INTRODUCTION

*Corresponding author
Ilochi Ogadinma

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C. nucifera is a popular fruit commonly called coconut [1], even though it is not a nut but a drupe [2]. C. nucifera is a mononuc perennial member of the family Arecaceae [3] and the only humanely edible species in the genus Cocos [4]. C. nucifera derived its name from a Spanish word ‘coco’ meaning ‘head’ or ‘skull’ [5]. C. nucifera produces a large quantity of clear liquid [4] which has an endospermal origin [2] and is called C. nucifera juice or ‘coconut juice’ [5].

The juice of coconuts is a popular drink in tropical countries [1], where it is usually sold commercially as fresh, canned or bottled [2] beverage. Some natives in Africa and South America allow coconut juice to ferment to coconut vinegar [6], which is a source of food [3]. Earlier studies have revealed the nutritional components of coconut juice [2, 3]. Some of these components include water, sugars, dietary fibres, essential and nonessential amino acids, vitamins B1, B2, B3, B5, B6, B9 and ascorbic acid[4], mineral calcium, iron, magnesium, manganese, potassium, zinc, sodium and selenium. In addition, recent studies listed some of the phytoconstituents of coconut juice, amongst which some agents that are believed to possess antioxidant [2], hypoglycaemic, immunostimulatory [1], hepatoprotective and anti-obesity [6] effects are included. Coconut juice has been reported to be of therapeutic importance in management of gastrointestinal [2], cardiovascular [3] and neuromuscular [4] disorders. The liver and kidney are both vital organs [7]. Without these organs, survival is impossible [8]. The liver is a metabolic organ [7]; the kidney is an excretory organ [8]. Every year, over 1 million individuals are diagnosed with either liver or kidney diseases [7, 8]. The cost of medical intervention for these diseases is above the average monthly income of a common man. The cost of dialysis in a developed country today is about 139 USD [8]. This estimated cost may be higher in a developing country. Besides the cost, some of these conventional methods of treatments have their own ‘baggage’. Conventional medicines are usually accompanied by adverse effects [9], may worsen the targeted condition or other existing conditions, may be effective for a short duration or may as well lack the ability to resolve the underlying disease condition [10]. Hepatic and renal diseases have remained a global challenge and are connected to increasing morbidity, mortality and healthcare expenditures [7, 8], and this is the impact statement of this study. It is therefore of immense importance to investigate the tendency or potential for C. nucifera to...
be therapeutically effective in the management of diseases relating to renal and hepatic functioning.

MATERIALS AND METHODS

Plant collection

Healthy unripe *C. nucifera* was purchased from a local market in Calabar, Cross River State.

Plant authentication

Each *C. nucifera* was identified and authenticated by Dr Ekeke Chimezie in the Herbarium for Department of Plant Science and Biotechnology, Faculty of Natural Sciences, University of Port Harcourt, Rivers State.

Preparation of juice

Each *C. nucifera* was properly washed with water and NaCl and then was cut open from one end of the exocarp with a Knife to expose the core of the fruit from which the juice was collected into a well covered transparent glass beaker. Apart from the sample taken to the laboratory for phytochemical screening, this preparation was done continuously before administration throughout the experimental period.

Phytochemical screening

The *C. nucifera* juice was taken to the laboratory of Pharmacognosy and Phytotherapy, Department of Pharmaceutical Sciences, Faculty of Pharmacy, Madonna University, Elele, Rivers State, for phytochemical screening. The phytoconstituents screened for include: Alkaloids, Flavonoids, Phenols, Tannins, Saponins, Carbohydrates, Steroids, oils and Terpenes. Phytochemical screening was done using standard laboratory methods [6].

Ethical consideration

Before the commencement of this study, an ethical approval and a recommendation was given by Madonna University Research Ethics Committee and Madonna University Clinical Research Approval Committee. An informed consent was also received from each sampled subject.

Experimental subjects

A survey was carried out on 180 male students resident in Madonna University. Simple random sampling technique was used, with each subject having an equal probability of selection (EPS). The age of the subjects sampled in this study was between 20 to 25 years. All test subjects were confirmed to be healthy by Dr Onyeso Godspower, a medical practitioner in Madonna University Teaching Hospital, Elele, Rivers State. In addition, the subjects were asked questions concerning their age, sex, weight, physical exercise and medical history.

The following pre-requisites served as exclusion criteria in sample selection:

- Ages below 20 years and above 25 years
- Weight below 60kg and above 80 kg
- Lifestyle report e.g. habits-alcoholics, drug abuse, drug dependence sedentary life style etc.
- Medical history indicating haemolytic diseases, liver and kidney infections etc.

Study design

The tests subjects were grouped into 3 (B, C, D) with 45 samples collected from each.

Treatment design

The subjects were grouped in 4, 45 subjects for each group.

- **A** = administered normal saline *ad libitum* and served as control group
- **B** = administered 50 ml of *C. nucifera* juice and served as Low dose group
- **C** = administered 100 ml of *C. nucifera* juice and served as Medium dose group
- **D** = administered 150 ml of *C. nucifera* juice and served as High dose group

Duration of study

This study lasted for a period of 42 days.

Blood sample collection

From each subject, 5ml of blood was collected into air tight bottles. Blood sample was collected from the median cubital vein. Blood sample collection was done on two (2) separate days;

- **Day 0** = early hours before onset of treatment on 0th day
- **Day 42** = early hours after onset of treatment on 42nd day

At the end of the experimental period, the blood samples were collected and subjected to centrifugation at 2000rpm using a Panasonic Autocentrifuge® and the serum was used for this study.

Biochemical analysis

The following biochemical analysis was conducted in the laboratory for hematology and chemical pathology, Madonna University and in the laboratory for clinical biochemistry, toxicology and biochemical pathology, University of Yaoundé.

Determination of serum liver function biomarkers

Liver function biomarkers assayed for include Aspartate transaminase (AST), Alanine transaminase (ALT) and alkaline phosphatase (ALP). This test was

carried out following standard procedures by Sandhya and Rajamohan, 2016 [7].

**Determination of kidney function biomarkers**

Kidney function biomarkers assayed for include creatinine (Cr), interleukin-18 (IL-18), Cystacin-C (CST3). This test was carried out using standard procedures by Clapp et al., 2009 [11].

**Statistical Analysis**

The statistical tool used for this study was SPSS version 20.0. Data was presented as Mean± Standard Error of Mean (SEM).

**RESULTS**

**Phytochemical compositions**

The phytoconstituents in *C. nucifera* juice include Alkaloids, Flavonoids, Saponins, Carbohydrates, Oils and Phenols.

**Basal level of liver enzymes**

Before the commencement of treatment, the table presents the basal level of the liver enzyme biomarkers. All values have no significant difference from the control values, A.

**Table-2: Phytochemistry of *C. nucifera* juice**

<table>
<thead>
<tr>
<th>Phytochemicals</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>++</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>+</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>-</td>
</tr>
<tr>
<td>Steroids</td>
<td>-</td>
</tr>
<tr>
<td>Oils</td>
<td>+</td>
</tr>
<tr>
<td>Phenols</td>
<td>++</td>
</tr>
<tr>
<td>Tannins</td>
<td>-</td>
</tr>
</tbody>
</table>

Key: + present – Absent

**Table-3: Effect of *C. nucifera* juice treatment on liver enzyme biomarkers (Day 0)**

<table>
<thead>
<tr>
<th>Treatments</th>
<th>AST (IU/L)</th>
<th>ALT(IU/L)</th>
<th>ALP (IU/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>23.2±0.2</td>
<td>34.3±1.4</td>
<td>122.3±2.1</td>
</tr>
<tr>
<td>B</td>
<td>21.4±0.4</td>
<td>35.4±2.3</td>
<td>119.6±2.0</td>
</tr>
<tr>
<td>C</td>
<td>24.3±1.4</td>
<td>34.2±1.0</td>
<td>124.1±1.4</td>
</tr>
<tr>
<td>D</td>
<td>23.1±0.1</td>
<td>34.2±1.3</td>
<td>123.1±1.7</td>
</tr>
</tbody>
</table>

Key: AST-aspartate transaminase, ALT-alanine transaminase, ALP-alkaline phosphatase

**Serum liver enzymes on day 42**

There was a significant decrease in liver enzyme biomarkers in treatment B for all liver enzymes AST, ALT and ALP at 24.2±0.70, 33.2±0.10 and 108.4±1.40 respectively, compared to treatment A and day 0 only for AST and ALT. There was also a significant decrease in all assayed liver enzymes compared to both treatment C and D. the values include 19.3±0.3a, 30.4±0.8a and 100.0±2.2a for AST, ALT and ALP respectively for treatment C. 18.0±1.0a, 27.2±0.4a and 87.2 ±1.7a for AST, ALT and ALP respectively for treatment D.

**Table-4: Effect of *C. nucifera* juice treatment on liver enzyme biomarkers (Day 42)**

<table>
<thead>
<tr>
<th>Groups</th>
<th>AST (IU/L)</th>
<th>ALT(IU/L)</th>
<th>ALP (IU/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>24.1±1.2</td>
<td>34.4±0.4</td>
<td>122.2±1.6</td>
</tr>
<tr>
<td>B</td>
<td>24.2±0.7</td>
<td>33.2±0.1</td>
<td>108.4±1.4</td>
</tr>
<tr>
<td>C</td>
<td>19.3±0.3</td>
<td>30.4±0.8</td>
<td>100.0±2.2</td>
</tr>
<tr>
<td>D</td>
<td>18.0±1.0</td>
<td>27.2±0.4</td>
<td>87.2 ±1.7</td>
</tr>
</tbody>
</table>

Key: AST-aspartate transaminase, ALT-alanine transaminase, ALP-alkaline phosphatase a0-statistically significant to A and day 0 P≤0.05, (H)- represents harmonic mean for each treatment group.

**Percentage change in liver enzymes from day 0 to 42**

For treatments C and D, there was a time dependent decrease in the liver enzymes AST, ALT and ALP at -20.6, -11.1 and -19.4 for treatment C respectively and -22.1, -20.5 and -29.2 for treatment D respectively. There was a time dependent decrease in only ALT and ALP for treatment B at -6.2 and -9.4 respectively.

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Table-5: Percentage change in liver enzyme biomarkers from day 0 to 42

<table>
<thead>
<tr>
<th>Treatments</th>
<th>AST (%c) 0→42</th>
<th>ALT (%c) 0→42</th>
<th>ALP (%c) 0→42</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>3.9</td>
<td>0.3</td>
<td>-0.08</td>
</tr>
<tr>
<td>B</td>
<td>13.1</td>
<td>-6.2</td>
<td>-9.4</td>
</tr>
<tr>
<td>C</td>
<td>-20.6</td>
<td>-11.1</td>
<td>-19.4</td>
</tr>
<tr>
<td>D</td>
<td>-22.1</td>
<td>-20.5</td>
<td>-29.2</td>
</tr>
</tbody>
</table>

Key: 0→42-day 0 to day 42

Kidney function enzymes day 0

Before the commencement of treatment, the table presents the basal level of kidney function biomarkers. All values have no significant difference from the control values, A.

Table-6: Effect of *C. nucifera* juice treatment on Kidney function biomarkers (Day 0)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Cr (μmol/L)</th>
<th>IL-18 (U/ml)</th>
<th>CST3 (U/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>29.4±0.4</td>
<td>16.2±3.2</td>
<td>13.1±2.0</td>
</tr>
<tr>
<td>B</td>
<td>28.7±0.2</td>
<td>15.4±2.1</td>
<td>13.2±2.4</td>
</tr>
<tr>
<td>C</td>
<td>29.0±1.0</td>
<td>16.4±3.1</td>
<td>12.7±2.7</td>
</tr>
<tr>
<td>D</td>
<td>29.1±0.4</td>
<td>16.7±2.4</td>
<td>13.4±2.3</td>
</tr>
</tbody>
</table>

Key: Cr- creatinine, IL-18- interleukin 18, CST3-cystatin C

Kidney function enzyme day 42

There was a significant decrease in treatments C and D compared to treatment A at 22.1±1.4a0, 12.1±0.3a0 and 10.4±1.2a0 for treatment C and 15.0±0.2a0, 10.2±0.4a0 and 10.2±1.4a0 for treatment D respectively.

Table-7: Effect of *C. nucifera* juice treatment on Kidney function biomarkers (Day 42)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Cr (μmol/L)</th>
<th>IL-18 (U/ml)</th>
<th>CST3 (U/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>29.6±1.4</td>
<td>16.2±0.4</td>
<td>13.2±2.1</td>
</tr>
<tr>
<td>B</td>
<td>28.0±1.0</td>
<td>15.0±1.0</td>
<td>13.1±1.4</td>
</tr>
<tr>
<td>C</td>
<td>22.1±1.4a</td>
<td>12.1±0.3a0</td>
<td>10.4±1.2a0</td>
</tr>
<tr>
<td>D</td>
<td>15.0±0.2a</td>
<td>10.2±0.4a0</td>
<td>10.2±1.4a0</td>
</tr>
</tbody>
</table>

Key: Cr- creatinine, IL-18- interleukin 18, CST3-cystatin C, a0-statistically significant to A and day 0 P≤0.05, (H)-represents harmonic mean for each treatment group.

Percentage change in kidney function enzymes from day 0 to 42

There was a time dependent decrease in kidney function biomarkers Cr, IL-18 and CST3 respectively.

Table-8: Percentage change in kidney function biomarkers from day 0 to 42

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Cr (%c) 0→42</th>
<th>IL-18 (%c) 0→42</th>
<th>CST3 (%c) 0→42</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>1.71</td>
<td>0</td>
<td>0.80</td>
</tr>
<tr>
<td>B</td>
<td>-2.43</td>
<td>-2.6</td>
<td>-0.80</td>
</tr>
<tr>
<td>C</td>
<td>-23.8</td>
<td>-26.2</td>
<td>-18.1</td>
</tr>
<tr>
<td>D</td>
<td>-48.5</td>
<td>-39.0</td>
<td>-24.0</td>
</tr>
</tbody>
</table>

Key: 0→42-day 0 to day 42

DISCUSSION

The phytoconstituents revealed in this study include alkaloids, flavonoids, saponins, carbohydrates, oils and phenols. The result of this phytochemical analysis is in agreement with earlier studies [1, 25]. Some of these phytoconstituents are believed to be hepatorenoprotective [11, 26], this has been reported by several scientists. From this study, there was no significant fluctuation in both liver and kidney biomarkers before commencement of treatments, so it was assumed that all samples were at basal level conditions [11, 12]. When dose of *C. nucifera* juice was increased, the level of these liver enzyme biomarkers reduced significantly. It can be deduced from this study that *C. nucifera* treatment has an inverse relationship with serum level of specific liver enzyme biomarkers.
AST, ALT and ALP. C. nucifera also caused a significant time-dependent decrease in liver enzymes AST, ALT and ALP, but this decrease was marked predominantly in treatments C and D, but only in ALT and ALP in treatment B. The juice significantly reduced kidney function biomarkers creatinine, interleukin-18 and cystatin-3. Creatinine is a breakdown product of creatine phosphate in muscle tissue [13]. It is an energy source [13] [14] as it replenishes the depleted adenosine triphosphate at the beginning of muscle contraction [15]. Elevated creatinine level signifies impaired kidney function [16] or the presence of kidney disease [15] [17]. As the kidneys become impaired for any reason, the creatinine level in the circulation will be elevated due to poor renal clearance [18]. Abnormally high levels of creatine thus warn of possible malfunction or failure of the kidneys [19]. Interleukin-18 is a proinflammatory cytokine. Its concentration increases in the kidneys after ischemia-reperfusion injury, alcohol ingestion and drug-induced renal injury [18, 19]. Cystatin is a protein encoded by CST3 gene [19]; it is a predominant biomarker of kidney function [20, 21]. If the kidneys are functionally normal, the blood creatinine, interleukin-18 and cystatin 3 level will be carefully regulated otherwise there will be an increase in its level [22, 23], which may be indicative of impaired glomerular filtration, renal tissue necrosis and hepatotoxicity [24, 25]. The time-dependent effective nature of C. nucifera may be caused by some of its bioactive phytoconstituents, as earlier reports have traced the therapeutic nature of various plants after prolonged treatments to their phytochemical components, most of which belongs to the classes of alkaloids, flavonoids, anthocyanins and phenols. From the results of this study, Whether C. nucifera will be effective or possess the potential in protecting the liver and kidney from pathologic changes will depend solely on the dose ingested. The dose determines the therapeutic efficiency of the C. nucifera juice. The mechanisms of action by which C. nucifera exhibits this protective function may probably be at mitochondrial level whereby it regulates metabolic activities within the cell and preventing cellular malfunction due to defective energy metabolism or at nuclear level whereby it affects the synthesis of essential proteins and membrane enzymes necessary for maintaining the integrity of the cell. Whatever effect C. nucifera juice has on the cell may be dependent on the dose ingested by the individual, and by other factors like duration and maybe the purity of the juice as the bioactivity of phytoconstituents of various plant materials may be inhibited by some foreign agents [26, 27]. This study provides scientific evidence that C. nucifera juice may be of medicinal importance in management of liver and kidney diseases.

CONCLUSION

From this study, C. nucifera juice has the ability to positively regulate some liver and kidney function markers in normal conditions. The phytoconstituents of C. nucifera may be responsible for its tendency to be hepatorenoprotective.

REFERENCES


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