

## Original Article

# Nephrocurative Effect of Flavonoids-Rich Fraction of *Irvingia gabonensis* Seed Extract against Acetaminophen-induced Kidney Damage in Mice

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## ABSTRACT

This paper investigated the nephrocurative effect of flavonoids-rich fraction of *Irvingia gabonensis* seed extract in acetaminophen-induced kidney damage in mice. The total flavonoids content of the different fractions of the extracts were reported to be higher in the Ethyl acetate fraction with a value of 47.20mg QE/g followed by Methanol 30.41mg QE/g, Chloroform 20.10mg QE/g, and Petroleum ether with 12.40mg QE/g. Further studies on the flavonoids-rich extract (Ethyl acetate fraction) revealed a significant difference ( $p < 0.05$ ) between the inhibitory power of the flavonoids-rich fraction of *Irvingia gabonensis* (IG) seed extract and that of Vit. C. Accordingly, the  $IC_{50}$  of the flavonoids-rich fraction of *Irvingia gabonensis* seed extract (47.97 $\mu$ g/ml) calculated was observed to be higher than that of Vit. C. standard (25.76 $\mu$ g/ml). Studies on the nephrocurative effect indicate a positive result with significant deference across the group and an ultimate normalization of the kidney biomarkers. The curative effect of the extract is assumed to be dose and time depended on a drastic decrease in kidney biomarkers after four weeks of extract administration. Toxicity study of the extract also revealed the extract to be safe at 3000mg/kgbw. This study to an extent has proven that *Irvingia gabonensis* seeds are a promising source of alternative raw material in pharmaceuticals for kidney toxicity treatment. Therefore, the flavonoids rich fraction of *Irvingia gabonensis* seed extract may have curative effect in the treatment of kidney diseases.

**Keywords:** Acetaminophen, Flavonoids, Inflammation, Medicinal, Nephrocurative

## INTRODUCTION

Exploration of plant metabolites has always been a promising approach in drug development. The quest for research in medicinal plants has continued to gain momentum and acceptance is increasing. Undeniably, the plant kingdom serves an important function in the provision of essential services to

mankind. Interestingly, humans have embraced the use of plants for medicinal purposes since ancient times. These medicinal plants are referred to as "herbs". It is well known that plants have been used as food, shelter, fuel and clothing. Medicinal plants are said to be one of the older sciences in existence and approximately more than one

tenth of different plant species are being used as raw materials in the pharmaceutical and cosmetics industries. These plants are said to possess active ingredients with medicinal properties. Unfortunately, medicinal plants used across the world either as traditional medicine or as active ingredients in conventional medicine are not evenly distributed across all geographical locations.<sup>1</sup>

Gurib-Fakim *et al.* define medicinal plants as plant varieties with medicinal properties and that these plants are a rich source of active compounds used by pharmaceutical industries in the development of synthetic drugs.<sup>2</sup> Noteworthy, different parts of these plants such as the root, stem, bark, leaf, fruit, flowers, seeds or even the whole plant can be utilized in disease treatment. Fortunately, the presence of active compounds in various parts of medicinal plants plays a direct or indirect role in disease treatment. That said, in several plant parts, certain metabolites or phytochemicals are produced and stored with active therapeutic and physiological effect on the human system.<sup>3</sup> Medicinal plants can sometimes work in synergy with another plant to elicit a therapeutic response, which can be more effective than when using a single medicinal plant. However, the utilization of more than one medicinal plant at the same time may cause the constituents of the plant to interact with each other, leading to either a beneficial or an adverse effect. Plant-derived compounds can dramatically improve hard-to-treat illnesses, such as cancer and other organ related diseases. Plant components are also characterized by their ability to prevent the development of certain diseases.

*Irvingia gabonensis* belongs to the *Irvingiaceae* family of plants. The plant is a large type of tree with large dense green leaves, edible seeds and fruit pulp. The plant is widely distributed in Africa, specifically in west Africa. The plant is important to the endogenous people of Africa as a source of food, medicine and fuel. The plant is rich in vitamin C and it is traditionally used in the treatment of many diseases. The seed of the plant is known as Goronor in Hausa, Ogbuno in Igbo, and Apon in Yoruba, Nigeria. Different parts of the plant have been exploited for medicinal properties, with the plant bark as the

commonly used part of the plant.<sup>4</sup> Shiembo *et al.* and Ayuk *et al.* reported the use of the bark of the plant to treat diseases such as hernia, yellow fever, diarrhea, dysentery, liver disease, sterility, and urethral discharge.<sup>5, 6</sup> In addition, Franzel *et al.* reported the use of the plant as a nutritional therapy for underweight babies, while Ngondi *et al.* stated the use of the plant in decreasing obesity and normalizing lipidemia.<sup>7, 8</sup> The plant has also been proven to have antioxidant activity as reported by Agbor *et al.*<sup>9</sup> Jamshidi *et al.* has attributed the utilization of plant traditional medicine in most parts of the world to its accessibility, affordability, acceptability, obtainability, and relatively lower level of toxicity.<sup>10</sup> In addition, the toxicity and adverse effects of conventional and allopathic medicines have also been important factors in the sudden increase in population demand for medicinal plants (herbs). This has resulted in an increase in the number of plant medicinal manufactured drugs followed by a reduction in the use of chemical drugs.<sup>2</sup> In the sequential research of *Irvingia gabonensis* medicinal potentials, Etta *et al.* investigated the effect of the plant seed on the liver and gonads of male albino rats while Ewere *et al.* investigated the plant's hypolipidemic therapeutic potential, all with promising results.<sup>11,12</sup>

### Acetaminophen-Induce Kidney Damage

N-acetyl-P-aminophenol commonly called "acetaminophen", is a commonly used painkiller. The drug is therapeutically safe when taken at the right dose. When taken in excess of the therapeutic dose, however, it can cause severe damage to major organs such as the liver and kidney in both humans and experimental animals.<sup>13</sup> Although a study by Eguia and Materson stated the episode of acetaminophen-induced liver damage to be more severe than kidney damage, the latter can also result in acute kidney damage.<sup>14</sup> Though many studies have been conducted on acetaminophen-induced liver damage, little has been done on kidney damage.<sup>15</sup> The step-by-step process of acetaminophen-induced kidney damage, as shown in figure 2, includes: The metabolism of acetaminophen (N-acetyl-P-aminophenol) to NAPQI (N-acetyl-para-amino-benzo quinoneimine) by cytochrome

p450 enzymes. While NAPQI can be converted to Glutathione conjugates by glutathione peroxidase, some of these molecules bind to the S3 segment of the distal tubules of the kidney, resulting in impairment in renal tubular structures, inflammation and toxicity. Further oxidative degradation of lipids takes place, leading to nephrotic cell membrane damage and reducing anti-inflammatory action.

In an era of seemingly limitless possibilities in the field of medicinal plant research, the prospect of obtaining an effective therapeutic effect from a single plant

constituent is enthralling. Therefore, this study investigated the

## MATERIALS AND METHODS

### Preparation of Plant Material

*Irvingia gabonensis* (ogbono) seeds were obtained from Station Round-about market in Kaduna metropolis, Chikun Local Government area of Kaduna State, Nigeria. The seeds were authenticated at the Biology Department, Ahmadu Bello University, Zaria, Kaduna State. A voucher number 113 was obtained and the plant was deposited in the Herbarium. The seeds were rinsed in running water and shade-dried for two weeks. The plant seeds were then pounded using mortar and pestle and weighed. The pounded plant material was sealed in a plastic bag and labelled as reported by Mukhtar and Tukur.<sup>16</sup>

### Defatting of Plant Material

The plant material was defatted using a Soxhlet extractor with n-hexane at a 1:3 plant material to n-hexane ratio, as

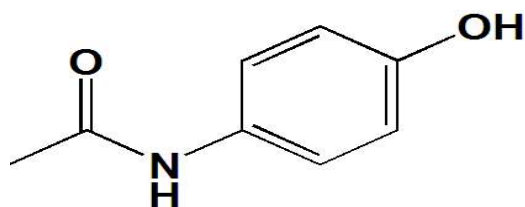
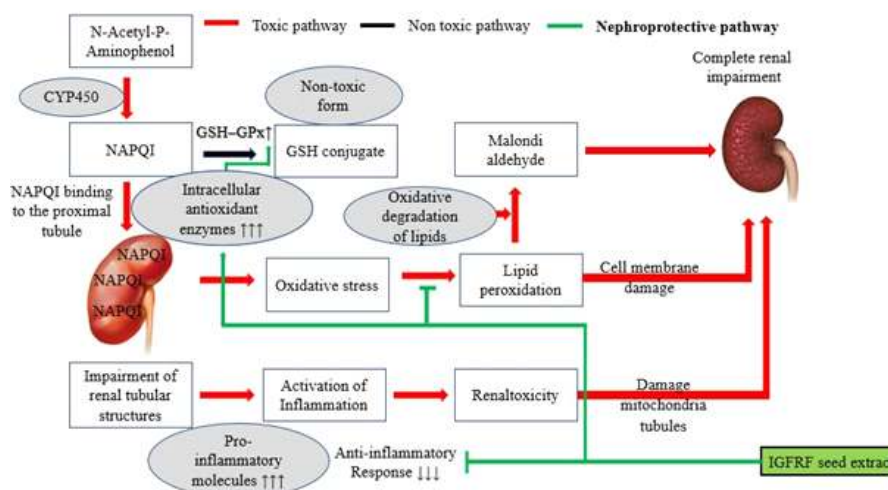


Figure 1: Structure of Acetaminophen



**Figure 2:** Hypothetical mechanism of action of flavonoids-rich fraction of *Irvingia gabonensis* (IG) seed extract against acetaminophen-induced kidney damage. It is believed that the flavonoids-rich fraction enhances the activity of GSH-GPx, increase intracellular antioxidant enzymes activity, inhibit the oxidative toxicity pathway and further down regulate the pro-inflammatory cascade of reaction involved in acetaminophen-induced kidney damage. Thus, these evidently deduced that the flavonoids-rich fraction of IG reduced the exacerbation of kidney damage and therefore, could be a potential nephron-curative agent. (Mechanism adopted with modification from karthivashan *et al.*<sup>15</sup>).

described by Efosa *et al.*<sup>17</sup>

### Crude Extraction of Plant Material

The crude extraction was done using microwave-assisted extraction (MAE). The defatted plant material was dissolved in 70% aq. ethanol and heated for one minute in a microwave at 80°C. The extract was then filtered using number 41 Whatman filter paper. The filtrate was then concentrated in a rotary evaporator under reduced pressure.<sup>16</sup>

### Fractionation of Crude Plant Extract

The crude plant extract was partitioned using different solvents of increasing polarity - petroleum ether, chloroform, ethyl acetate and methanol with the aid of Soxhlet apparatus at 40°C for 2-3 hours. Solvent extracts obtained were concentrated under reduced pressure by a rotary evaporator and subsequently labelled.

### Determination of Total Flavonoids Content (TFC)

The total flavonoids content in each solvent fraction was determined by Aluminum chloride colorimetric assay as reported by Laghari *et al.*<sup>18</sup> Quercetin was used as a standard. The flavonoids content was calculated as mean  $\pm$  SD (n=3) and expressed as mg/g of quercetin equivalent of dry *Irvingia gabonensis* seed extract.

The Flavonoids content of each solvent fraction was estimated using the linear regression equation obtained from the standard curve of quercetin and the formula below.

$y = 0.578x + 0.0954$ ,  $r^2 = 0.995$ ,  $y =$  Absorbance of each solvent fraction.

Flavonoids Content =  $c \times c / M$

Where C = concentration of quercetin from calibration curve in mg/cm<sup>3</sup>, V = volume of extract in cm<sup>3</sup> and M = weight of extract in grams.

### Isolation of Crude Flavonoids fraction

Having discovered the total flavonoids content to be higher in the ethyl acetate fraction, the fraction was dissolved in water and further extraction was done using

chloroform in a separating funnel. Sodium chloride (10%) was added drop wise to the aqueous layer to precipitate out tannins. The supernatant liquid was then partitioned with ethyl acetate and the solvent was evaporated to yield the crude flavonoids fraction.<sup>18</sup>

### Free Radical Scavenging Activity by DPPH Method

The free radical scavenging activity of different concentrations of solvent extracts of the flavonoids-rich extract and of standard ascorbic acid solution was evaluated by using the DPPH radical scavenging method as reported by Owaisi *et al.*<sup>19</sup> with slight modifications. The percentage of radical scavenging activity of tested extracts and positive control ascorbic acid was calculated using the formula below.

Free radical scavenging activity (%) =  $(Ac - As) / Ac \times 100$

Where Ac = Absorbance of control at 517nm and As = Absorbance of sample. The concentration of sample required to scavenge 50% of DPPH free radical (IC<sub>50</sub>) was determined from the curve of percent inhibitions plotted against the respective concentration.

### Acute Toxicity (LD<sub>50</sub>) Study

The LD<sub>50</sub> was determined using Lorke's method with slight modification.<sup>20</sup>

The LD<sub>50</sub> was calculated using the formula:

$LD_{50} = (D_0 \times D_{100})$

D<sub>0</sub> = Highest dose that gave no mortality,

D<sub>100</sub> = Lowest dose that produced mortality.

### Induction of Kidney toxicity in Mice

Nephrotoxicity was induced according to the method of Abdulmumin *et al.*<sup>21</sup> Acetaminophen 800mg/Kg was dissolved in aqueous solution and administered orally to the mice after animal's acclimatization and feeding with pellet and water ad libitum for 2 weeks.

### Experimental Animals

Twenty-seven (27) experimental male albino mice weighing between 25 and 40g were purchased from

Kaduna State University's pharmaceutical science department. The mice were selected randomly into nine groups of three mice each (Group I, II, II, IV, V, VI, VII, VIII and IX). The mice were acclimatized for fourteen days under standard temperature (23°C) and humidity conditions with a 12-hour light/dark cycle. The mice were fed standard feed and water was given to them ad libitum. The animal handling protocols were adapted based on the guidelines of the National Institute of Health (NIH, 1998) for laboratory animal care. Mice in group I served as the normal control group (no extract, no acetaminophen administered), but were fed with pellets ad water ad libitum throughout the experimental period, while mice in group II served as the negative control group. Mice in this group were administered with acetaminophen but no extract was given to this group. Mice in group III served as the positive control groups. Acetaminophen was administered, followed by daily administration of Captopril 0.8mg/cm<sup>3</sup>/Kg body weight dissolved in distilled water. Group IV-IX belongs to test groups. Mice in group IV – VI were administered acetaminophen, followed by 25, 50, 100mg/kg of the

extract for two weeks, while mice in group VII-IX were administered 25, 50, 100mg/kg of the extract for four weeks. Blood samples were collected from each experimental animal via the heart vessels into non-heparinized bottles for organ function analyses at 24 hours after the last feeding at two weeks and four weeks respectively.

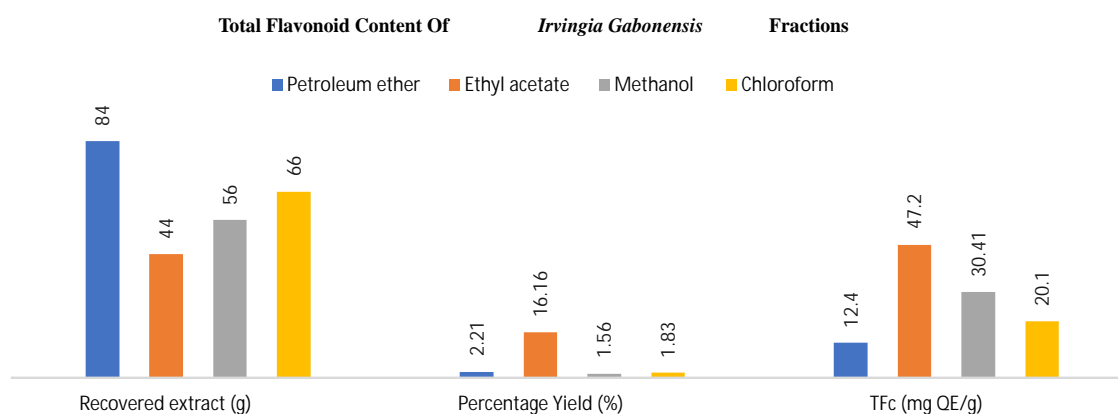
#### Determination of Biochemical Parameters

Serum Sodium Concentration by Maruna, Serum Potassium Concentration by Henry, Serum Creatinine Concentration by Bartels and Bohmer (1972), Serum Bicarbonate Concentration by Forrester et al., Serum Chloride concentration by Tietz and Serum Urea Level by Weatherburn respectively.<sup>22-27</sup>

#### Statistical Analysis

Statistical analysis was done using one way analysis of variance (ANOVA) using SPSS 2014 software at 5% level of significance ( $p < 0.05$ ). The data is given as the Mean  $\pm$  standard deviation.

## RESULTS AND DISCUSSION



**Figure 3: Total recovery of *Irvingia gabonensis* seed extract in grams, Percentage (%) yield and total flavonoids content (TFC) of the different solvent fraction of the plant seed extract.**

The result above shows the total flavonoids content (TFC) of the various crude extracts. The highest TFC was recorded in the Ethyl acetate extract (47.20mg QE/g) with the lowest flavonoids content reported in petroleum ether extract. Previous research by Mgbemena *et al.* confirmed the presence of a high concentration of

flavonoids in *Irvingia gabonensis* seed.<sup>28</sup> Furthermore, the above result is consistent with Thavamoney *et al.* report, in which they recovered the highest total flavonoids content of *Dacryodes rostrata* fruit in the ethyl acetate fraction after several solvent fractionations.<sup>29</sup>

**Table 1: Effect of the Flavonoids rich extract (ethyl acetate fraction) of *Irvingia Gabonensis*(IG) on 1.1 Diphenyl -2-Picryl -Hydrazyle (DPPH) Radical Activity – Compared with Vitamin C.**

Concentration (µg/ml)	IG Absorbance (Mean±SD)	Vit.C Absorbance (Mean±SD)	IG% radical scavenging activity (%)	Vit.C% radical scavenging activity (%)
20	1.09±0.45 <sup>a</sup>	1.98±0.45 <sup>a</sup>	44±12.7 <sup>b</sup>	26±12.7 <sup>bc</sup>
40	1.26±0.11 <sup>ab</sup>	1.48±0.11 <sup>b</sup>	60±10.6 <sup>b</sup>	45±10.6 <sup>c</sup>
60	0.50±0.55 <sup>b</sup>	1.60±0.55 <sup>b</sup>	74±6.4 <sup>ab</sup>	65±6.4 <sup>b</sup>
80	0.22±0.84 <sup>c</sup>	1.89±0.84 <sup>a</sup>	90±14.1 <sup>a</sup>	70±14.1 <sup>b</sup>
100	1.09±0.16 <sup>a</sup>	2.41±1.16 <sup>a</sup>	95±3.5 <sup>a</sup>	90±3.5 <sup>a</sup>

Values are measure of triplicate estimation Means in the same column followed by different letters are significant at  $P < 0.05$ ,  $IC_{50}(IG) = 25.76$  (µg/ml),  $IC_{50}(Vit. C) = 47.97$  (µg/ml).

The DPPH scavenging activity of the extract as seen above (Table 1) indicates a significant difference ( $p < 0.05$ ) between the inhibitory power of the flavonoids-rich fraction of *Irvingia gabonensis* (IG) seed extract and that of Vit. C. Similarly, the  $IC_{50}$  of the flavonoids rich Fraction of *Irvingia gabonensis* (IG) seed extract calculated was observed to be higher than that of the Vit. C. standard. From an earlier study in 2008, Shirley

attributed the functions of high content of flavonoids in seed to acting in defending the seed against pathogens and predators.<sup>30</sup> These same physiological functions might be attributed to the reason behind the high antioxidant activity recorded in Ethyl acetate fraction with the highest flavonoids content. Agbor *et al.* and Ezeabara and Ezeani stated the plant has high antioxidant capacity, leading to its medicinal use.<sup>9,31</sup> Furthermore, the presence of Lupeol,

**Table 2: Serum Kidney Function Indices of Acetaminophen – induced Kidney Damage mice after the Two Weeks and Four Weeks Oral Administration of Flavonoids Rich Fraction of *Irvingia gabonensis* (IG)**

Groups	Na <sup>+</sup> (mmol/L)	K <sup>+</sup> (mmol/L)	Creatinine (mmol/L)	HCO <sub>3</sub> <sup>-</sup> (mmol/L)	Cl <sup>-</sup> (mmol/L)	Urea (mmol/L)
<b>Group I</b>	147.93±8.56 <sup>a</sup>	5.65±0.52 <sup>a</sup>	27.29±2.96 <sup>a</sup>	31.33±4.04 <sup>a</sup>	91.83±4.34 <sup>b</sup>	12.87±2.42 <sup>d</sup>
<b>Group II</b>	168.27±3.59 <sup>b</sup>	8.49±1.42 <sup>a</sup>	51.50±7.76 <sup>b</sup>	35.57±4.94 <sup>a</sup>	117.29±13.18 <sup>a</sup>	25.93±2.10 <sup>a</sup>
<b>Group III</b>	129.43±9.68 <sup>a</sup>	4.21±0.88 <sup>a</sup>	34.28±3.00 <sup>a</sup>	32.96±1.76 <sup>a</sup>	75.87±7.50 <sup>c</sup>	18.58±0.90 <sup>b</sup>
<b>Test group After two weeks administration</b>						
<b>Group IV</b>	152.00±6.24 <sup>b</sup>	6.53±1.27 <sup>a</sup>	41.66±5.25 <sup>b</sup>	34.62±6.46 <sup>a</sup>	98.27±11.62 <sup>b</sup>	22.18±2.76 <sup>b</sup>
<b>Group V</b>	147.67±3.06 <sup>a</sup>	6.99±1.74 <sup>a</sup>	39.02±5.76 <sup>a</sup>	34.02±5.76 <sup>a</sup>	95.53±6.46 <sup>b</sup>	19.67±3.23 <sup>b</sup>
<b>Group VI</b>	143.67±3.79 <sup>a</sup>	6.73±1.99 <sup>a</sup>	35.39±8.14 <sup>a</sup>	33.06±4.10 <sup>a</sup>	91.04±8.49 <sup>b</sup>	19.17±2.11 <sup>c</sup>
<b>After four weeks administration</b>						
<b>Group VII</b>	136.80±2.08 <sup>a</sup>	5.93±2.05 <sup>a</sup>	36.46±6.61 <sup>a</sup>	32.69±2.00 <sup>a</sup>	93.22±4.19 <sup>b</sup>	18.57±3.16 <sup>c</sup>
<b>Group VIII</b>	132.80±3.65 <sup>a</sup>	5.37±1.96 <sup>a</sup>	31.92±9.98 <sup>a</sup>	31.92±9.98 <sup>a</sup>	84.36±7.48 <sup>b</sup>	14.68±0.90 <sup>cd</sup>
<b>Group IX</b>	127.33±8.74 <sup>a</sup>	4.97±1.70 <sup>a</sup>	29.27±2.36 <sup>a</sup>	30.89±2.79 <sup>a</sup>	79.70±11.59 <sup>b</sup>	11.03±1.53 <sup>d</sup>

Values are measure of triplicate estimation Means in the same column followed by different letters are significant at  $P < 0.05$ , IG = *Irvingia gabonensis*.

a constituent of *Irvingia gabonensis*, as reported by Ekpe *et al.* might be another reason behind the higher flavonoid's concentrations that led to better antioxidant activity than ascorbic acid.<sup>32</sup> This can, in addition to the presence of high flavonoids contents be another reason

behind the above result. Hence, the plant seed have the potential to serve a medicinal purpose in the treatment of kidney diseases.

Alteration in the level of kidney serum biomarkers after administration of *Irvingia gabonensis* flavonoids-rich extract for two-weeks (Group 4 – Group 6) and four-weeks (Group 7 – Group 9).

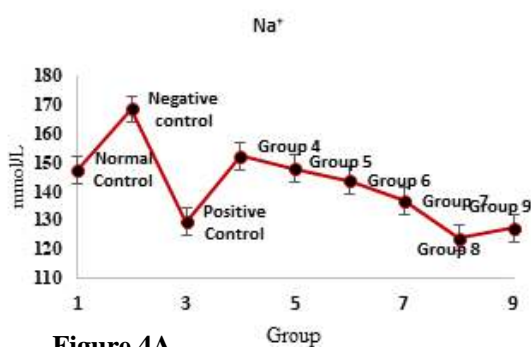


Figure 4A

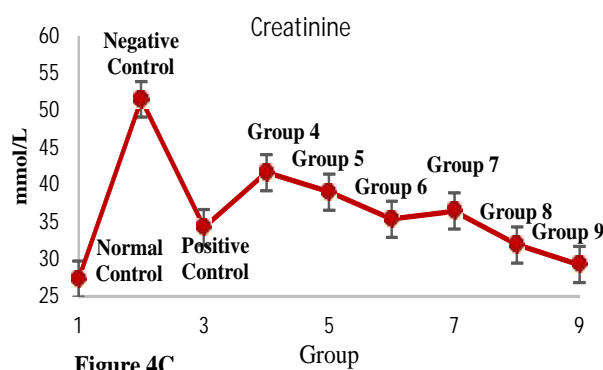


Figure 4C

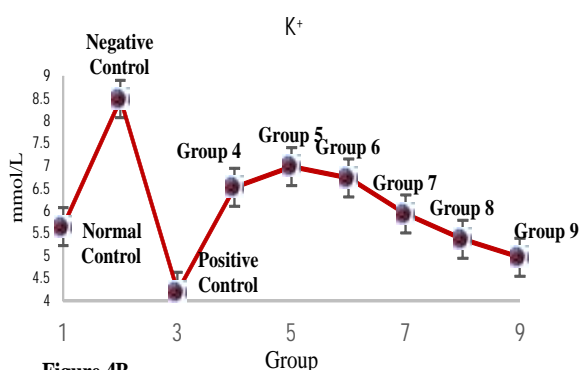


Figure 4B

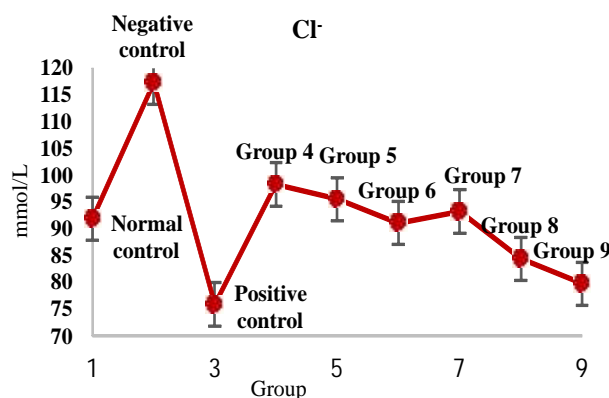


Figure 4E

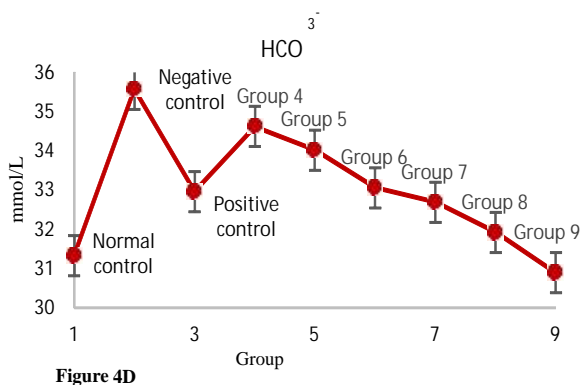


Figure 4D

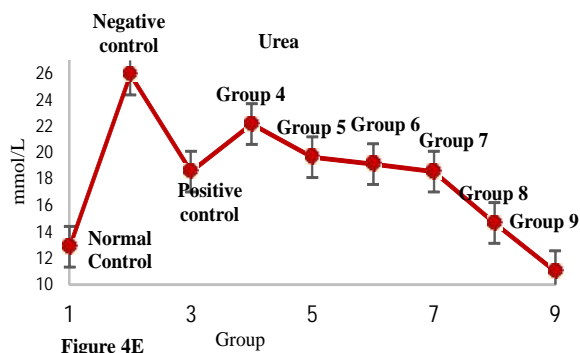


Figure 4E

**Figure 4A-E:** Showing the gradual alteration in the level of the kidney serum biomarkers after the administration of *Irvingia gabonensis* flavonoid-rich extract.

**Figure 4A-E:** The flavonoids-rich fraction of *Irvingia gabonensis* seed extract minimize acetaminophen-induced kidney damage. The serum biomarkers obtained from mice treated orally with acetaminophen revealed significant elevation ( $p < 0.05$ ) in sodium ( $\text{Na}^+$ ;  $147.93 \pm 8.56$  mmol/L), Potassium ( $\text{K}^+$ ;  $5.65 \pm 0.52$  mmol/L), Creatinine ( $27.29 \pm 2.96$  mmol/L), serum bicarbonate ( $\text{HCO}_3^-$ ;  $31.33 \pm 4.04$  mmol/L), serum chloride ( $\text{Cl}^-$ ;  $91.83 \pm 4.34$  mmol/L) and serum urea ( $12.87 \pm 2.42$  mmol/L). However, Mice treated with the flavonoids-rich fraction (groups IV to IX) record a promising decline in the kidney biomarkers level. The reduction in the biomarkers were found to be dose-dependent and time-dependent when compared withing groups treated with the extract. The normalization of the kidney biochemical parameters ( $\text{Na}^+$ , Creatinine, and  $\text{Cl}^-$ ,  $\text{K}^+$ ,  $\text{HCO}_3^-$  and urea) can be attributed to the flavonoids-rich content of the extract owing to the anti-inflammatory potential of flavonoids and the duration of extract administration on the mice. From figure 2 above, the effect of acetaminophen toxicity in the kidney is as a result of the oxidative degradation of lipids to malondialdehyde leading to the reduced anti-inflammatory function, followed by the activation of inflammation due to renal impairment of the kidney tubules. Based on this mechanism, the flavonoids-rich is assumed to have elicited a dose and time depended on anti-inflammatory response, thereby normalizing the increasing amount of serum kidney biomarkers. The medicinal potential of *Irvingia gabonensis* plant has been studied extensively.<sup>6, 33, 34, 35</sup> Therefore, the nephrocurative potential of *Irvingia gabonensis* seed is promising and should be harnessed by the pharmaceuticals in the development of either nutraceutical or drugs for kidney disease treatment.

Toxicity study of the plant reported no death from any of the experimental mice even at the highest dose of 3000mg/kgbw after 24hours of treatment and after the 7-

**Table 2: Acute Lethal Dose (LD50) Analysis on Experimental mice in phase I and II of Acute Toxicity Study**

Experimental Groups	Treatment	No. of deaths after 24 hours
<b>Phase I</b>		
1	10mg/kg bw IG extract	0/3
2	100mg/kg bw IG extract	0/3
3	1000mg/kg bw IG extract	0/3
Control	0	0/3
<b>Phase II</b>		
1	1000mg/kg bw IG extract	0/1
2	2000mg/kg bw IG extract	0/1
3	30000mg/kg bw IG extract	0/1

Mg/kg.bw = milligram/kilogram body weight, IG = *Irvingia gabonensis* LD<sub>50</sub> <= 30000mg/kg

day period of observation and no sign of toxicity was observed. Also, no sign of weight lost was recorded. Therefore, it is assumed that there is no percentage change in body weight. The oral LD<sub>50</sub> was therefore assumed to be higher than 3000mg/kgbw.

## CONCLUSION

The results obtained from this study have given an insight on the nephrocurative potentials of flavonoids-rich extract of *Irvingia gabonensis*. This reveals that *Irvingia gabonensis* seeds are promising source of alternative raw material for the pharmaceutical industries. Therefore, the flavonoids rich fraction of *Irvingia gabonensis* seed extract can be said to have curative effect on kidney diseases.

## Recommendation

Based on the promising findings of the study, further research into the various classes of flavonoids in *Irvingia gabonensis* seed and their mechanisms of action will shed light on the efficacy of the plant's medicinal effect. Consumption of the *Irvingia gabonensis* seed in traditional meals might be encourage especially in people already predisposed to kidney diseases.



### Conflict of interest statement

The authors declared that there are no conflicts of interest.

### Ethical Approval

All authors declare that "Principles of laboratory animal care" (NIH publication No. 85-23, revised 1985) were duly observed, as well as specific national laws where applicable.<sup>36</sup> All experiments have been examined and approved by the appropriate ethics committee.

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## REFERENCE

1. Mokaila, Aone. "Traditional Vs. Western Medicine-African Context". Drury University, Springfield, Missouri (2001). Archived from the original on 14 September 2006. Retrieved 11 March 2010.
2. Gurib-Fakim, T. Brendler, L. D. Phillips, and L. N. Eloff, Green Gold. Success Stories Using Southern African Plant Species, AAMPS Publishing, Mauritius. (2010); 25(3):472.
3. Manach C, A. Scalbert, C. Morand, C. Rémésy, and L. Jiménez, "Polyphenols: food sources and bioavailability," American Journal of Clinical Nutrition, Volume 79, Issue 5, (2004), Pages 727–747, <https://doi.org/10.1093/ajcn/79.5.727>
4. Betti, J. L.; Medicinal plants sold in Yaounde markets, Cameroon, African Study Monographs: (2004); 23(2), 47-64. From: <https://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.574.8891&rep=rep1&type=pdf>
5. Shiembo, P. N.; Newton, A. C.; Leakey, R. R. B; Vegetative propagation of *Irvingia gabonensis*, a West African fruits tree. Forest Ecology and Management. (1996); 87, 185-192, [https://doi.org/10.1016/S0378-1127\(96\)03781-4](https://doi.org/10.1016/S0378-1127(96)03781-4)
6. Ayuk E. T, Duguma B, Franzel Kengue, S, Mollet S. M, Tiki-Manga T, Zenkekeng P, Uses, management and economic potential of *Irvingia gabonensis* in the humid lowlands of Cameroon. Forest Ecology and Management, (1999); 113: 1-9. From: <http://www.sidalc.net/repdoc/A3487i/A3487i.pdf>
7. Franzel, S., Jaenicke, H. and Jansseen, W. 1996: Choosing the Right Trees: Setting Priorities for multipurpose Tree Improvement. ISNAR Res. Rep. 8, The Hague, International Service for National Agricultural Research; (1996). Retrieved from <https://www.researchgate.net/publication/275155129>
8. Ngondi, J.L., Oben, J.E. & Minka, S.R. The effect of *Irvingia gabonensis* seeds on body weight and blood lipids of obese subjects in Cameroon. Lipids Health Disiseas. (2005); 4, 12. <https://doi.org/10.1186/1476-511X-4-12>
9. Agbor, G. A. and Oben, J. E. Antioxidant Capacity of some Herbs/Spices from Cameroon: A comparative Study of two Methods. Journal of Agriculture Food Chemistry. (2005); 53, 6819-6824. <https://doi.org/10.1021/jf050445c>
10. Jamshidi-Kia F, Lorigooini Z, and Amini-Khoei H. Medicinal plants: past history and future perspective. Journal of Herbmmed Pharmacology; (2018); 7(1):1-7, <https://doi.org/10.15171/jhp.2018.01>
11. Etta H. E, Olisaeke C. C, Iboh, C. I. Effect of *Irvingia gabonensis* (Aubry-Lecomte ex O'Rorke) Seeds on the Liver and Gonads of the Male Albino Rats. Journal of Biology, Agricultural and Healthcare. Vol. 4(1), (2014): 10-15. <https://www.iiste.org/Journals/index.php/JBAH/article/view/10226>
12. Ewere G. E, Okolie N. P, Avan E. D and Umoh P. E. Comparative effects of ethanol leaf stem back extracts of *Irvingia gabonensis* (Bush Mango) on

- sodium arsenite-induced lipid profile perturbations in wistar rats. Springer, Clinical Pytoscience, (2021); 7:3; DOI: 10.1186/s40816-020-00241-5. <https://doi.org/10.1186/s40816-020-00241-5>
13. Karthivashan G, Arulsevan P, Tan SW, Fakurazi S. The molecular mechanism underlying the hepatoprotective potential of *Moringa oleifera* leaves extract against acetaminophen induced hepatotoxicity in mice. *Journal of Functional Foods* (2015); **17**:115-126. <http://dx.doi.org/10.1016%2Fj.jff.2015.05.007>
  14. Eguia L, Materson BJ. Acetaminophen acute renal failure without fulminant liver failure. *Pharmacotherapy*; (1997) 7:363-370. <https://pubmed.ncbi.nlm.nih.gov/9085330/>
  15. Karthivashan, G., Kura, A. U., Arulsevan, P., Md Isa, N., & Fakurazi, S. The modulatory effect of *Moringa oleifera* leaf extract on endogenous antioxidant systems and inflammatory markers in an acetaminophen-induced nephrotoxic mice model. *Peer Journal*, (2016); 4, e2127. <https://doi.org/10.7717/peerj.2127>
  16. Mukhtar, M. D. and Tukur, A. In-vitro Screening for Activity of *Pistia Stratiotes*. Extracts. *NISEB Journal* (1999); 1(1):51-60
  17. Efosa, G.E., Emmanuel, U. & Usunomena, U., 'Phytochemical composition, in-vitro antioxidant activity and acute toxicity of *Irvingia gabonensis* (O'Rorke) baill ethanolic leaf extract', *International Journal of Biological Research*, (2016). 4(1), 36-41. <https://doi.org/10.14419/ijbr.v4i1.5939>
  18. Laghari, Abdul, Memon, Shahabuddin and Nelofar, Aisha. Extraction, Identification and Antioxidative Properties of the Flavonoid-Rich Fractions from Leaves and Flowers of *Cassia angustifolia*. *American Journal of Analytical Chemistry*. (2011): 4236. <http://dx.doi.org/10.4236/ajac.2011.28100>
  19. Owaisi MA, Hadiwi NA, and Khan SA. GC-MS analysis, determination of total phenolics, flavonoid content and free radical scavenging activities of various crude extracts of *Moringa peregrina* (Forssk.) Fiori leaves. *Asai Pacific Journal of Tropical Biomedicine*. (2014); 4:964-970. <https://doi.org/10.12980/APJTB.4.201414B295>
  20. Lorke D. A new approach to practical acute toxicity testing. *Archive Toxicology*. (1983); 54:275-87. <https://doi.org/10.1007/bf01234480>
  21. Abdulmumin Y, Matazu K. I., Wudil A. M., Alhassan A., J., and Imam A. A. Nephrocurative effect of aqueous stem bark extract of *Boswellia Papyrifera* (DEL) against acetaminophen-induced kidney damage in Rats. *Ife Journal of science*. (2014); (16) Page no. 3. <https://www.ajol.info/index.php/ij/s/article/view/131685>
  22. Maruna, R.F.L. Colometric determination of sodium in human serum and plasma; *Clinical Chemistry Acta* (1959); 2:581
  23. Henry, R.F. *Clinical Chemistry Principle and Techniques*, 2<sup>nd</sup> edition, Harper and Row, Hagerstown, (1974); 1128-1135.
  24. Bartels H. and Bohmen M. A Colorimetric Method for Determination of Serum Creatinine. *Journal of Clinical Chemistry; Acta* (1972); 37:193, [https://doi.org/10.1016/0009-8981\(72\)90432-9](https://doi.org/10.1016/0009-8981(72)90432-9)
  25. Forrester RL, Wataji LJ, Silverman DA, Pierre KJ. Enzymatic method for determination of CO<sub>2</sub> in serum. *Clinical Chemistry*. (1976) Feb;22(2):243-5. PMID: 1248128. Retrieve from: <https://pubmed.ncbi.nlm.nih.gov/1248128/>
  26. Tietz, N.W. *Fundamental of Clinical Chemistry*, 2<sup>nd</sup> Edition, W.B. Saunders Co. Philadelphia. (1976); 874.
  27. Weatherburn MW. Phenol hypochlorite reaction for determination of serum urea. *Analytical Chemistry*. (1967) 39 (8):939-971
  28. Mgbemena, Nkoli Marynnn, Ilechukwu, Ifenna, Okwunodolu, Felicia Uchekukwu, Chukwurah, Joe-Vera Ogugua and Lucky, Isioma Blessing. "Chemical composition, proximate and phytochemical analysis of and peels, seed coat, leaves and seeds" *Ovidius University Annals of Chemistry*, vol.30, no.1, (2019); 65-69.

- <https://doi.org/10.2478/auoc-2019-0012>
29. Thavamoney, N., Sivanadian, L., Tee, L. H., Khoo, H. E., Prasad, K. N., & Kong, K. W. Extraction and recovery of phytochemical components and antioxidative properties in fruit parts of *Dacryodes rostrata* influenced by different solvents. *Journal of food science and technology*, (2018); 55(7), 2523–2532. Available from <https://doi.org/10.1007/s13197-018-3170-6>
30. Shirley, B. Flavonoids in seeds and grains: Physiological function, agronomic importance and the genetics of biosynthesis. *Seed Science Research*, (2008); 8(4), 415-422. <https://doi.org/10.1017/S0960258500004372>
31. Ezeabara, C. A. and Ezeani, D. S. Comparative study of phytochemical and nutrient contents of various parts of *Irvingia gabonensis*. *International Journal of Pharmacy and Chemistry*, (2016) ;2(2):10 ?14. DOI: 10.11648/j.ijpc.20160202.11
32. Ekpe Onot O.1; Nwaehujor Chinaka O.1; Ejiofor Charles E.2; Arikpo Peace W.1; Woruji Eliezer E.1 and Amor Emmanuel T.1. *Irvingia gabonensis* seeds extract fractionation, its antioxidant analyses and effects on red blood cell membrane stability; *Pharmacologyonline*, (2019); (1), 337-353. [https://pharmacologyonline.silae.it/files/archives/2019/vol1/PhOL\\_2019\\_1\\_A033\\_Ekpe.pdf](https://pharmacologyonline.silae.it/files/archives/2019/vol1/PhOL_2019_1_A033_Ekpe.pdf)
33. Ainge L, Brown N, A State of Knowledge Report Undertaken for the Central African Regional Program for the Environment. Oxford Forestry Institute, Department of Plant Sciences University of Oxford United Kingdom. (1998); pp. 24.
34. Okolo, chinedu O. Hohnson, and Patricia, B. Analgesic effect of *Irvingia gabonensis* stem bark extract. *Journal of Ethnopharmacology* (1995); Pp:45, 125-129.
35. Egras A. M, Hamilton, W. R T. L. Lenz, M. S. Monaghan, An evidence-based review of fat modifying supplemental weight loss products. *Journal of Obesity*, (2011); 1–7. <https://doi.org/10.1155/2011/297315>
36. NIH, Care and Use of Laboratory Animals. National Institute of Health Publication, Washington DC., (1998); 85-123. <https://grants.nih.gov/grants/olaw/guide-for-the-care-and-use-of-laboratory-animals.pdf>