HEPATOPROTECTIVE EFFECTS OF **Canna indica** L. RHIZOME AGAINST ACETAMINOPHEN (PARACETAMOL)

F. Longo¹, A. Teuwa¹, S. Kouam Fogue², M. Spiteller³ and L.S. Etoundi Ngoa¹

¹Laboratory of Animal Physiology, Department of Biological Sciences, Higher Teachers’ Training College, University of Yaounde I. PO Box 47 Yaounde-Cameroon.
²Laboratory of Chemistry, Department of Chemistry, Higher Teachers’ Training College, University of Yaounde I. PO Box 47 Yaounde-Cameroon.
³Institute of Environmental Research (INFU) of the Faculty of Chemistry TU Dortmund, Otto-Hahn-Str. 6, D-44221 Dortmund, Germany.

**ABSTRACT**

Self-medication often leads to poisoning the body, especially the liver. *Canna indica* is a Cannaceae empirically used to treat various types of hepatitis in Cameroon. Liver toxicity was observed four hours after oral administration of a single dose of Acetaminophen (APAP) 400mg.kg⁻¹. Effects of the aqueous extract of the rhizomes at doses 8, 16, 32, 64 and 128mg.kg⁻¹ bw, and that of the antidote N-acetylcysteine [Mucomyst] were evaluated in male rats, pre-treated with APAP. Results showed that *Canna indica* failed to increase rats’ body weight (P>0.05), normalised rats’ behaviour (aggression and sensitivity to touch and noise), and provoked a decrease (P<0.001) of the relative liver weight. *Canna indica* also modelized, at all doses, the liver’ damages caused by inflammation, through a stimulation of hepatocyte regeneration and a healing of inflamed areas. Its antioxidant effect was revealed by a lower plasma activity of alanine aminotransferase transaminase. The dose 8mg.kg⁻¹ and 16mg.kg⁻¹ showed the most effective activity. The presence of flavonoids and fatty acids, revealed by phytochemical analysis and two bioactive molecules: Carotol and 9-Cedranone (Cedran-9-one) using high performance liquid chromatography, known to posses anti-inflammatory activities, confirms the anti-inflammatory properties of *Canna indica*. *Canna indica* at doses comprises between 8 and 16mg.kg⁻¹ bw could be strongly recommended.
KEYWORDS: Canna indica L., Liver toxicity, Acetaminophen, N-Acetylcystein, ALT, Rat.

INTRODUCTION
Self-medication is increasingly on the rise in Cameroon. When out of control, it causes an overdose in the body, leading to a liver toxicity. Drugs overdose such as acetaminophen (Paracetamol) and alcohol-based products (beer, whiskey, etc.), can provoke drug-induced hepatitis.\cite{1,2} In fact, before tackling this disease, it’s worth mentioning that, drug intake could lead to the development of various forms of liver diseases including, among others hepatitis, cholestasis (liver tumor), steatosis (fat accumulation in the liver), phospholipidosis (accumulation of phospholipids in the liver), sclerosing cholangitis and vascular disease. The frequency of drug-induced liver disease is increasing.\cite{3,4} After absorption of a toxic dose, severe acute hepatitis can occur with a procession of nonspecific clinical signs such as nausea and vomiting in the early hours, followed by abdominal pain predominantly in the right hypochondrium and jaundice.\cite{5}

Cells have several defence mechanisms to protect against potentially toxic reactive metabolites coming from the drug metabolism: antioxidant enzymes [superoxide dismutase (SOD) and glutathione peroxidase], the absorption of vitamin E, ascorbic acid, and other free radical scavengers and by a repairing mechanism of DNA damage.\cite{6} Because of their lipid solubility, many drugs cannot be eliminated in native form by the body. They must firstly being metabolized to water-soluble substances, before being excreted in urine or sweat. Drug metabolism usually involves two phases: Phase I includes a variety of enzymatic systems which mainly consists of the oxidation of the native molecule, under the control of a complex enzyme system that includes cytochrome P$_{450}$ and Phase II consists of a conjugation reaction (mainly with glucuronic acid, under the control of uridine-glucuronosyltransferases (UGT)).\cite{6} Acetaminophen (Paracetamol), is a combination of para-acetyl-amino-phenol and N-acetyl-para-aminophenol (APAP); It is the active ingredient in many specialised drugs class of non salicylates antipyretic analgesics.\cite{7} APAP is devoid of anti-inflammatory properties, and has no effects on platelet aggregation, with the advantage to possess less restricted indications (as age), and is devoid of serious side effects when used at recommended doses. However, in case of overdoses, APAP becomes very noxious to the organism especially to the liver.\cite{8,9} It primarily acts in the central nervous system,\cite{10} by inhibiting the production of prostaglandins involved in the processes of pain and fever, through an inhibitory action on the prostaglandin H$_2$ synthase (PGHS). Other mechanisms of action have been invoked to explain the analgesic
and antipyretic activity of APAP such as a central serotonergic mechanism of action is suspected by potentiating the effect of serotonergic neurons in the spinal cord, thus exerting an inhibitory control on pain pathways. It may also act by limiting the release of beta-endorphins. Adverse reactions have been reported in most cases. The main adverse effects are found such as rash rash or urticaria, probably allergic, thrombocytopenia and asthma, hypotension anaphylactic shock, vascular purpura rectal ulceration, agranulocytosis and acute pancreatitis usually occurs when Paracetamol is combined with other drugs such as codeine. Chronic active hepatitis, granulomatous hepatitis and rhabdomyolysis are others secondary effects of Paracetamol. In very young children, Paracetamol may increase the risk of developing asthma.

_Canna indica_ L. belongs to the family of Cannaceae; with 10 species mostly are used for decoration because of the beauty of their leaves and flowers (yellow, orange or red). _C. Indica_ is well known and widely used in traditional medicine for its diuretic and inhibitory effects on enzymes involved in the mechanism of inflammation, among various diseases. In the western region of Cameroon (Mbouda), _C. indica_ is commonly used to treat hepatitis. The decoction of its roots, in combination with fermented rice, is used in the treatment of gonorrhoea and amenorrhea and the leaves burned off a smoke-owned insecticide. The purpose of the following tests is therefore to assess the anti-inflammatory effects of the aqueous extract of the rhizomes of _Canna indica_ L., on rats’ liver and on the activity of ALT (alanine aminotransferase), one of the main blood indicative of liver toxicity.

**MATERIAL AND METHODS**

The rhizomes of _Canna indica_ L. were collected in a Yaounde suburb (Oyom-Abang). Extraction was done as recommended by the traditional healer and the recommendations of medicinal plants’ uses. The botanical identification of the plant specimen was made at the national Herbarium of Cameroon in Yaounde by Mr Christian YOMBO, and found to be identical to the voucher specimen N°42058HNC, deposited by Biholong in May 1979.

**2-1. Plant materials**

**2-1.1. preparation of plant rhizomes’ extract**

After cleaning, 03kg of fresh _C. indica_ rhizomes were boiled in 4L of water during 30min; after filtration and lyophilization, the 33.85g of a caramel colour powder (yield = 1.128%) obtained was use for experiments after dilution in distilled water.
2-1.2. Phytochemical Analysis
Phytochemical analysis of the aqueous extract of the rhizomes of *Canna indica* (ACI) was conducted,[22] to detect the presence of reducing substances: saponins and triterpene glycosides, tannins (gallic and catechin); Chemical hydrolysis tests were also done to detect phenols, coumarins glycosides, flavonoids, alkaloids, fatty acids, poly-uronides and polysaccharides.

2-1.3. High performance liquid chromatography fingerprint
For HPLC analysis, the sample powder was weighed (1.0g) into a 100mL flask and 100% methanol was added to the volumetric mark. The mixture was sonicated for 60min at room temperature. After extraction, the mixture was filtered through a 0.2μm membrane filter. The resulting solution was used for HPLC analysis. The qualitative assessment was performed by HPLC coupled to a mass spectrophotometer, using a gradient method. Constituents were detected at wavelength 205nm according to their through their retention time, relative abundance and masses. The mass spectrum was obtained with Xcalibur 2.0.7., and structures were drawn using CS Chemdraw ultra.[23]

2-2. Animals
Six to eight-week-old pathogen-free male Wistar rats were bred in our laboratory and provided with water and food *ad libitum*. Experimental procedures were carried out according to guidelines for the care of laboratory animals from the Cameroon National Ethics Committee (Ref. N° FWA-IRB00001954) and with the NIH Guidelines for the Care and Use of Laboratory animals.[24]

2-3. Experimental Design
After fasting during eighteen hours[25] and divided into eight treatment groups (*n* = 5), except in the healthy group, acute liver injury was induced (*per os*) with 400mg.kg of APAP. (Tablet of 500mg, Lot N°2F72134 were purchased in a local drugstore), and diluted in warm phosphate-buffered saline (PBS, pH 7). Healthy and APAP groups received 1ml PBS; The reference group (NAC) received N-acetylcysteine, and test groups (ACI₁ – ACI₅) received ACI (8, 16, 32, 64 and 128mg.kg⁻¹) *per os* during 72Hrs.[7]

2-4. Autopsy and diagnostic
On the behavioural changes: aggression, sensitivity to noise and touch, mortality and water consumption were recorded throughout the experiment. Rats’ weights were recorded before
and four hours after the final treatment and euthanized using ethyl ether. The artero-venous blood of each rat was collected in a 5mL heparinized tube for transaminase analysis (ALT).[26] The liver was quickly isolated, weighed, filmed and the morphology was recorded and a biopsy of the left liver was preserved in 10% neutralized formalin for histological analysis. The relative weight of all livers was expressed in% [(liver weight /b.w.)*100].

2.5. Histological analysis
Liver biopsies were dehydrated, included in paraffin, cut (5μ), de-paraffined, rehydrated and coloured with hematoxylin-eosin,[27] and observed under a light microscope (Olympus) at x400 magnification.

2.6. Evaluation of Alanine amino-transferase enzyme (ALT)
The ALT assessment was performed using a KIT (FORTRESS diagnostics, ALT (GPT) IFCC KINETIC UV). Optical densities were measured using a spectrophotometer Genesis brand, at the wavelength of 340nm.

2.7. Statistical analysis
Statistical analyses were performed using Sigma-Stat 3.2. Software; Means values ± S.E.M. and the comparison between treated and controls groups were calculated by “t” student test. Multiple groups’ comparison was also done by One Way ANOVA test, using Mann-Whitney and Turkey methods, according to values. Results were considered significantly different at P<0.05.

RESULTS
3.1. Phytochemical analysis and HPLC-SM
The phytochemical analysis of ACI, revealed the presence of phenolic compounds, catechin gallic tannins, anthocyanins, alkaloids, fatty acids, coumarins and anthraquinons (tab.2).

Table 2: qualitative composition of the aqueous extract of Canna indica’s rhizomes

<table>
<thead>
<tr>
<th>Canna Indica</th>
<th>Polyuronids</th>
<th>Polyses</th>
<th>Saponins</th>
<th>Phenols</th>
<th>Gallic tannins</th>
<th>Catechin tannins</th>
<th>Anthocyanins</th>
<th>Triterpenes</th>
<th>Flavonoids</th>
<th>Alkaloids</th>
<th>Fatty acids</th>
<th>Coumarins</th>
<th>Anthraquinons</th>
<th>Glycosides</th>
<th>Anthraenique</th>
</tr>
</thead>
<tbody>
<tr>
<td>Canna Indica</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>
HPLC fingerprint coupled to the mass spectrometer after 28 min, showed a chromatogram spectrum with three main peaks corresponding respectively to: compound (A): retention time 0.61 and 0.7 min, RT 0.61, [M+H]⁺: m/z 360.1503; (calcd. for C_{27}H_{20}O 360.1504) and at the retention time of 6.66 min, two compounds appears: (B_1) [M+H]⁺: m/z; (calcd. For [M+H]⁺ 214.8362 C_{3}H_{11}O_{11} and (B_2) [M+H]⁺: m/z; (calcd. for [M+H]⁺ 212.8391C_{3}H_{11}O_{11}). Two known bioactive sesquiterpenes former isolated from the essential oil of *C. Indica*^{28}, named (C) Carotol: (3R,3aS,8aR)-3-Isopropyl-6,8 a-dimethyl-2,3,4,5,8,8a-hexahydro-3a(1H)-azulenol) RT: 12.18, m/z 222.2054 (calcd. For [M+H]⁺, m/z 223.2055 C_{15}H_{27}O) and (C’) 9-Cedranone (Cedran-9-one) RT: 5.93, m/z 221.3583; (calcd. for (C_{15}H_{27}O, [M+H]⁺ m/z 221.3552), were identified (Fig.3).

**Figure 3:** chromatogram profile of *Canna Indica* rhizomes: (A): RT 0.61, [M+H]⁺: m/z 360.1503; (calcd. for C_{27}H_{20}O 360.1504); (B_1) RT 6.66, [M+H]⁺: m/z ; (calcd. For [M+H]⁺ 214.8362 C_{3}H_{11}O_{11} , (B_2) (A): RT 6.66, [M+H]⁺: m/z 360.1503; (calcd. for C_{27}H_{20}O 360.1504); (C) Carotol: (3R,3aS,8aR)-3-Isopropyl-6,8 a-dimethyl-2,3,4,5,8,8a-hexahydro-3a(1H)-azulenol) RT: 12.18, m/z 222.2054 (calcd. For [M+H], m/z 223.2055 C_{15}H_{27}O) and (C_1) 9-Cedranone (Cedran-9-one) RT: 5.93, m/z 221.3583; (calcd. for (C_{15}H_{27}O, [M+H]⁺ m/z 221.3552).^{28}
3.2. *Canna indica* effects on animal behaviour

After APAP *per os* administration, rats were gregarious with minimal movements; Four hours later, they presented a cough coupled to breathing disorder. *C. Indica* caused few behavioural disorders (ACI₄ and ACI₅) which disappeared 72 hours later, similarly to NAC [Tab.3].

Table 3: Rats’ behavioural effects, 24 and 72 hours after administration of NAC and the aqueous extract of rhizomes of *Canna indica*.

<table>
<thead>
<tr>
<th>Doses (mg/Kg)</th>
<th>4 h after Acetaminophen</th>
<th>NAC</th>
<th>24h</th>
<th>72h</th>
<th>24h</th>
<th>72h</th>
<th>24h</th>
<th>72h</th>
<th>24h</th>
<th>72h</th>
<th>24h</th>
<th>72h</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>400</td>
<td>1330</td>
<td>8 (ACI₁)</td>
<td>16 (ACI₂)</td>
<td>32 (ACI₃)</td>
<td>64 (ACI₄)</td>
<td>128 (ACI₅)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Locomotion</td>
<td>N D N N N N N N D N D N</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sensitivity to noise</td>
<td>N D N N N N N N N N N</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sensitivity to touch</td>
<td>N D N N N N N N N N N</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aggression</td>
<td>N D N N N N N N D N D N</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stool</td>
<td>G G G G G G G G G G G G</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Breathing</td>
<td>N BD N N BD N RD N BD N BD N</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cough</td>
<td>NC C NC NC C NC C NC C NC C NC C NC</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*N=normal, C= Cough, NC= No Cough, D=decreased, G=granulous P=present, A=absent, RD= Breathing disturbance*

3.3. *C. indica* Effects on rat’s body weight

ACI caused failed to modify (P> 0.05) rat’s weight gain similarly to NAC.

3.4. *C. indica* on rat liver’s relative weight

ACI decreased rat’s liver relative weight, at all doses (*vs* APAP), especially at 8mg.kg⁻¹ [4.54 ± 0.49% (P<0.05)] equivalent to -29.61% (*vs* NAC -4.72 ± 0.45% (P <0.05)], lower than NAC [fig.5].
3.5. *Canna indica* effects on ALT

APAP caused a plasmatic ALT increase up to 1870.16IU/L. ACI decreased (P<0.05) the plasmatic level of ALT similarly to NAC. Meanwhile, ALT decrease induced by *C. indica* occurred inversely proportional to the dose: the dose of 8mg/kg induced the highest decrease value of 451.05 IU/L (P<0.001), while the dose of 128mg/kg induced a decrease of 1458.855IU/L. In groups ACI2 and ACI4, *C. indica* induced highest decreases equivalent to 71.98% and 75.88% respectively, compared to APAP group [1870.16IU/L]. Likewise, compared to NAC group [520.308IU/L], ACI1 and ACI2 showed the highest efficiencies with values inferior to NAC, while from ACI3 to ACI5, *C. indica* caused weak decreases. Comparatively to Healthy rats [111.55IU/L], ALT values induced by ACI and NAC stayed upper [fig.6].
Figure 2: *Canna indica* rhizomes’ aqueous extract and NAC effects on ALT plasmatic rate. Each bar represents mean ±S.E.M. n=6; *P<0.05; **P<0.01; ***P<0.001: Significant difference vs Paracetamol group (P); §P<0.05; §§§P<0.001: Significant difference vs NAC group.

3.6. *Canna indica* effects on the liver morphology and histology

APAP provoked a liver tissue damage characterised by numerous nodules (oedemas) [vs. H group] [fig.7H]. Histological analysis showed a cell degeneration at the portal vein with a membrane’ lesion, resulting to a loss of cytoplasm [fig.7P], while the healthy liver (H) showed a normal liver tissue architecture, with normal hepatocytes, sinusoids with their lining cells, normal portal vein, hepatic artery and the bile duct [fig.7H]. NAC administration revealed a liver with a texture similar to a healthy liver. However, the presence of healing points and small nodules [fig.7NAC] were recorded; at the tissue level, few inflamed hepatocytes have also been recorded nearest the portal vein (fig.7NAC').

*C. indica* significantly improved the inflammation caused by APAP 400mg/kg: *On the morphology* of all rats (ACI1 to ACI5), livers were no longer shown nodules, but the presence of healing points was observed, with a dark red colour [fig.7ACI1-5]. *On the histology*, rats’ liver from group ACI1 to ACI3 presented normal hepatocytes [fig.7ACI1-3]; Group ACI4 (64mg/kg) presented a lack of hepatocyte vacuolation (no degeneration) accompanied by a slight inflammation and abnormalities [fig.7ACI4] and group ACI5 (128mg/kg) presented a liver tissue with a structure similar to that of inflamed liver (APAP’) with the presence of bi-nucleated hepatocyte with membrane lesions and numerous inflammation [fig.7ACI5'].
DISCUSSION

Evaluation of the anti-inflammatory effect of the aqueous extract of *Canna indica* rhizomes was made on liver toxicity induced by APAP (Acetaminophen). Oral administration of an acute dose of 400mg/kg APAP caused lesions resulting in the presence of granulomas, oedema (small tumours of inflammatory origin) and numerous polymorphic cells, cells or specific items scattered throughout the liver tissue; On the liver tissue, acetaminophen caused a massive hepatocyte necrosis at the centrilobular region, with a dark red colour).\[^{5,29}\] *C. indica* significantly improved the alteration induced by APAP. The improvement in aggression, sensitivity to noise and locomotion demonstrated a tranquilizing effect exerted on the central nervous system.\[^{30}\] Similarly, the liver’s relative weight decrease also revealed a sign of behavioural improvement, especially at doses 8 and 16mg/kg. *C. indica* caused a decrease in relative liver weight below that of the NAC group; Indeed, it is known that, inflammation in general, and granulomas that appeared on liver tissue in particular, comes from the fluid and cellular infiltration: inflammatory cells: leukocytes, macrophages, Kupffer cells\[^{12,6,30}\] which certainly contributed to the relative liver weight increase on APAP livers. In case of overdose, N-acetylcystein (NAC) is used to strengthen the body defence towards the
toxic metabolites by reducing their level. Improvements in liver tissue damage by *C. indica* were similar to NAC, especially at dose 64mg/kg, characterised by residual inflammatory cells and some granulomas. In contrast, at doses 8 and 16mg/kg, the effects of *C. indica* were significantly higher than that of NAC, with a complete elimination of inflammation, demonstrated by the presence of scars, and the absence of nodules and granulomas.

Transaminases are usually measured in serum to detect pathogen. Generally, liver toxicity is detected by assaying only the TGP (glutamic pyruvic Transaminase also called ALT), which is preferentially localized in the liver (organ specificity). Serum ALT levels were elevated in APAP rats, up to 6 times higher than that of NAC group. In addition, it has been shown that NAPQI, a molecule produced during APAP poisoning, creates adducts (compounds produced by addition to a DNA molecule), which can bind to liver protein. Degradation of membrane lipids and disruption of calcium homeostasis can be resulted, causing necrosis and cytolytic hepatitis up to the kidney by the same mechanism (1 to 2% of cases). Thus, oedemas and necrosis of hepatocytes observed in the livers are evidence of an actual presence of APAP poisoning. The fulminate hepatitis induced by a dose of 400mg/kg of APAP also led to a considerable rise in plasma alanine aminotransferase concentration, up to 1870.16 IU / L. The ¾ decrease (P<0.05) clearly demonstrated an antioxidant effect (anti-inflammatory effect) of *C. indica*. This result is similar to results which also showed a decrease in plasma transaminase (ALT), and a reduction/absence of inflammation in the evaluation of hepatoprotective effects of different extracts from medicinal plants in APAP hepatotoxicity cases.

Among the bioactive molecules from the rhizomes of *C. indica*, revealed by the phytochemical analysis, phenolic compounds like coumarins and flavonoids detected are the major antioxidants regularly founded in natural products. In addition, among all those compounds, anthocyanins and alkaloids detected, are also known for their anti-oxidative activity. The presence of fatty acids in rhizomes is also proven by the discovery of a red oil called "Cannabiol" in red flowers of *C. indica*. Those molecules have shown anti-ischemic, anti-platelet, anti-inflammatory and anti-lipoperoxide, with biological antioxidants effects mainly: radical-scavenging activity and enzyme inhibition through the action of enzymes involved in oxidation systems such as: 5-lipoxygenases, cyclooxygenases, the monooxygenases and an anti-lipoperoxidation), could be responsible of the so-observed biological activities of *C. indica*, alone or synergistically. In addition, an antioxidant activity of *C. indica* has already been demonstrated confirming once again, the anti-inflammatory/antioxidative activity of *C. indica* rhizomes. The major components has been
identified as steroids and the identification of Carotol\textsuperscript{[28]} and 9-Cedranone, earlier discovered in \textit{Marchantia Convoluta} leaves with a proven cytotoxicity activity on human liver and lung cancer cells\textsuperscript{[40]} thus, those components of the essential oil of \textit{C. indica} rhizomes, could take part on the anti-inflammatory effect observed.

**CONCLUSION**

The behaviour normalization by \textit{C. indica} showed that it affects the central nervous system. Although, the lack of effect observed on rat’ body weight, linked with a decrease in liver weight, contributed to demonstrate its curative effects without affecting animal weight. The presence of healing marks and the decrease on ALT also confirmed its anti-inflammatory effects. The extract revealed a greater efficiency at doses 8 and 16mg/kg. In addition, previous work has determined to 22.56mg/L, the median lethal dose (LD\textsubscript{50}) of the same extract\textsuperscript{41}. This, once again, confirmed our results which showed that the dosage 8 and 16mg/kg, actually belongs to the range of effective doses 100 (ED\textsubscript{100}) and lethal dose\textsubscript{50} (LD\textsubscript{50}) of the aqueous extract of rhizomes of \textit{Canna indica} L.

The most characteristic is that, \textit{C. indica} rhizomes’ extract was more effective at low doses. Indeed, the doses 8 and 16mg/kg induced greater reductions in the liver relative weight, with a total regeneration of hepatocytes, and a complete reduction of inflammation. This result is also confirmed by tests on plasma ALT levels, which showed that at both doses, the extract induced the greatest oxydative activity.

**ACKNOWLEDGMENTS**

Special thanks to Dr D. Nzeufiet of the Laboratory of Animal Physiology (Faculty of Sciences-UYYI) for histological analysis, and to Dr G. Agbor and Mr R. Ajang Ngidi of the Phytochemistry Laboratory of the Institute of Medical Research and Study of Medicinal Plants of Cameroon (IMPM) for phytochemical analysis.

This work was supported, in part, by the German Academic Exchange Service (DAAD) initiative “Welcome to Africa” and the Cameroonian High Education Ministry through the \textit{special allocation} account for the modernization of \textit{University research} in Cameroon.

**REFERENCES**


20. Sofowora A. Medicinal Plants and Traditional Medicine in Africa. 1984: p.44.


