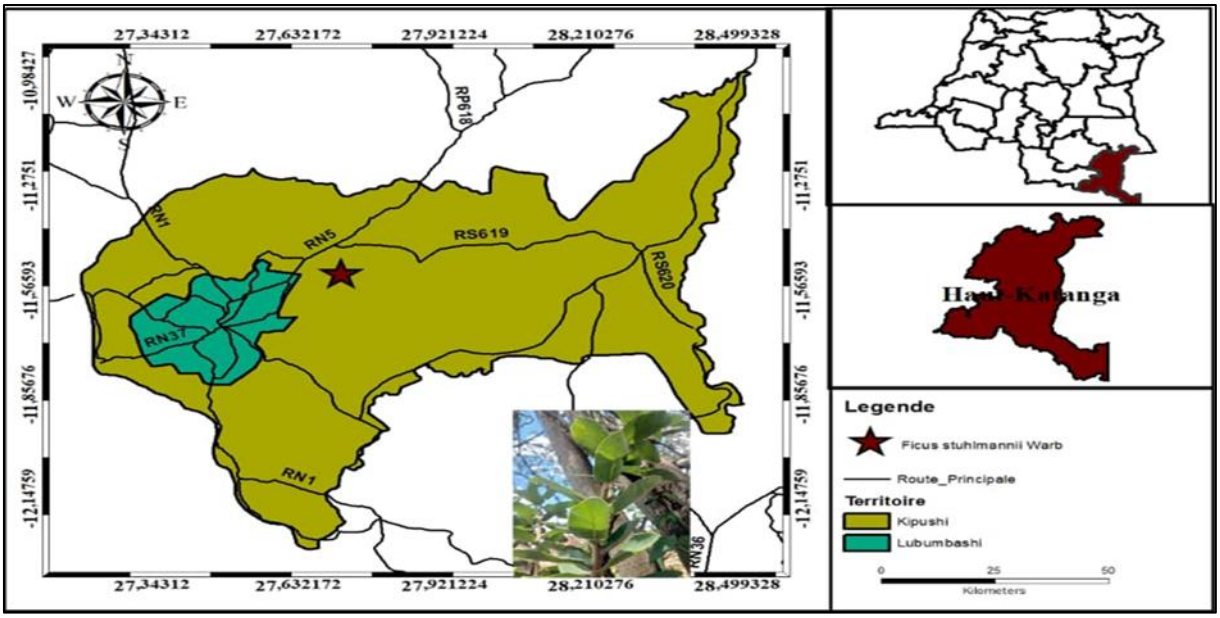

Figure 1 Site where *Ficus stuhlmannii* Warb. leaves were harvested.

The collection and identification of *Ficus stuhlmannii* Warb. leaves were carried out at a site located at an altitude of 1,223 meters, at the following geographical coordinates: latitude 11.85676° south and longitude 27.34312° east. The harvest was followed by drying and storage of the drug away from sunlight at room temperature, then pulverization using an electric mill (HC-500) at 5000 rpm.

2.4. Extraction

Various extracts were obtained from dried, powdered *Ficus stuhlmannii* Warb. leaves, as shown in Figure 2. Solid-liquid extraction was performed by maceration using four solvents of increasing polarity to gradually exhaust the plant material. Depending on the solvent used, four extracts were obtained: petroleum ether (PEE), dichloromethane (DCME), ethyl acetate (AcOEtE), and methanol (MeOHE) extracts. Specifically, 500 g of *Ficus stuhlmannii* Warb. leaf powder was macerated twice in 2.5 L of petroleum ether for 24 hours, and the same operation was repeated for the other solvents: dichloromethane, ethyl acetate, and methanol. The solutions obtained were filtered and then concentrated using a rotary evaporator at a temperature below 50°C. The protocol used is summarized in Figure 2 below.

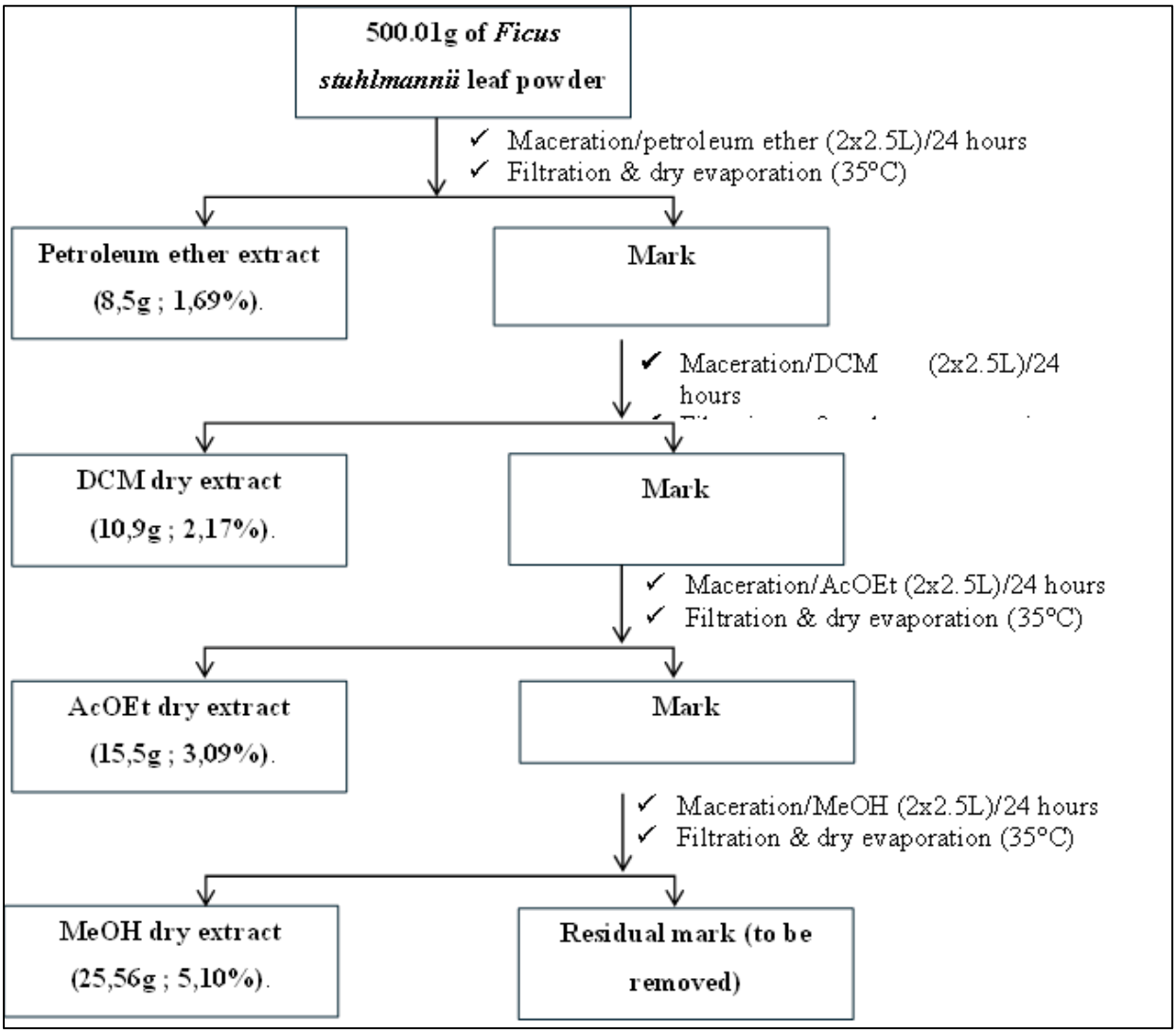

Figure 2 Solid-liquid extraction diagram for *Ficus stuhlmannii* Warb. leaves

The acute toxicity study will focus on the methanolic extract, which was selected based on data from previous studies [7,9–11].

2.5. Preparation of the solution to be administered.

A stock solution was prepared from the methanolic extract. Each time, this stock solution was prepared at a single concentration: 40mg/mL, by dissolving 12 g of the dry extract in 300 mL of distilled water. This concentration allowed the acute toxicity study to be conducted for animals that were to receive the extracts at doses of 1000, 2000, 3000, 4000 and 5000 mg/kg of body weight, respectively. The volume to be administered by gavage was calculated taking into account the animal's weight in kg, the dose of extract administered expressed in mg/kg of body weight, and the concentration in mg/mL using the mathematical relationship below [12,13]

$$\textbf{Volume (mL)} = \frac{\text{Dose}\left(\frac{g}{Kg}\right) \text{x Weight (kg)}}{\text{Concentration}\left(\frac{g}{mL}\right)}$$

2.6. Treatment of animals

The animals used were white mice weighing between 50 and 245 g, supplied by breeders. These mice were placed in cages and had free access to water and food. The solutions administered were prepared extemporaneously and the doses of extract used for toxicity assessment are expressed in mg of dry extract per kg of body weight of each animal [14,15].

2.7. Acute toxicity assessment

The toxicity study was conducted using the method described by [16] which consists of administering increasing doses of the substance to groups of mice of similar weight. The dose administered is expressed in mg/kg or mL/kg of body weight, and the difference between adjacent doses must be constant. For each group, the percentage of mortality within one hour or at the end of the allotted time is recorded. The study of subacute oral toxicity, with a view to identifying clinical and/or biological signs of poisoning, was carried out according to the protocol described by Musa et al. [17] with a few minor modifications.

The acute toxicity study was conducted on a total of 18 mice, divided into six groups of three animals each. Prior to administration of the substances, the mice were fasted for 12 hours, with free access to water, in accordance with standard protocols. Each group was then treated according to a specific experimental regimen, detailed below:

- Group I: mice received physiological solution (0.9% NaCl) at a volume of 2 mL/kg body weight;
- Group II: mice received the extract at a dose of 1000 mg/kg body weight;
- Group III: mice received the extract at a dose of 2000 mg/kg body weight;
- Group IV: mice received the extract at a dose of 3000 mg/kg body weight;
- Group V: mice received the extract at a dose of 4000 mg/kg body weight;
- Group VI: mice received the extract at a dose of 5000 mg/kg body weight.

We used concentrations close to those used by traditional healers, based on the approximate calculation model for herbal medicines proposed by Chiffoniere [18].

All treatments were administered by gastric gavage in a single daily dose, according to the batches, for 14 days. The mice were observed for signs of poisoning or clinical signs (feeding, locomotion, general appearance, abdominal condition). Immediately after administration of the substances at different doses according to the batches, the mice were deprived of food for 1 hour. In addition, their body weight was measured at the end of each week throughout the treatment period.

At the end of the treatment period, the mice were sacrificed. Blood samples were taken on the 15th day using a syringe in the jugular vein after puncturing the area behind the mandibular angle on each mouse. The blood sample was collected directly into two tubes containing 2 mL of blood each, one containing an anticoagulant, ethylenediaminetetraacetic acid (EDTA), and the other without anticoagulant, to measure hematological and biochemical parameters indicative of toxicity, respectively.

The blood collected in this way was centrifuged to obtain plasma for measuring biochemical parameters, including transaminases (ALT, AST), urea, and creatinine. The principles and operating procedures are presented below.

2.8. Data analysis

Statistical tests and graphs were performed using GraphPad software version 8. Results are presented as mean ± standard deviation. The data were evaluated using ANOVA (one-way analysis of variance) followed by Tukey's multiple comparison test at a 5% threshold to assess the significance of the differences observed. If $P<0.05$, the difference between the values is considered significant, and if $P>0.05$, the difference is not significant.

3. Results

3.1. Evaluation of behavioral change after gavage with methanolic extract from *Ficus stuhlmannii* Warb leaves and during the 14-day observation period

After administration of the methanolic extract, the animals were returned to their metal cages where they had access to food and water. They were observed immediately after administration of the product, then every 30 minutes thereafter, and regularly for 14 days. During this period, no signs of acute toxicity were noted in the animals in the groups. Immediately after administration of the extract, we observed partial hypoactivity and decreased appetite in the treated mice, but after a few minutes the animals resumed their movements. These signs were observed in all groups (table I).

Table 1 Signs of acute toxicity observed after oral administration of the extract (n=3)

Extract	Batch	Dose (mg/Kg)	Number of deaths	Signs of acute toxicity
Physiological solution (NaCl 0.9%)	I	0	0/3	None
MeOHE	II	1000	0/3	None
MeOHE	III	2000	0/3	None
MeOHE	IV	3000	0/3	None
MeOHE	V	4000	0/3	None
MeOHE	VI	5000	0/3	None

3.2. Changes in the body weight of the animals

The animals given the extract, as well as those in the control group, all showed a positive weight gain during the 1-day to 7-day administration period and a slight weight loss on the 14th day (Table II).

Table 2 Variation in average animal weights during acute toxicity assessment

	Dose (mg/kg)	Day 1	Day 7	Day 14
Physiological solution (NaCl 0.9%)	0	166.33 ± 12.22^{ns}	169.33 ± 10.44^{ns}	168.00 ± 12.00^{ns}
MeOHE	1000	141.33 ± 3.06^{ns}	145.67 ± 3.06^{ns}	138.00 ± 6.24^{ns}
MeOHE	2000	153.00 ± 6.67^{ns}	157.00 ± 7.33^{ns}	147.00 ± 8.00^{ns}
MeOHE	3000	166.33 ± 9.11^{ns}	170.00 ± 10.00^{ns}	157.67 ± 8.00^{ns}
MeOHE	4000	151.33 ± 6.22^{ns}	160.67 ± 4.44^{ns}	143.67 ± 11.11^{ns}
MeOHE	5000	127.00 ± 11.33^{ns}	129.67 ± 9.78^{ns}	126.67 ± 11.33^{ns}

ns: no-significant difference ($p > 0.5$)

3.3. Biochemical profile of *Mus musculus* after administration of high doses of methanolic extract from the leaves of *Ficus stuhlmannii* Warb.

This section will discuss the results of the biochemical parameters of acute toxicity of the methanolic extract of *Ficus stuhlmannii* leaves, namely urea, creatinine, direct bilirubin, indirect bilirubin, alanine aminotransferase, aspartate aminotransferase and alkaline phosphatase in treated mice and controls (Table III).

Table 3 Biochemical profile of control *Mus musculus* and treated groups (n=3)

Markers	**Standards [19–21]**		**Dose mg/kg**				
		Control	**1000**	**2000**	**3000**	**4000**	**5000**
AST (UI/L)	3.12-38	15.73±10.34	20.20±6.67^{ns}	6.76±2.54^{ns}	7.40±2.71^{ns}	1.37±1.02^{ns}	2.42±1.43^{ns}
ALT (UI/L)	3-40	3.77±0.29	9.51±5.83^{ns}	1.08±1.16^{ns}	5.59±2.65^{ns}	0.61±0.15^{ns}	1.76±1.06^{ns}
ALP (UI/L)	17-77	50.41±45.84	41.54±27.53^{ns}	22.06±8.09^{ns}	70.83±52.14^{ns}	30.96±13.14^{ns}	36.81±33.06^{ns}
TB (mg/dL)	2-11	2.01±2.01	1.05±0.40^{ns}	1.05±0.30^{ns}	0.99±0.45^{ns}	2.98±3.50^{ns}	4.63±5.71^{ns}
DB (mg/dL)	6-25	1.39±1.07	6.96±9.13^{ns}	6.09±5.63^{ns}	0.81±0.25^{ns}	0.66±0.40^{ns}	9.95±11.55^{ns}
UREE (mg/L)	9-31.5	11.90±0.80	15.14±3.18^{ns}	10.94±1.30^{ns}	10.26±1.32^{ns}	13.76±2.15^{ns}	10.55±0.61^{ns}
CREA (mg/dL)	0.6-1.4	0.88±0.13	0.85±0.8^{ns}	0.73±0.09^{ns}	0.58±0.07^{ns}	0.73±0.17^{ns}	0.88±0.11^{ns}

ns not significant

3.4. Haematological profile of *Mus musculus* after administration of high doses of methanolic extract from the leaves of *Ficus stuhlmannii* Warb.

The control group showed normal values. The same was true for the groups that received the aqueous extract at doses ranging from 1000mg/kg to 5000mg/kg of body weight. The haematological profile results are shown in Table IV.

Table 4 Evaluation of the acute toxicity of the MeOH extract on several haematological markers in treated mice (n= 3) and controls

Markers	**Standard [19–21]**	**Control**	**1000mg/Kg**	**2000 mg/Kg**	**3000 mg/Kg**	**4000 mg/Kg**	**5000 mg/Kg**
SR (mm/h)	1.1-14	6.67±1.15	4.33±4.93^{ns}	5.00±3.00^{ns}	4.67±2.08^{ns}	5.00±4.00^{ns}	6.00±2.65^{ns}
MCM (µm^3)	71-96	109.47±19.31	98.17±11.06^{ns}	110.10±40.10^{ns}	82.17±12.15^{ns}	68.23±5.20^{ns}	119.57±41.16^{ns}
PLT (x10^3 /µL)	140-813	316.33±177.56	261.00±144.00^{ns}	270.00±124.00^{ns}	250.00±125.30^{ns}	214.67±85.70^{ns}	236.67±151.19^{ns}
RBC (x10^6/mm^3)	5-12.5	4.73±0.51	5.63±0.38^{ns}	5.07±1.56^{ns}	6.43±0.91^{ns}	6.10±1.60^{ns}	3.27±0.64^{ns}
WBC (×10^3 /mm^3)	3-13	3466.67±222.22	3533.33±1444.44^{ns}	5866.67±955.56****	4700.00±733.33****	6266.67±1888.89****	4433.33±444.44*
HTC %	36-52	50.87±4.45	54.97±1.74^{ns}	43.70±10.74^{ns}	51.97±4.95^{ns}	43.30±9.10^{ns}	34.73±2.73^{ns}
Hb g/dL	9-15	15.14±1.55	16.69±0.37ns	14.36±3.03ns	14.85±0.24^{ns}	9.29±5.88^{ns}	10.47±0.49^{ns}
basophils (x10^3/µL)	0 - 0.01	0.67 ±0.89	0^{ns}	0^{ns}	1.67±1.11^{ns}	0^{ns}	0.33±0.44^{ns}
Leukocyte (x10^3/µL)	7-18	58.67±1.78	54.00±4.00^{ns}	58.67±7.56 ^{ns}	60.00±13.33 ^{ns}	46.00±21.33 ^{ns}	46.00±21.33 ^{ns}
Monocytes (%)	5-10	2.67±2.22	4.33±2.89^{ns}	5.67±1.78^{ns}	3.00±2.67^{ns}	5.33±2.22^{ns}	1.33±1.78^{ns}
Eosinophils (x103 /µL)	0 - 0.11	0	0ns	0ns	0ns	0ns	0ns

*= p< 0,05; ** = p< 0,01; ***= p< 0,001; ****= p<0,0001 (significant difference compared to the control group was observed in some groups); ns: not significant. MeOHE: methanol extract.

4. Discussions

No signs of toxicity or death were observed during our experiment, as shown in Table I above. In this study, the Lorke procedure was chosen because it offers the advantage of obtaining adequate information regardless of the type of material tested and the route of administration chosen. There were no deaths or signs of acute toxicity in mice treated with all doses up to 5000 mg/kg of *Ficus stuhlmannii* Warb leaves, indicating that the oral LD_{50} of *Ficus stuhlmannii* Warb is greater than 5000 mg/kg body weight.

In addition to the absence of clinically and biologically observable toxicity, no deaths were recorded in animals after oral administration of the methanolic extract of *Ficus stuhlmannii* Warb leaves. This finding suggests that the lethal dose 50 (LD_{50}) of the extracts examined is higher than the dose administered. It was then reported that, according to the LD_{50}, the extract can be considered highly toxic if the LD_{50} is equal to or less than 1 mg/kg, very harmful if the LD_{50} is between 1 mg/kg and 50 mg/kg, moderately toxic if the LD_{50} is between 50 mg/kg and 500 mg/kg, and practically non-toxic if the LD_{50} is greater than or equal to 1500 mg/kg. The LD_{50} is equal to or less than 1 mg/kg, indicating that the extracts are low in toxicity. If the LD_{50} is between 500 mg/kg and 5000 mg/kg, the extracts are considered moderately toxic. However, if the LD_{50} is between 5 g/kg and 15 g/kg, the extracts are practically non-toxic. Finally, if the LD_{50} is greater than or equal to 15 g/kg, the extracts are safe. [16,22]. Consequently, these results confirm the classification of the methanolic extract of *Ficus stuhlmannii* Warb. leaves as practically non-toxic. This table shows the variation in weight between treated and untreated mice. We found no significant difference between the two groups. The administration of methanolic extract from *Ficus stuhlmannii* Warb leaves therefore does not cause any change in weight in exposed subjects. This finding is similar to the work of Ojo et al. (2013) who reported LD_{50} value for aqueous *Ocimum gratissimum* leaf extract to be 4242.64mg/kg [16,23]. On day 14, a slight weight loss (< 1.5 g), potentially related to diet, was recorded in some groups. Statistical analysis ($p > 0.05$) revealed no significant variation between the treated groups and the control group during the study. These results indicate that the methanolic extract does not induce any significant adverse effects on the animals' appetite or growth. Comparing the initial average weight on day 0 of the treated group with the final average weight on day 14, no significant difference in weight was observed ($p > 0.05$). A slight weight loss was observed in animals treated with methanolic extract (Lot 1 = 3.3 g; Lot 2 = 6 g; Lot 3 = 8.63 g; Lot 4 = 7.63 g and Lot 5 = 0.33 g) did not show any significant difference ($p > 0.05$). The observed increase or loss in body weight of the test animals suggests that they did accumulate energy from their normal diet [24]. However, no difference was observed between the control animals and the treated animals. In fact, an extract is generally considered toxic when it causes weight loss of approximately 10% or more [25]. Weight change is used as a general indicator of the adverse effects of chemical compounds [26]. As a result, weight loss is linked to the animal's physiological functioning and can be explained not only by anorexia but also by changes in the animals' metabolism [27,28].

The values for the negative control group are within the normal range (Table III). The same is true for the groups that received the methanolic extract of *Ficus stuhlmannii* Warb.

Analysis of biochemical parameters after administration of a single dose shows that there are no significant differences ($p>0.05$) between the control animals and those that received the extract. The AST marker decreased slightly at doses of 1000 to 5000 mg/kg from 20.20±6.67 to 2.42±1.43 IU/L; this was also observed with ALT parameters from 9.51±5.83 to 1.76±1.06 IU/L; PAL from 41.54±27.53 to 36.81±33.06 IU/L and UREA from 15.14±3.18 to 10.55±0.61 mg/L. However, we observed a slight increase in parameters at doses of 1000 to 5000 mg/kg: creatinine 0.85±0.8 to 0.88±0.11 mg/dL; BLT from 1.05±0.40 to 4.63±5.71 mg/dL and BD from 6.96±9.13 to 9.95±11.55 mg/dL.

Analysis of the results (table IV) of haematological parameters after 14 days of treatment in mice shows a significant difference in white blood cell count in groups (2-5) compared to the control group and a non-significant difference ($p >0.05$) in blood platelet count, red blood cell count, mean corpuscular volume, haematocrit, sedimentation rate of haematocrit and haemoglobin compared to healthy controls. Some parameter values were within the pre-established normal range. These were: PLT (214.67±85.70 to 270.00±124.00x103/µL) ; GR (5.07±1.56 to 6.43±0.91 x106/µL); GB (3533.33±1444.44 to 6266.67±1888.89 x 103/mm3) except at a dose of 5000mg/Kg; HCT (34.73±2.73 to 51.97±4.95%) at a dose of 1000 mg/kg; Hb (9.29±5.88 to 14.85±0.24 g/dL) at a dose of 1000 mg/kg; Basophils (0 to 0.33±0.44 x103/µL) at a dose of 3000mg/kg and eosinophils (zero).

Acute toxicity tests, the first step in the toxicological evaluation of drugs, involve administering a single dose to animals over a short period of time and observing any adverse effects and mortality [29]. This study shows that the methanolic extract of *Ficus stuhlmannii* Warb. leaves did not induce mortality at a dose of 5000 mg/kg during the 14-day observation period. Slight changes in behaviour, breathing and hypoactivity were observed for a few minutes after administration of the dose in both the treated and control groups, indicating mild trauma from the conditions of administration. These slight behavioural deviations are thought to be due to stress and fear during force-feeding, a

highly unusual treatment for animals. In the work of Sanchez-Motoya et al, for certain natural products, a reduction in respiratory rate and convulsions were observed during intoxication with phenolic compounds; hypoactivity could be attributed, among other things, to the anaesthetic, sedative and, above all, psychoactive properties of the compounds, such as various alkaloids [30].

Although many secondary metabolites are present in the methanol extract, they are not toxic on their own or are present in non-toxic doses.

The acute oral toxicity threshold test is set at 5 g/kg body weight. If no mortality is observed at this dose level, a higher dose is generally not necessary [31]. The absence of mortality at this dose level indicates that there are no serious adverse effects associated with phytomedication and that methanolic extract of *Ficus stuhlmannii* Warb. leaves up to 5,000 mg/kg is not toxic under the conditions of this study. These results corroborate those of Mpiana et al.; Ngbolua et al. [32,33] who conducted studies on the aqueous decoction of Drepanoalpha® plant powder and its lyophilisate. Consequently, their results suggest that the median lethal dose (LD_{50}) of the contents of Drepanoalpha® capsules is greater than 5000 mg/kg. Our results are similar, showing that the methanolic extract of *Ficus stuhlmannii* Warb leaves is 'practically non-toxic' after acute exposure in mice, according to the classification of Lu & Kacew (2003); furthermore, according to the OECD scale [31,34], The methanolic extract of *Ficus stuhlmannii* Warb leaves has a toxicity index of 5, which also corresponds to 'practically non-toxic [35].

Abbreviations

ACOETE	Ethyl acetate extract
ANOVA	Analysis of Variance
DCME	Dichloromethane extract
PEE	Petroleum ether extract
MEOHE	Methanolic extract
SR	Sedimentation rate
MCM	Mean Corpuscular Volume
PLT	Platelets
RBC	Red Blood Cells
WBC	White Blood Cells
HTC	Haematocrit
Hb g/dL	Haemoglobin
AST (UI/L)	Aspartate aminotransferase
ALT (UI/L)	alanine amino transferase (
ALP (UI/L)	Alkaline phosphatase
TB (mg/dL)	Total bilirubin
DB (mg/dL)	Direct bilirubin
CREA (mg/dL)	Creatinine

5. Conclusion

The objective of this study was to contribute to determining the subacute in vivo toxicity of the methanolic extract of *Ficus stuhlmannii* Warb. leaves in mice.

Determining acute toxicity makes it possible to highlight anatomopathological changes following a single administration of an active substance. In this study, oral administration of methanolic extract from the leaves of *Ficus stuhlmannii* Warb. at different doses (1000, 2000, 3000, 4000 and 5000 mg/kg bw) to mice observed for 15 days showed different effects depending on the dose administered but was not toxic.

The results obtained show that the methanolic extract of *Ficus stuhlmannii* Warb. leaves is not toxic to treated mice, even at a concentration of 5000 mg/kg.

Compliance with ethical standards

Acknowledgments

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Disclosure of conflict of interest

The authors declare that they have no conflicts of interest.

Statement of ethical approval

The principles governing the use of laboratory animals as set out by the OECD, the University of Montreal's Animal Use Ethics Committee, and the Canadian Council on Animal Care protocol were duly observed. All animal experiments were conducted in accordance with the Guide for the Care and Use of Animals and in accordance with the protocol approved by the Ethics Committee of the University of Lubumbashi.

Authors' contributions

Mbayo Kalubandika Glauber conducted the experiment and drafted the manuscript. All authors read, corrected and approved the final version of the document. Lumbu Simbi supervised the work and validated the manuscript.

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