# Determination of the Mineral Composition of the Pulp and Evaluation of the Acute Toxicity of the Seeds of Carica Papaya L. Consumed in Lubumbashi and Kinshasa in the Democratic Republic of the Congo

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**Method Article**

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# Abstract

**Background:** This work aimed on the one hand to determine the mineral and phytochemical composition of Carica papaya in order to guarantee the food safety of consumers, and on the other hand to evaluate the acute toxicity of papaya seeds.

**Methods:** The papayas were bought on the Mzee market in Lubumbashi and that of Selembao in Kinshasa. Fruit sampling was done according to the ISO 7002 standard on agricultural and food products; the papayas were firm, mature, without stains or physical damage.

**Results:** The analysis results of the papaya pulp showed both for the samples from the city of Lubumbashi and for the city province of Kinshasa that it contains respectively 85.87% and 84.46% water; 0.59% and 0.53% ash content. The mineral evaluation of our two samples presented a potassium content of 200±8 mg, magnesium 13.12±3mg, calcium 22.15±2mg, sodium 3mg±0.5 for the sample from Lubumbashi and 192, 32±8mg of potassium, 14.458±3mg of magnesium, 20.58±2mg of calcium and sodium 3.58±0.5mg for the sample from Kinshasa in macroelements. Concerning the trace elements, after analysis we found as zinc content (0.29 ± 0.1 mg and 0.12 ± 0.1 mg), copper (0.02 ± 0.01 mg and 0.14 ± 0.01mg), iron (2.22±0.5 mg and 2.04±0.5 mg) respectively for Lubumbashi and Kinshasa.

The chemical screening indicates the presence of alkaloids, saponosides, tannins catechics, flavonoids and anthocyanins in the palm wine and ethanolic extract of the seeds of Carica papaya and an absence of cyanogenic glycosides and gallic tannins.

**Conclusion:** With mild toxicity, the seeds of the fruit of Carica papaya L. can be used with moderate risk by the population.


# I. INTRODUCTION

For centuries and even millennia, men have used plants to relieve their pain, heal their ailments and heal their wounds. Currently, despite the progress of pharmacology, the therapeutic use of medicinal plants is very present in some countries of the world and especially in developing countries. Indeed, there are about 500,000 species of plants on earth, 80,000 of which have medicinal properties (1).

Among these plants is the species *Carica papaya* which belongs to the family of *Caricaceae*. The latter is known for its multiple biological activities, including its antioxidant and antimicrobial properties (2). This allows it to have an important role in the body's defense and in the fight against infections (Muhamad, 2005). The Carica papaya species is widely used in traditional medicine (leaves, roots, bark and seeds), to treat and relieve digestive and abdominal disorders such as: dyspepsia, gastric hyperacidity, dysentery and constipation (3). The plant is also used as an anti-inflammatory (4).

The plant produces a tasty fruit; highly appreciated by many consumers and is one of the fruits whose consumption is recommended. This fruit also has a high mineral density which allows it to reinforce mineral intake, without the risk of excess calories (5). Its multiple pharmacological properties are due to the richness of the plant in primary and secondary metabolites and to its mineral composition. Compounds present in the plant are among the most important groups of natural products due to their biological properties and structural diversity (6).

However, many plant species have their own toxicity, direct or indirect, which must be known before any use. Perfect knowledge of the constituents of a plant is therefore necessary.

Thus, this study aims to determine the mineral composition of papaya pulp as well as to evaluate in vivo the acute toxicity of the ethanolic extract of its seeds and of the seed macerate in palm wine from samples collected in Lubumbashi and Kinshasa in the Democratic Republic of the Congo, in order to guarantee food security for consumers.

# II. MATERIALS AND METHODS

The plant material consisting of fruits (papayas) was obtained at the main supply points, the Mzée market in Lubumbashi and Selembao market in Kinshasa.

The sampling was carried out according to the ISO 7002 standard for agricultural and food products. The selection of fruits took into account the quality criteria of the latter, in particular firm, mature papayas, without stains or physical damage. The botanical identification of these fruits was carried out in the laboratory of plant physiology and ecology, of the faculty of agronomic sciences of the University of Kinshasa.

The pulp of the fruit was used for the determination of the composition in mineral elements and the seeds for the determination of the acute toxicity. Moisture was determined by oven drying (AOAC 1997) at 105°C to constant mass. The total ashes were measured by calcination in a muffle furnace at 550°C (AFNOR, 1977b) until the white residues were obtained. Mineral elements were quantified by inductively coupled plasma spectrometry (7).

Phytochemical screening made possible to search for bioactive compounds (secondary metabolites) in particular alkaloids, coumarins, flavonoids, quinones, saponins, steroids, tannins and terpenoids. In addition, cyanogenic glycosides have been researched because they provide information on possible toxicity due to cyanide released by thermal or enzymatic hydrolysis.

The acute oral toxicity study of papaya seed extract in palm wine and ethanol was conducted according to OECD (Organisation for Economic Cooperation and Development) 425 guidelines. To do this, female mice of the NMRI SUISSE species were used. The method of Kraber and Behrens allowed the determination of the lethal dose 50 (LD50) by approximation by close calculation. The level of toxicity of the seeds was determined according to the Hodger and Sterner scale after calculating the LD50 (8).

# III. RESULTS

### 1. Determination of water content

The results above show that the pericardium of Lubumbashi papaya has a slightly higher water content than that of Kinshasa.

### 2. Determination of total ash content

The results above show that the pericardium of papaya from Lubumbashi has a slightly higher ash content than that from Kinshasa.

### 3. Determination of mineral content

Table 1
Mineral composition of dry matter of Carica papaya linn pulp

| Mineral elements | Lubumbashi (mg/100g) | Kinshasa (mg/100g). |
|---|---|---|
| Calcium | 22,15 ± 2 | 20,58 ± 2 |
| Cobalt | 0,00 | 0,00 |
| Chrome | 0,02 ± 0,01 | 0,01 ± 0,01 |
| Copper | 0,02 ± 0,01 | 0,14 ± 0,01 |
| Iron | 2,22 ± 0,5 | 2,04 ± 0,5 |
| Potassium | 200 ± 8 | 192,32 ± 8 |
| Magnésium | 13,12 ± 3 | 20,58 ± 3 |
| Sodium | 3 ± 0,5 | 3,58 ± 0,5 |
| Lead | 0,00 | 0,00 |
| Zinc | 0,29 ± 0.1 | 0,12 ± 0.1 |

The table above shows the contents of mineral elements present in the pericardium of papaya samples from Lubumbashi and Kinshasa

### 4. Extract Yields

Extraction by maceration of *Carica papaya* seeds in palm wine and ethanol had a yield of 34.07g or 154.86% for palm wine and 6.22g or 28.27% for the ethanolic extract. It should be noted that the value greater than 100% of the yield of the palm wine extract is due to the presence of other metabolites in the palm wine.

### 5. Phytochemical screening

Table 2
Phytochemical screening of Carica papaya Lubumbashi and Kinshasa seeds

| Code | Alk | Cou. | Anth | Flv. | Tannins | | Sapo. | Quinones | Ster. et terp | Hcn |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Catch. | Gal. | | | | |
| SLub | + | + | + | + | + | - | + | - | + | - |
| SKin | + | + | + | + | + | - | + | - | + | - |
| SLub: Sample from Lubumbashi; SKin: Sample from Kinshasa, Alc.: Alkaloids; Cou.: Coumarins; Anth. : Anthocyanins; Flv. Flavonoids; Sapo. : Saponins; Ster and terp. : Steroids and terpenoids; Catch. : Catechists; Gal.: Gallics, Hcn: Cyanogenic glycosides; +: presence; - : Absence | | | | | | | | | | |

**6. Assessment of acute toxicity**

• **Evolution of the weight growth of mice**

Table 3
Evolution of the weight growth of the mice during the treatment

| | J0 | J2 | J4 | J6 | J8 | J10 | J12 | J14 |
|---|---|---|---|---|---|---|---|---|
| **Groups** | **Weight in g** | | | | | | | |
| Control 1 | 19,23 | 19,2 | 19,35 | 19,80 | 20,63 | 20,75 | 20,72 | 20,02 |
| Control 2 | 19,20 | 19,3 | 20,32 | 20,65 | 19,96 | 20,56 | 20,81 | 20,54 |
| **1st batch of mice** | | | | | | | | |
| B1AE1 | 19,02 | 18,78 | 19,45 | 20,45 | 20,84 | 20,68 | 20,23 | 20,82 |
| B1AE2 | 19,23 | 20,14 | 20,12 | 20,05 | 21,32 | 21,06 | 21,25 | 21,01 |
| B1AE3 | 18,56 | 19,23 | 19,05 | 20,25 | 20,79 | 21,04 | 20,25 | 21 |
| **2nd batch of mice** | | | | | | | | |
| B2PW1 | 17,54 | 19,23 | 19,78 | 18,45 | 20,76 | 20,96 | 21,03 | 21,03 |
| B2PW2 | 19,72 | 19,03 | 19,98 | 20,58 | 20,59 | 20,75 | 21,45 | 21,36 |
| B2PW3 | 18,06 | 19,86 | 19,75 | 21,03 | 20,53 | 21,09 | 21,23 | 21,36 |
| - B1AE1, B1AE2, B1AE3: Alcoholic Extract of the seeds of *Carica papaya* | | | | | | | | |
| - B2PW1, B2PW2, B2PW3: Palm Wine Extract of the seeds *Carica papaya* | | | | | | | | |

• **Effect of treatment on biochemical parameters**

Table 4
Effect of palm wine and ethanolic extracts of Carica papaya seeds on the biochemical parameters of mice

| **Parameters** | **CREAT (mg/l)** | **UREA (g/l)** | **GOT (UI/l)** | **GPT (UI/l)** |
|---|---|---|---|---|
| Witness (control) | 5,50 ± 0,70 | 0,52 ± 0,02 | 106,60 ± 4,10 | 30,70 ± 0,14 |
| Batch 1 / ethanolic extract | 5,00 ± 1,00 | 0,56 ± 0,18 | 198,50 ± 51,08 | 47,30 ± 42,69 |
| Batch 2 / palm wine extract | 4,33 ± 0,57 | 0,59 ± 0,12 | 158,70 ± 35,17 | 25,33 ± 4,25 |
| p-value | 0,340 | 0,879 | 0,122 | 0,622 |
| **Decision** | NS | NS | NS | NS |
| S: significant difference, NS: non-significant difference | | | | |

### 7. Determination of LD50

After 14 days of observation, no death was observed in the treated mice, which did not allow the determination of the LD50. Oral administration of a single dose of 5000 mg/kg of palm wine and ethanol extract of Carica papaya seeds did not cause significant changes in NMRI mice. Indeed, no signs of toxicity such as a decrease in sensitivity to pain or noise or locomotion were observed during the 4 hours following the administration of the extracts. According to the classification of chemical products by Hodger and Sterne, our extracts are on scale 5 of products corresponding to a slightly toxic product (500 to 5000mg/kg).

# IV. DISCUSSION

Analysis of papaya pulp from the city of Lubumbashi (Mzée Market) and that of Kinshasa (Selembao Market) shows a water content of 85.87% and 84.46% respectively. This shows that this climacteric fruit is very perishable, hence the many losses observed in the fruit markets. The moisture content of papaya is therefore included in the range of water contents of fresh fruits; this result is almost similar to that of Morris at al (9); and Muhamad at al (10) who worked on the same variety of papaya and found a water content of 83.53%.

*Carica papaya* fruit pulp collected in Lubumbashi and Kinshasa contains 0.53% and 0.59% total ash content respectively. These results show that papaya could be an excellent source of minerals. Our results are lower than those of Larraurie at al (11) and Martinez et al (12) who found respective contents of 2.85% and 5% on mango and papaya in 100g of pulp.

With regard to the mineral elements, the analysis of the samples revealed at the level of the macroelements for 100 g of dry matter, a potassium content of 200 ± 8 mg, magnesium 13.12 ± 3 mg, calcium 22.15 ± 2 mg, sodium 3 ± 0.5mg for papaya from Lubumbashi (Mzee Market) and 192.32 ± 8mg of potassium, 14.458 ± 3mg of magnesium, 20.58 ± 2mg of calcium and sodium 3.58 ± 0.5mg for papaya from Kinshasa (Selembao Market). These results show some inequalities in terms of content which would surely be due not only to the difference in soil composition but also to different cultivation and climatic conditions (13). With a potassium (200 ± 8mg; 192.32 ± 8mg) and calcium (22.15 ± 2 mg; 20.58 ± 2mg) levels, our results are slightly higher than those found by Lobo and Pastor,2012 who have found as potassium value 182mg and 18.61mg as calcium content on papaya pulp.

Regarding the trace elements, the analyzes showed as zinc content (0.29 ± 0.1 mg and 0.12 ± 0.1 mg), copper (0.02 mg ± 0.01 and 0.14 ± 0.01 mg), iron (2 ,22 ± 0.5mg and 2.04 ± 0.5mg). The results are almost different between the two sampling sites because of the environmental parameters mentioned above. Compared to the results found by Lobo and Pastor (14), some of the results of this study are superior as is the case of copper and iron. The contents of other elements are lower than those of Lobo and Pastor (14), namely: sodium (9.61mg), zinc (1.97mg).

The yield of extracts obtained after maceration of papaya seeds in palm wine is much higher than that of ethanol.The yield obtained after maceration of papaya seeds in palm wine of 34.07g or 154.86% was higher than that of ethanol 6.22g or 28.27%. These results are contrary to those of the studies conducted by Amazu at al. (15), and Rasha at al (16) in which the extraction of papaya seeds with palm wine had a higher yield than those of methanol but lower than that of an extraction made with absolute ethanol (15, 16).

Phytochemical screening revealed the presence of alkaloids, saponosides, tannins (catechics), flavonoids and anthocyanins in the palm wine and ethanol extract of Carica papaya seeds and an absence of cyanogenic glycosides. These results confirm those reported by Mangambu at al. who found the same result in papaya seeds (17).

Oral administration of a single dose of 5000 mg/kg of the palm wine extract and the ethanolic extract of the seeds of *Carica papaya* did not cause significant changes and no deaths were reported in mice. No signs of toxicity such as decreased sensitivity to pain, noise or locomotion were observed. The extracts have a toxicity index equivalent to 5, according to the scale of toxicity of a chemical substance according to the LD50 and the route of administration established by Hodger and Sterner (8). Other studies on aqueous and ethanolic extracts of *Carica papaya* leaves at a dose of 5000 mg/kg did not lead to any toxic effect. Administration of palm wine and ethanol extracts of papaya seeds for 14 days promoted weight gain in mice. These results correspond to those found by Etame at al, whose palm wine extract had promoted weight gain in rats (18).

The biochemical analyzes carried out showed an increase in the enzymatic activity of ASAT for the two extracts and a decrease in the activity of ALAT. These results relate to those found by Etame et al. (18). After biochemical analyses, the latter found an increase in ASAT in both sexes and at all doses as well as a decrease in ALAT in rats. ALAT activity is more specific for liver damage than ASAT activity. However, in our study, the variations in enzyme activity were not significant, compared to the values in the group of control mice.

The serum urea and creatinine assay revealed that the administration of the extracts did not cause any significant change. These results also relate to those found by Etame et al. (18). Serum urea and creatinine are considered the main markers of nephrotoxicity (19).

# V. CONCLUSION

This study aimed to evaluate the mineral composition of the pulp as well as the acute toxicity of the seeds of the fruits of *Carica papaya linn* consumed in Kinshasa and Lubumbashi in DR Congo in order to guarantee the food safety of consumers and to contribute to the prevention of certain pathologies. Specifically, it involved quantifying mineral elements (macroelements and trace elements) and determining the phytochemical composition as well as evaluating the acute toxicity of *Carica papaya linn* seeds on laboratory mice.

The results of this study show that the fruit pulp of *Carica papaya* contains macro and trace elements with very distinct contents in potassium, magnesium, calcium, sodium, zinc, copper and iron in the dry matter. These contents differed insignificantly between the samples from Lubumbashi and Kinshasa. The results of the chemical screening show the presence of alkaloids, saponosides, catechic tannins, flavonoids, anthocyanins, coumarins, steroids and terpenoids in the palm wine and ethanol extract of the seeds of *Carica papaya linn*.

Extracts from the seeds of the fruit of *Carica papaya L*. can be used with a moderate risk for the population. Indeed, the oral administration of a single dose of 5000 mg/kg of the palm wine extract and the ethanolic extract of papaya seeds did not cause any sign of acute toxicity in mice. At this dose, no deaths were noted. This observation makes it possible to determine the LD50 of the extracts and to assign them a slight toxicity, according to the scale of Hodger and Sterner. No hepatic and renal damage was observed in mice.

# Declarations

### Ethics approval and consent to participate

Our Laboratory as a reseach one has the permission of the national ethics committee to use mice and human sample but the results in the last case are strictly confident according to the declaration of Helsinki.

### Consent for publication

Not applicable

### Competing interests

The authors declare that there is no competing interest regarding the publication of this paper

### Authors’ contributions

IG and JA drafted the manuscript. MM, DA, JA, NO, LM, MN, and KKK read, commented

the draft versions, and approved the final manuscript. All the authors read and

approved the final manuscript

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### Availability of data and materials

The datasets used and/or analysed during the current study are available

from the corresponding author on a reasonable request

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# Figures

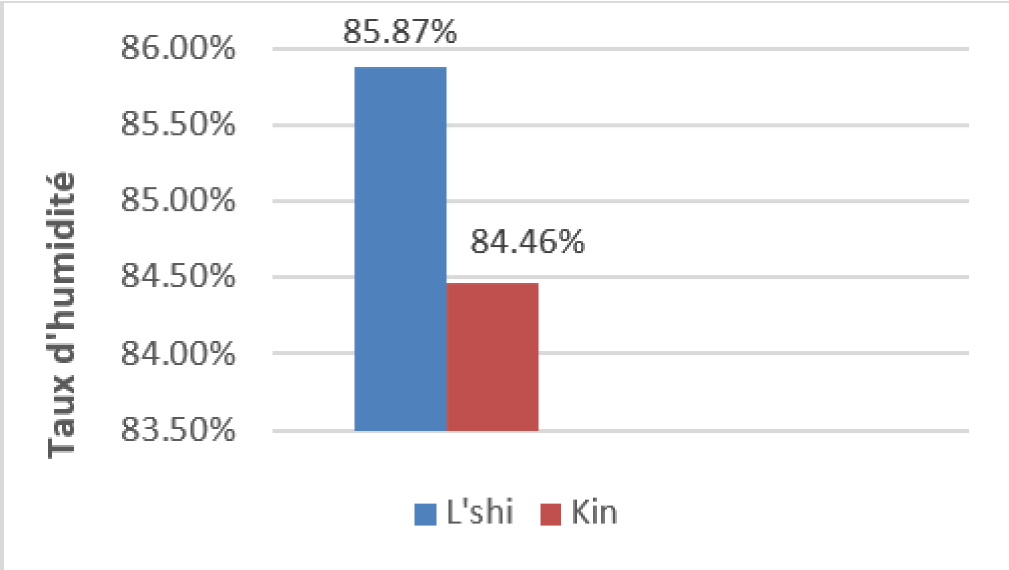


**Figure 1**

Water content of the pericardium of *Carica papaya linn* from Kinshasa and Lubumbashi

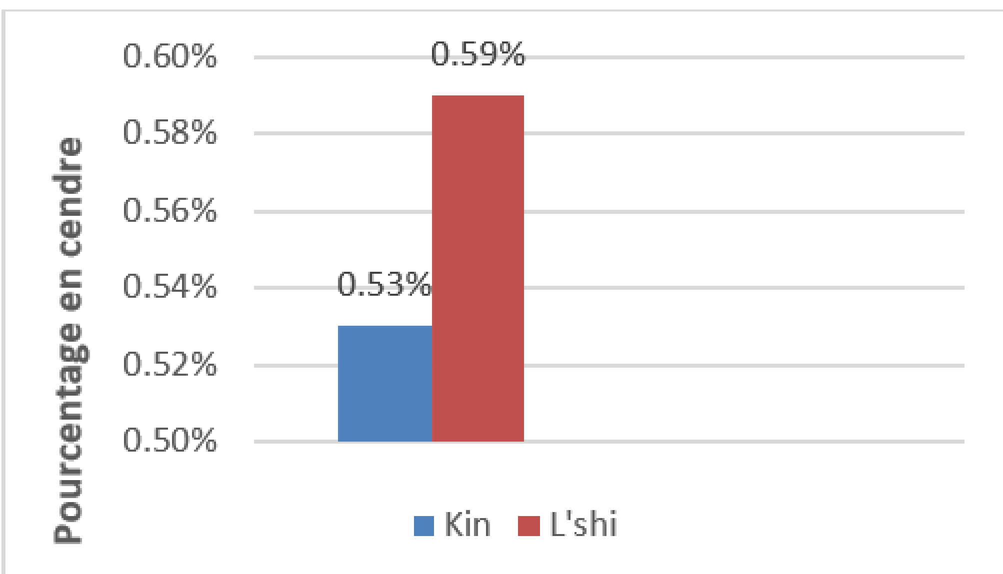


**Figure 2**

total ash content of the pericardium of *Carica papaya linn* from Kinshasa and Lubumbashi