

## COMPARISON OF THE EFFECT OF METHYLENE CHLORIDE AND AQUEOUS EXTRACT OF *GARCINIA KOLA* (HECKEL) SEED ON GASTRIC ACID SECRETION IN RATS

Anowi CF<sup>1</sup>, Ononiwu IM<sup>2</sup>, and Utoh-Nedosa UA<sup>3</sup>

<sup>1</sup>Department of Pharmacognosy and Traditional Medicine, Faculty of Pharmaceutical Sciences, Nnamdi Azikiwe University, Awka, Anambra State, Nigeria.

<sup>2</sup>Department of Pharmacology, Faculty of Medicine, University of Port Harcourt, Port Harcourt, Rivers State, Nigeria.

<sup>3</sup>Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, Nnamdi Azikiwe University, Awka, Anambra State, Nigeria.

### ABSTRACT

A study reported that intraperitoneally injected crude extracts of *Garcinia kola seed* and *kola acuminata seed* produced dose-dependent increases in gastric acid secretion in rats. This study investigated and compared the effects of aqueous or methylene chloride extract of *Garcinia kola seed* on gastric acid secretion in albino rats. Varying doses (2mg/ml; 4mg/ml; 6mg/ml; 8mg/ml and 10mg/ml doses) of aqueous or methylene chloride extract of *Garcinia kola seed* (equivalent of 13mg/kg, 26mg/kg, 40mg/kg, 53mg/kg and 67mg/kg) were infused into the stomach of 5 groups of randomly selected urethane-anesthetized male and female albino rats weighing 150-220g (test group, N=5; control group, N=5), through gastric tubes. The infusion moved at the rate of 15-20 drops per minute. The control animals were infused with 0.9% normal saline. The effluent collected after flushing the stomach of the test rats with the different doses of the extract (or the control rats with saline), were titrated against 0.001M sodium hydroxide to obtain the concentrations of HCl in millimoles in the effluents. Graphs of the acidity of the gastric contents in the effluents (in millimoles) plotted against the test doses of the extract (in mg/ml) showed that aqueous extract of *Garcinia kola seed* did not affect the basal gastric acid secretion in rat stomach (of 0.3mM acidity) but inhibited large increases in acidity of gastric contents beyond basal gastric acid secretion (increases above 1.53m M acidity). On the other hand, methylene chloride extract of *Garcinia kola seed* produced increases in both basal gastric acid secretion and large increases in gastric acid secretion far above basal levels (from 2mM to 5mM acidity). These results show that methylene Chloride used as a solvent for *Garcinia kola seed* extract had a direct gastric Hydrochloric acid (HCl) production-stimulation effect. The results also demonstrated that an organic solvent like methylene chloride can have direct gastric acid secretion-enhancement effects in a situation where the plant extract dissolved in it is exhibiting gastric acid secretion-inhibitory effects.

### INTRODUCTION

*Garcinia kola* seeds have bitter, astringent and gummy liquid extracts which are used in traditional African medicine to treat cough;

irritant, hoarse or sore throat; respiratory tract inflammation; dysentery and diarrhoea; emesis; diabetes; post partum haemorrhage; cuts; parasitic skin diseases; Guinea worm

infestation; liver cirrhosis. It is also used as a molluscicide; snake repellent; bitter tonic and astringent tonic. *Garcinia kola* extract/constituents have demonstrated anti-oxidant, anti-diabetic and liver-protective properties<sup>1, 2, 3</sup>.

This study compared the effects of stomach-infused 2mg/ml; 4mg/ml; 6mg/ml; 8mg/ml and 10mg/ml doses of aqueous extract of *Garcinia kola* seed on gastric acid secretion with those of infused 2mg/ml; 4mg/ml; 6mg/ml; 8mg/ml and 10mg/ml doses of methylene chloride extract of *Garcinia kola* seed.

### MATERIALS AND METHODS

Randomly selected male and female albino rats weighing 150-220g were anesthetized with intramuscularly administered 0.6/kg urethane and their stomachs were infused with 2mg/ml, 4mg/ml, 6mg/ml, 8mg/ml and 10mg/ml aqueous or methanol extract of *Garcinia kola* seed. The sampling size for each dose of aqueous or methylene chloride extract of *Garcinia kola* seed was N=5 for test group and N=5 for control group. The administered doses were equivalents of 13mg/kg; 26mg/kg; 40mg/kg; 53mg/kg and 67mg/kg aqueous or methylene chloride extracts of *Garcinia kola* seed.

Urethane used for anesthetizing the rats was made by dissolving 0.6mg/kg urethane crystals in normal saline. The point of establishment of anesthesia in each rat was judged as the point of the animal lost righting reflex.

Two per cent stock solutions of aqueous or methylene chloride extracts of *Garcinia kola* seed (10g in 100ml), were made by dissolving the *Garcinia kola* seed extract powder first in 5ml of dimethylsulphoxide (DMSO) and making up the volume with distilled water or methylene chloride. From the stock solution, serial dilutions were made to obtain 2mg/ml, 4mg/ml, 6mg/ml, 8mg/ml and 10mg/ml (which were equivalents of 13mg/kg; 26mg/kg; 40mg/kg; 53mg/kg and 67mg/kg) of aqueous or methylene chloride extracts of *Garcinia kola* seed.

The test and control rats were incised midsection at the throat and the trachea of each rat shunted with a flexible tube to maintain respiration. A tube was inserted into the oesophagus at a point proximal to the gastric (cardiac) sphincter (for in-flow of the extract or normal saline) and another inserted just after the pyloric sphincter (for out-flow of the gastric effluents). An infusion tube was connected to the oesophageal tube and its rate of discharge set

at 15-20 drops per minute. Normal saline was run into each animal for 20 seconds before the experiment to ensure that the tubing system was not blocked.

The test rats were infused with 13mg/kg; 26mg/kg; 40mg/kg; 53mg/kg or 67mg/kg) of aqueous or methylene chloride extract of *Garcinia kola* seed and the control animals were infused with 0.9% normal saline of the same dosage as the test rats. At the end of the experiment with each test dose, the gastric contents were flushed out and the effluent fluids were collected and titrated against 0.001M sodium hydroxide with phenolphthalein as the indicator.

Blank titrations were also run with the five doses of the aqueous *Garcinia kola* extract against 0.001M NaOH using Phenolphthalein as indicator.

The concentrations of HCl in millimoles secreted in the stomach of the test rats during the experiments were calculated using the equation:

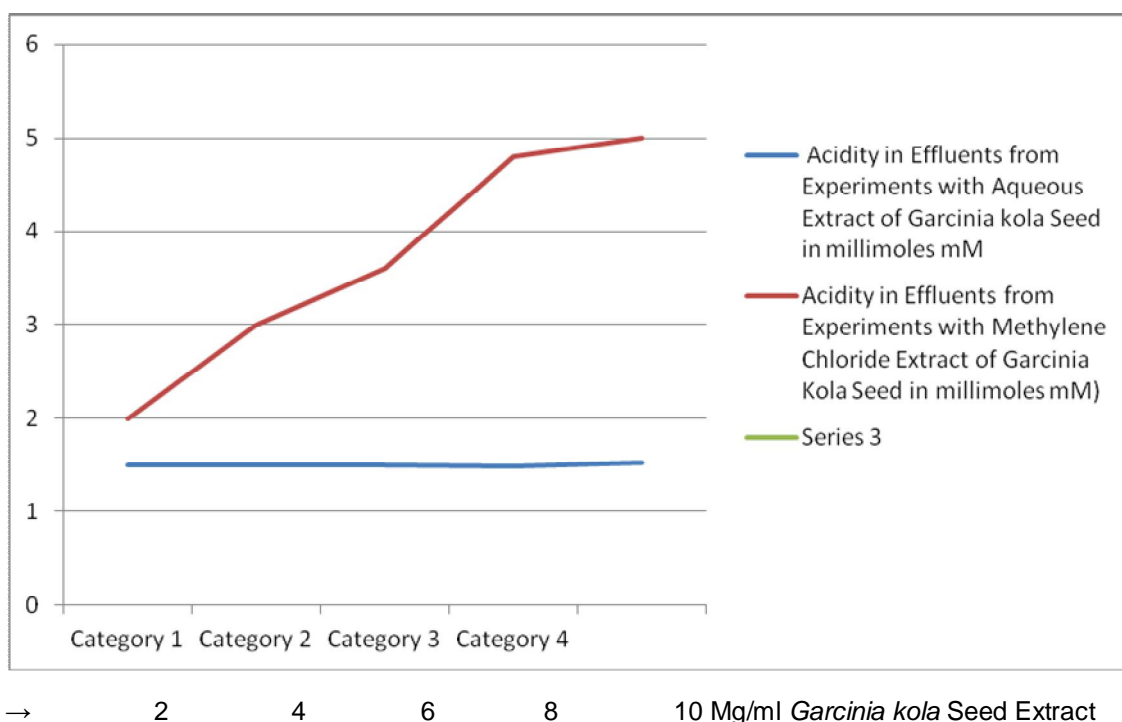
$$C_1 V_1 = C_2 V_2$$

(Where  $C_1$  and  $C_2$  are concentration of acid in effluent and concentration of Base respectively and  $V_1$  and  $V_2$  are volume of acid and volume of base respectively).

A Graph of the concentrations of secreted acid in the effluent fluids plotted against the 2mg/ml; 4mg/ml; 6mg/ml; 8mg/ml and 10mg/ml doses of methylene chloride extracts of *Garcinia kola* seed was made.

### RESULTS

The graphs of mean acidity in the effluent fluids of the five experiments plotted against the 2mg/ml, 4mg/ml, 6mg/ml, 8mg/ml and 10mg/ml doses of aqueous or methylene chloride extract of *Garcinia kola* shown in figure 1 demonstrated that aqueous extract of *Garcinia kola* seed had no effect on basal gastric acid secretion by the stomach of the tested rats (which increased the concentrations of acid in the effluent fluids from 1.5 millimoles [mM] to 1.53 millimoles [mM] and that aqueous *Garcinia kola* extract inhibited further increases in gastric acid secretion in the stomach beyond 1.5mM. Methylene chloride extract of *Garcinia kola* seed produced dose-dependent increases in gastric acid secretion beyond basal gastric acid secretion as it raised the acidity in the gastric effluents from 2 millimoles [mM] to 5 millimoles [mM]). In comparison, no effect on gastric acid secretion was observed in the control groups that received 0.9% normal saline.



**Fig. 1: Graph of Acidity of Effluents from Experiments with Aqueous or Methylene Chloride Extract of Garcinia Kola Seed against doses of the Extracts Used**

## DISCUSSION

*Garcinia kola* seed extracts have been shown to contain biflavonoids<sup>6,9</sup> which have demonstrated antioxidant<sup>1,3,4,5</sup> and scavenging properties<sup>5</sup>. They have also demonstrated antidiabetic<sup>3</sup>, testicular damage protective<sup>7</sup> and liver damage-protective<sup>2,10</sup> and antispasmodic<sup>11,12</sup> properties. Inhibition of increases in gastric acid secretion in rats by aqueous extract of *Garcinia kola* seed observed in this study is in agreement with the antioxidant<sup>1,3,4,5</sup> and immune defence scavenging properties<sup>5</sup> of *Garcinia kola* seed extract demonstrated in its antidiabetic<sup>3</sup>, testicular damage protective<sup>7</sup> and liver damage-protective<sup>2,10</sup> and antispasmodic<sup>11</sup> properties.

This shows that the stimulation of increases in gastric acid secretion in the test rats (but not in control rats) by methylene chloride extract of *Garcinia kola* seed observed in this study is a direct action of the solvent, methylene chloride, on gastric acid secretion.

## CONCLUSION

From the findings of this study, it is concluded that aqueous extract of *Garcinia kola* seed inhibited increases in gastric acid secretion in rats while methylene chloride extract of *Garcinia kola* seed stimulated increases in gastric acid

secretion in the test rats (when no inhibition or stimulation of gastric acid secretion was observed in control rats that received 0.9% normal saline) and therefore the stimulation of gastric acid secretion by methylene chloride extract of *Garcinia kola* seed observed in this study is due to a direct action of the solvent, methylene chloride, on gastric acid secretion.

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