



Evaluation of antioxidant and antacid activities of crude methanol and peptide extracts of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas*

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ABSTRACT

Background: There are many uses for therapeutic-based compounds in the food and pharmaceutical industries, and natural products are one of the best sources of these compounds. *Momordica charantia* (Family Cucurbitaceae), *Luffa cylindrica* (Family Cucurbitaceae), and *Jatropha curcas* (Family Euphorbiaceae) have been documented for various medicinal applications.

Objective: This study aimed to assess the anti-ulcer and antioxidant properties of *M. charantia*, *L. cylindrica* and *J. curcas* extracts and their crude peptide.

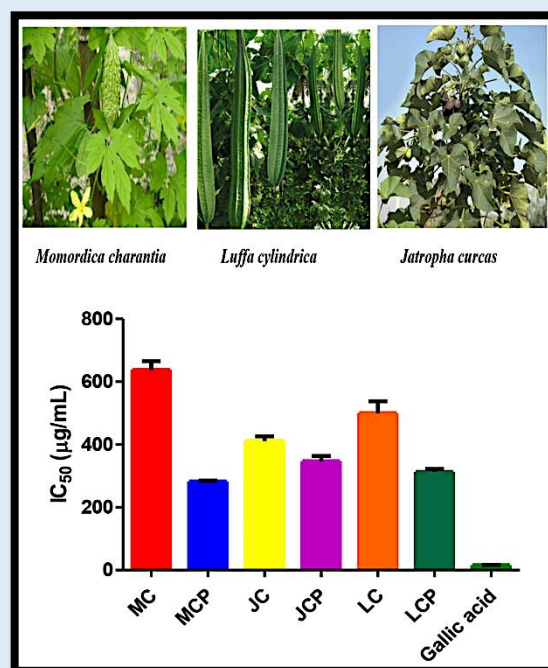
Methods: The plant samples were collected, identified, and authenticated at Forest Herbarium, Ibadan, Nigeria. The samples were allowed to air dry, were pulverized, and then were extracted using methanol. The peptidic principles were extracted using Dichloromethane: Chloroform: Water mixtures in the ratio 1:1:0.5 and concentrated *in vacuo*. The peptidic extracts were screened for peptide and peptide-related compounds by TLC bioautography using Ninhydrin, Pyridine, and modified G-250 reagents. The extracts were assessed for antioxidant (2,2-diphenyl-1-picrylhydrazyl, DPPH), free radical scavenging activity, total phenolic content (TPC), total flavonoid content (TFC), and antiulcer activity. The antiulcer activity was evaluated using the artificial gastric acid neutralizing activity.

Results: It was observed that all *M. charantia*, *L. cylindrica*, and *J. curcas* and their crude peptide extracts contain phenols and flavonoids. The TPC values of the crude methanol extracts were greater than those of the corresponding peptide extracts for all three species. The TFC of *M. charantia* and *L. cylindrica* crude methanol extracts were lower than their crude peptide extracts, while the TFC of *J. curcas* crude methanol extract was higher than the TFC value for its crude

peptide extract. The DPPH radical scavenging activity of the crude extracts and crude peptide extract of *M. charantia*, *L. cylindrica*, and *J. curcas* was low when compared to the standard (Gallic acid). The *M. charantia* crude peptide extract had the lowest IC₅₀ value of 282.4 µg/mL, indicating that it had better scavenging efficacy compared to the other extracts that were examined, while the lowest IC₅₀ value was obtained from *J. curcas* methanol extracts (411.7 µg/mL). The result from this study shows that *M. charantia*, *L. cylindrica*, and *J. curcas* and their crude peptide possess artificial gastric acid neutralizing activity, which implies that they can be used as antacids to reduce the acidity of the stomach. Hence, the plants were evaluated for their efficacy in managing peptic ulcers by their ability to reduce the acidity of the stomach by raising the pH of the stomach. This is the first report on the antacid potential of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas* in literature.

Conclusions: The results from this study support the ethnomedicinal claims that *M. charantia*, *L. cylindrica*, and *J. curcas* can be used in the management of peptic ulcers, and these plants could serve as sources of lead compounds to produce natural antiulcer drugs. There were traces of the presence of linear peptides and cyclic peptides (cysteine-rich cyclotides).

Keywords: Antiulcer, Antioxidant, Peptides, *Momordica charantia*, *Luffa cylindrica*, *Jatropha curcas*



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INTRODUCTION

In advanced countries, medicinal plant use has rekindled interest, while in 80% of developing countries, it is the primary treatment approach. Traditional herbal therapy is used to treat health issues by most people worldwide (87.5%) [1].

A unique class of pharmacological drugs known as therapeutic peptides is made up of many well-ordered

amino acids, most of which possess molecular weights ranging from 500 to 5000 Da [2]. Basic research on the physiological functions of naturally occurring human hormones, including insulin, oxytocin, vasopressin, and gonadotropin-releasing hormone (GnRH), in the human body, served as the foundation for the research into therapeutic peptides [3]. Following the 1921 synthesis of insulin, the first therapeutic peptide, significant

advancements have been made, resulting in the approval of more than 80 peptide medications on a global scale. Therefore, one of the most popular subjects in pharmaceutical research these days is the development of peptide medications [3].

Peptides are finding fast usage in the fight against a wide range of harmful microbes, including viruses, bacteria, fungi, and yeast. Tolerance to current medications, a dearth of efficient treatments, or newly discovered pathogens are the causes of this occurrence. For instance, most of this class of peptides are anti-microbial peptides (AMPs), which provide an alternative to the growing resistance to current antibiotics [4].

Open sores on the skin or mucous membranes, known as ulcers, are identified by an exposed epithelial cell lining and a deep lesion in a particular area that vitiates and causes bleeding [5]. A gastric ulcer occurs when it forms in the stomach. One of the most common gastrointestinal conditions worldwide, peptic ulcers are estimated to afflict 10% of people worldwide [5]. This type of ulcer is caused by the *H. pylori* bacteria. When taking antacids as a treatment for stomach ulcers, patients typically have improved symptoms after two to three weeks [6-7].

Thus, standard medications are often utilized to treat ulcers. A superficial tissue loss is indicative of an ulcer. Antiulcer medications and antacids generate 6.2 billion rupees in revenue and account for 4.3% of the Indian pharmaceutical market [5].

When taking antacids as a therapy for stomach ulcers, patients often have improved symptoms after two to three weeks. *Momordica charantia*, *L. cylindrica*, and *J. curcas* are traditionally used for the management of peptic ulcers in Nigeria and therefore selected for antacid activity.

Free radical damage to cells is prevented by antioxidants. It has been demonstrated that antioxidants can prevent or slow down the oxidation of other substances [8]. By eliminating radical intermediates and

oxidizing themselves, they have the power to stop chain reactions and prevent oxidation processes [9-10]. The body is rich in components that may either prevent or lessen the harm caused by free radicals.

Jatropha curcas L., sometimes referred to as fig nut, termite plant, and Barbados nut [11], belongs to the family Euphorbiaceae, and it produces edible oil seeds from its large shrub or small tree. This plant's parts are used to treat a variety of ailments in both people and animals [11].

The tropical vine known as bitter gourd (*Momordica charantia* L.), often called bitter melon or bitter apple from family Cucurbitaceae, is widely grown as a medicinal and vegetable crop in China, Southeast Asia, and India [12].

Luffa cylindrica, often known as the sponge gourd plant, is a member of the Cucurbitaceae family. The plant produces fiber-covered fruits that are encircled by a huge number of flat, blackish seeds that are native to Nigeria, Africa, and India. It has been used to treat fever, sinusitis, and asthma due to its nutritional and therapeutic qualities [13].

The purpose of this study was to assess the antioxidant and anti-ulcer potentials of the plant extracts of *M. charantia*, *Luffa cylindrica*, and *J. curcas* as well as that of their peptides.

MATERIALS AND METHODS

Plant collection, identification, and authentication:

Momordica charantia L., *Luffa cylindrica* (L.) M. Roem., and *Jatropha curcas* L., were collected at Orogun, Ibadan, Southwestern Nigeria. Plant identification and authentication were carried out at the Forest Herbarium Ibadan, (FHI) with corresponding voucher numbers FHI 112642, FHI 112643, and FHI 112839, respectively.

Preparation of extracts: The collected plants were dried and pulverized. The powdered samples were extracted with distilled methanol to obtain the crude extract. The

peptidic principles were extracted using Dichloromethane: Chloroform: Water mixtures in the ratio 1:1:0.5 and concentrated *in vacuo*. The peptidic extracts were screened for peptide and peptide-related compounds by TLC bioautography using Ninhydrin, Pyridine, and modified G-250 reagents. Crude extracts (250 mg and 100 mg) of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas* leaves and their corresponding crude peptide extracts were weighed separately and dissolved in 5 mL of methanol and then topped off with 250 mL of purified water.

Measurement of total phenolic content (TPC): The TPC of all the crude extracts of *M. charantia*, *L. cylindrica*, and *J. curcas* was evaluated following standard protocol [14]. The TPC was expressed as mg gallic acid equivalent (GAE)/g. Briefly, Folin-Ciocalteu 2.5 mL (diluted 10 folds) was added into 0.5 mL aliquot of each crude extract at 200 µg/mL and was then given a 3 min standing period after which 2 mL of (75 g/L) Na₂CO₃ solution in distilled water was added. Seven concentrations of standard gallic acid were prepared: 200, 100, 50, 25, 12.5, 6.25, and 3.125 µg/mL. Triplicates of the experiment were prepared. After the mixture's contents were well mixed, it was let to sit at room temperature for 30 min. Methanol (0.5 mL), 2.5 mL of Folin-Ciocalteu, and 2 mL of (75 g/L) Na₂CO₃ were used to set up the blank. After 30 min of incubation, the mixture's absorbance was measured at 765 nm using a UV-VIS spectrophotometer (Spectrumlab 752S). Using absorbance readings of gallic acid concentrations (200-3.125 µg/mL) at a wavelength of 765 nm, a linear dose-response regression curve was generated. The TPC in the plant extract was calculated using the formula:

$$\text{TPC} = \text{CV}/\text{M}$$

Where C is the equivalent gallic acid concentration determined from the calibration curve (µg/mL), V is the mixture volume (5 mL), M is the weight of the plant

extract (0.03 g), and TPC is the total phenolic contents mg GAE/g of the dry weight of extracts.

Measurement of total flavonoid contents (TFC): This study's aluminum chloride colorimetric approach followed standard procedures [15]. The calibration curve was compared to quercetin as a standard. Three milligrams of quercetin were dissolved in methanol and diluted in the range of 200, 100, 50, 25, 12.5, 6.25, and 3.125 µg/mL. Separately, 1.5 mL of methanol, 0.1 mL of 10% aluminum chloride, 0.1 mL of 1 M potassium acetate, and 2.8 mL of distilled water were combined with the diluted standard solution (0.5 mL). Three milligrams of test samples (*M. charantia*, *L. cylindrica*, and *J. curcas* crude extracts of the leaves and their corresponding crude peptide extracts) were dissolved in 20 mL of methanol, and then 0.5 mL of each extract and the reagents from the standard were thoroughly combined in a test tube. The same volume of distilled water was used in place of 10% aluminum chloride in the blank. The UV-VIS spectrophotometer (Spectrumlab 752S) was used to measure the absorbance of the reaction at 415 nm after the reaction was incubated for 30 min at room temperature. The presence of flavonoids was indicated by a yellow color. There were three runs of the experiment. The calibration curve was used to determine the flavonoid concentration based on the measured absorbance. The content of the flavonoid in the extract was expressed in terms of quercetin (mg quercetin/g of extract). The formula below was used to determine the plant extract's total flavonoid content:

$$\text{TFC} = \text{CV}/\text{M}$$

Where V is the volume of mixture (5 mL), M is the weight of plant extract (0.03 g), TFC is the Total Flavonoid Contents mg QE/g of dry weight of extracts, and C is the concentration of equivalent quercetin determined from calibration curve µg/mL.

DPPH radical scavenging activity: The free radical scavenging activity of the crude extracts (*M. charantia*, *L. cylindrica* and *J. curcas* crude extracts of the leaves and their corresponding crude peptide extracts) was evaluated using a standard method [16]. Using gallic acid as a positive control, the extract's capacity to donate hydrogen or scavenge radicals was used to assess its free radical scavenging activity. Three milliliters (0.004%) of freshly made 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH) from Sigma Aldrich were combined separately with 1 mL of methanol solution containing test samples and standard (gallic acid) at various concentrations (200, 100, 50, 25, 12.5, 6.25 µg/mL). Three milliliters of DPPH and 1 mL of methanol were combined to create the blank. The reaction mixtures were placed in the dark for 30 min while being incubated at room temperature. Using a UV-VIS spectrophotometer (Spectrumlab 752S), the absorbance was measured at 517 nm after 30 min and converted to a percentage of antioxidant activity. The results were recorded in triplicates. The degree of decolorization of DPPH from purple to yellow indicates the scavenging efficiency of the extract. Using the graph's equation and a calibration curve, the concentration of sample needed to scavenge 50% of the DPPH free radical (IC₅₀) was calculated. The percentage of inhibition of DPPH (%) was calculated as follows:

$$\% \text{ Inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of test sample}}{\text{Absorbance of control}} \times 100$$

The antioxidant activity of each sample was expressed in terms of IC₅₀.

Gastric acid neutralizing activity: The artificial gastric acid neutralizing assay was carried out [17-18].

Preparation of extracts: Crude extracts (250 mg and 100 mg) from *M. charantia*, *L. cylindrica* and *J. curca* leaves and their corresponding crude peptide extracts were separately weighed and dissolved in 5 mL of methanol and then topped off with 250 mL of purified water.

Artificial gastric juice preparation: In 500 mL of water, two grams of salt and 3.2 milligrams of pepsin enzymes were dissolved. After that, sufficient water and 7.0 mL of hydrochloric acid were added to yield a 1000 mL solution of artificial gastric acid in the stomach at a pH of 1.20.

Determination of pH of the prepared extracts: Out of the prepared extracts, 90 mL was measured in a conical flask and used for the pH determination at temperatures ranging from 25 °C – 37 °C. The pH values of the control solution were also determined according to a standard method [19].

Determination of the neutralizing effects on artificial gastric acids: Artificial gastric juices (100 mL) at pH 1.2 were mixed with 90 mL of each test solution. The pH values were calculated to investigate the neutralizing effect [19].

In vitro titration method of Fordtran's model for determination of the neutralization capacity: A 250 mL beaker containing around 90 mL of test samples was heated to 37°C. Using a magnetic stirrer, aeration was administered at 136 air bubbles per min to simulate peristaltic movements. Artificial gastric juice was used to titrate the test samples until the endpoint was reached at pH 3. The volume (V) of artificial gastric juice consumed was recorded. The measurement of the total hydrogen ion consumed (millimole) was 0.06309 (m Mol) × V (mL) [19].

Statistical analysis: For the statistical calculations, Graph Pad Prism Version 4.00 for Windows (Graph Pad Software Inc.) was employed. The collected experimental data were presented as mean ± SEM, where SEM stands for standard error of mean. The Dunnett Multiple Comparisons Test was used to do a one-way analysis of variance (ANOVA) and compare the groups. When p<0.01, the differences were statistically significant.

RESULTS

Total phenolic content (TPC) and total flavonoid content (TFC):

The Gallic acid equivalent/g of the extract was used to calculate the TPC using the Folin-Ciocalteu technique by comparison with the standard. Using the standard curve as a reference, the TFC are expressed as mg quercetin equivalent/g of extract. The study examined the total flavonoid and phenolic content of crude peptide extracts and methanol extracts of *M. charantia*, *L. cylindrica*, and *J. curcas*. The TPC of *M. charantia* methanol extract at 6.82 mg GAE/g is higher than its crude peptide extract which gave 5.84 mg GAE/g of extract. The TPC of the *L. cylindrica* methanol extract with 4.40 mg GAE/g has similar phenolic content as its crude peptide with 4.08 mg GAE/g of extract. The TPC of the *J.*

curcas methanol extract at 5.17 mg GAE/g is higher than its crude peptide extract which gave 2.47 mg GAE/g of extract as shown in Table 1.

The TFC of the methanol leaf extracts of the plants and their crude peptide extracts were expressed as mg QE/g of extract. The TFC of methanol extract of *M. charantia* leaves, at 5.90 mg QE/g is lower than its crude peptide extract which gave 8.48 mg QE/g of extract, The TFC of *L. cylindrica* methanol extract with 5.90 mg QE/g is lower in flavonoid content when compared to its crude peptide which had 8.13 mg QE/g of extract. The TFC of *J. curcas* methanol extract at 7.39 mg QE/g is similar to the TFC of its crude peptide extract which is 6.54 mg QE/g of extract as shown in Table 1.

Table 1. Total Phenolic Content (TPC) and Total Flavonoid Content (TFC) of the crude methanol extract and crude peptide extracts of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas*

Extract	TPC (mg GAE/g)	TFC (mg QE/g)
Momordica charantia methanol extract	6.82±0.04	5.90±1.38
Momordica charantia peptide extract	5.84±0.38	8.48±1.47
Jatropha curcas methanol extract	5.17±0.25	7.39±0.22
Jatropha curcas peptide extract	2.47±0.25	6.54±0.59
Luffa cylindrica methanol extract	4.40±0.12	5.90±1.56
Luffa cylindrica peptide extract	4.08±0.45	8.13±0.05

DPPH radical scavenging assay: Figure 1 illustrates the concentration-dependent 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging activity of the three plants' crude peptide and methanolic extracts. The scavenging activity is expressed as a percentage of the ratio of the decrease in absorbance of the test solution to that of the DPPH solution without the plant extracts. It was shown that the plant extracts' capacity to scavenge free radicals depended on concentration, with 200 µg/mL concentration having the highest activity for all extracts, while 6.25 µg/mL had the lowest across all the samples

tested; so, with an increase in concentration, an increase in activity was observed.

The crude methanol extracts of the plants and their crude peptide extracts showed scavenging activity with IC₅₀ values ranging from 282.4 to 637.7 µg/mL.

The methanol extract of *M. charantia* had the highest IC₅₀ value (637.7 µg/mL), indicating minimal scavenging activity, while the crude peptide extracts of the species had the lowest IC₅₀ value (282.4 µg/mL), indicating better scavenging activity than the other extracts screened.

For the percentage inhibition, *Luffa cylindrica* crude peptide extracts had the highest free radical scavenging activity (FRSA) of 36.65% at 200 $\mu\text{g}/\text{mL}$. *J. curcas* crude

peptide extract had the lowest free radical scavenging activity (FRSA) of 14.27% at 200 $\mu\text{g}/\text{mL}$ (Table 2).

Table 2. Percentage Free radical scavenging activity (FRSA) of the crude methanol extracts and crude peptide extracts of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas*

Percentage FRSA at different concentrations (%)						
Samples	200 $\mu\text{g}/\text{mL}$	100 $\mu\text{g}/\text{mL}$	50 $\mu\text{g}/\text{mL}$	25 $\mu\text{g}/\text{mL}$	12.5 $\mu\text{g}/\text{mL}$	6.25 $\mu\text{g}/\text{mL}$
LC	27.75 \pm 2.32	25.70 \pm 0.59	16.83 \pm 4.794	21.35 \pm 7.32	13.46 \pm 3.99	15.76 \pm 3.23
LCP	36.65 \pm 2.04	21.63 \pm 2.12	14.05 \pm 1.22	11.71 \pm 1.37	10.86 \pm 3.62	6.811 \pm 2.12
JC	28.25 \pm 3.03	14.17 \pm 3.34	10.52 \pm 2.19	8.91 \pm 3.811	0.80 \pm 2.31	0.09 \pm 6.34
JCP	14.27 \pm 1.01	8.96 \pm 0.65	5.78 \pm 0.80	6.81 \pm 2.70	2.57 \pm 2.09	0.94 \pm 0.48
MC	26.07 \pm 2.51	15.03 \pm 3.77	19.48 \pm 3.098	14.08 \pm 1.04	14.19 \pm 3.74	8.95 \pm 1.25
MCP	35.16 \pm 0.92	24.56 \pm 0.55	17.55 \pm 2.75	10.79 \pm 1.62	7.48 \pm 1.31	3.31 \pm 1.01

Key: JC- *Jatropha curcas*; JCP- *Jatropha curcas* Crude Peptides; MC- *Momordica charantia*; MCP- *Momordica charantia* Crude Peptides; LC- *Luffa cylindrica*; LCP- *Luffa cylindrica* Crude Peptides

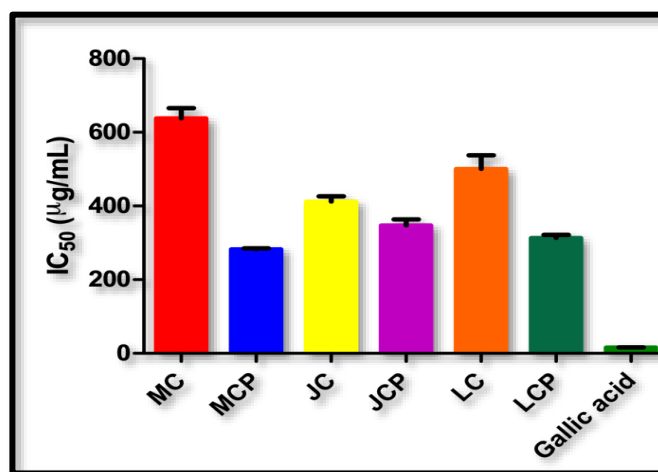


Figure 1. IC₅₀ values of the DPPH Free Radical Scavenging Activity of the crude methanol extracts and crude peptide extracts of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas*

Artificial gastric acid neutralizing activity: The artificial gastric acid neutralizing activity of *M. charantia*, *L. cylindrica* and *J. curcas* and their crude peptide extracts is shown in Table 3. *Luffa cylindrica* methanol extract (100 mg/250 mL) showed the highest artificial gastric acid

neutralizing activity by increasing the pH of the artificial gastric acid from 1.2 to 1.63 \pm 0.01, which is comparable to the pH increase of the artificial gastric acid from 1.2 to 1.54 \pm 0.04 achieved with standard NaHCO₃ (100 mg/250 mL). *Jatropha curcas* crude peptide extract (200 mg/250

mL) was next in activity which raised the pH of artificial gastric acid from 1.2 to 1.58 ± 0.02 , while the crude peptide extract of *M. charantia* (100 mg/250 mL) and *J. curcas* methanol extract both had the lowest artificial gastric acid neutralizing activity raising the pH from 1.2 to 1.49 ± 0.01 . The negative control (distilled water) raised the pH of artificial gastric acid from 1.2 to 1.54 ± 0.04 (Table 3).

Luffa cylindrica crude peptide extracts had the highest action efficiency against artificial gastric acid (the amount of artificial gastric acid needed to bring the pH of the plant extract down to 3). It consumed 150.00 ± 0.00 mL of artificial gastric acid. *L. cylindrica* methanol extract (100 mg/250 mL) consumed 149.00 ± 1.00 of artificial gastric acid for its pH to adjust to 3, while *M. charantia* methanol extract has the lowest in that it consumed 95.50 ± 0.50 artificial gastric acid (Table 3).

Table 3. Artificial gastric neutralizing activity of the crude methanol extracts and crude peptide extracts of *Momordica charantia*, *Luffa cylindrica* and *Jatropha curcas*

Extract/Standard	Concentration (mg/250 mL)	pH	Neutralizing efficiency	Action efficiency (Amount of AGT consumed in mL)	Number of H ⁺ ion consumed
Sodium carbonate	200	8.48	1.51 ± 0.04	143.5 ± 5.50	9.06 ± 0.35
	100	8.16	1.54 ± 0.04	138.75 ± 2.25	8.76 ± 0.15
JC	200	6.45	1.53 ± 0.04	100.00 ± 0.00	6.31 ± 0.00
	100	6.47	1.49 ± 0.01	97.50 ± 0.50	6.15 ± 0.03
JCP	200	6.22	1.58 ± 0.02	99.00 ± 1.00	6.25 ± 0.07
	100	6.4	1.56 ± 0.03	99.50 ± 0.50	6.28 ± 0.03
MC	200	6.53	1.55 ± 0.05	95.50 ± 0.50	6.03 ± 0.04
	100	6.51	1.51 ± 0.01	99.00 ± 1.00	6.25 ± 0.65
MCP	200	6.2	1.53 ± 0.03	90.00 ± 5.00	5.68 ± 0.32
	100	6.2	1.49 ± 0.01	145.00 ± 5.00	9.15 ± 0.32
LC	200	6.3	1.55 ± 0.01	149.00 ± 1.00	9.40 ± 0.06
	100	6.41	1.63 ± 0.01	144.90 ± 0.10	9.15 ± 0.01
LCP	200	6.09	1.55 ± 0.01	99.00 ± 1.00	6.25 ± 0.07
	100	6.26	1.52 ± 0.00	150.00 ± 0.00	9.46 ± 0.00
*Water	100	6.19	1.30 ± 0.00		

Key: JC- *Jatropha curcas*, MC- *Momordica charantia*, LC- *Luffa cylindrica*, JCP- *Jatropha curcas* Crude Peptides, MCP- *Momordica charantia* Crude Peptides, LCP- *Luffa cylindrica* Crude Peptides

DISCUSSION

The most significant bioresource of pharmaceuticals for traditional medicine, contemporary medicine, nutraceuticals, dietary supplements, and other uses has been found in medicinal plants over time. In national healthcare programs, the World Health Organization (WHO) supports, advises, and promotes traditional and herbal therapies since they are widely accessible, inexpensive, safe, and well-received by the public. Scientific research on medicinal plants was considered in the literature to determine their economic and medicinal significance [20]. Medicinal plants have been proven to possess phytochemical and antioxidant activities [6, 10, 15-16, 21]. This study aimed to assess the antioxidant and antiulcer capabilities of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas* extracts as well as that of their peptides.

In the study, it was observed that all *M. charantia*, *L. cylindrica* and *J. curcas* and their crude peptide extracts contain phenols and flavonoids. The TPC of the crude methanol extracts was higher than their corresponding peptide for all three species. The TFC of *M. charantia*, *L. cylindrica* crude methanol extracts were lower than their crude peptide extract, while the TFC of *J. curcas* crude methanol extract was higher than the TFC value for its crude peptide extract (Table 1).

A recent study showed a high TPC in *M. charantia* aqueous extract. The DPPH result demonstrated that, in comparison to the standard ascorbic acid used in the study, the aqueous extract at higher doses significantly inhibited DPPH [20].

In another investigation, the TPC values for *M. charantia*'s aqueous fruit extract were comparable to those for the plant's crude methanol and crude peptide extracts. The TFC value of the crude methanol extracts and crude peptide extract of *M. charantia* was found to be greater in this study than it was in the leaf ethanolic extract [22]. Previous research also confirmed the antioxidant efficacy of *M. charantia* extract [23-24].

Additionally, it was noted that the crude peptide and crude methanol extracts of *L. cylindrica* had TPC values that were comparable to those of the ethyl acetate extract of *L. cylindrica* fruits previously reported [25]. This variation is probably due to the usage of different morphological parts and solvent of extraction. The TPC values obtained for crude methanol and crude peptide extracts of *J. curcas* were similar to the values observed for the leaves and stem bark of *J. curcas* in another research [26]. The TFC values obtained were significantly higher than the values reported [26] for the leaf and stem bark of *J. curcas*. This variation is probably due to the usage of different morphological parts. These findings from the current investigation indicated that the phenolic and flavonoid compounds that were recovered in the various solvents varied greatly in terms of kind and nature. Strong antioxidants found in plants have been demonstrated to improve human health whether taken orally or medicinally, delaying the aging and cancer-causing processes [27]. Researchers are presently focusing on natural antioxidants rather than synthetic antioxidants since the former is more effective than the latter [28-29]. The antioxidant activity of *J. curcas* was previously reported [30-32]. The IC₅₀ value, or the amount required to scavenge 50% of DPPH, is a measure of an antioxidant compound's capacity to scavenge radicals and is calculated using the DPPH technique. The antioxidant capacity of plant materials may be measured using a variety of techniques. No single testing technique can give a complete review of a sample's antioxidant profile due to the intricacy of the oxidation/antioxidation process. For the percentage inhibition, *L. cylindrica* crude peptide extracts had the highest free radical scavenging activity while *Jatropha curcas* crude peptide extract had the lowest free radical scavenging activity (FRSA).

The radical scavenging activity (DPPH) of the crude extracts and crude peptide extract of *M. charantia*, *L. cylindrica* and *J. curcas* when compared to the standard

(Gallic acid) was low. The IC₅₀ of the peptide extract of all three species was lower than the methanol extract of the plant species which implies more activity than the methanol crude extract. The IC₅₀ value obtained for *M. charantia* is higher than the reported IC₅₀ value of DPPH scavenging capacities of ethanolic fruit extract of *M. charantia* from previous research [33] which implies less activity. *J. curcas* has an IC₅₀ value that is comparable to a previous report [34]. In the investigation by [25], the stem bark showed limited activity (high IC₅₀).

The phytochemical screening revealed that *M. charantia*, *L. cylindrica*, and *J. curcas* have low antioxidant properties, which may be related to their low flavonoid and their phenolic content. The most substantial phytochemical components produced by plants are phenolic compounds, which include flavonoids, phenolic acids, and tannins [35].

The result also revealed that these three plants have high antiulcer activity, suggesting that other constituents working in synergism with their low flavonoid and phenolic content are what give these plants their antiulcer properties [36].

The antiulcer activity of *M. charantia*, *L. cylindrica*, and *J. curcas* methanol extracts and their crude peptide extracts was evaluated using the artificial gastric acid neutralizing activity.

Antacids neutralize stomach acid to treat ulcers; they do not, however, lessen the number of gastric secretions. When prescribing antacids, physicians should consider that different antacid medications differ significantly in their *in vivo* and *in vitro* potencies. However, there are other factors to consider while selecting an antacid apart from potency. Other significant factors are price, taste, salt content, bowel habits, and side effects. Despite being widely utilized, antacid side effects and medication combinations pose a serious clinical issue. As a result, interest in using traditional

Chinese herbal treatments to treat ulcer illness has grown recently [19].

The findings of this investigation indicate that the crude peptides of *M. charantia*, *L. cylindrica*, and *J. curcas* can neutralize artificial gastric acid, suggesting that they may be utilized as antacids to lessen stomach acidity. Hence, the plants were evaluated for their suitability for managing ulcers by their ability to reduce the acidity of the stomach by raising the pH of the stomach. This is the first report on the antacid potential of *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas* in literature. Despite reports that the three plant species have been used ethnobotanically to treat ulcers, no thorough investigation has been conducted to assess the plants' antacid properties. However, there are other antiulcer reports on all three plant species; there was a previous report of the antiulcer activity of *M. charantia* methanol extract in rats [37]. The antiulcerogenic activity of ethyl acetate leaf extracts of *L. cylindrica* has also been reported [38], while another study investigated the antiulcer activity of methanolic extract of *J. curcas* on Aspirin-induced gastric lesions in Wistar strain rats [39] with few other studies on antiulcer activity of medicinal plants [40-42].

The results demonstrated that all the methanol crude extracts of *M. Charantia*, *L. cylindrica*, and *J. curcas* and their crude peptide extracts had artificial gastric acid neutralizing activity in that they were able to raise the artificial gastric acid pH from 1.2 to 1.53±0.04 for *J. curcas*, 1.58±0.02 for *J. curcas* crude peptide, 1.55±0.05 for *M. charantia*, 1.53±0.03 for *M. charantia* crude peptide, 1.63±0.01 for *L. cylindrica*, and 1.55±0.01 for *L. cylindrica* crude peptide when compared to that of the standard NaHCO₃ (1.51±0.04). Our research indicates that the crude peptide extracts of *M. charantia*, *L. cylindrica*, and *J. curcas* have an artificial stomach ulcer neutralizing effect, suggesting that they may be utilized to treat ulcers.

Additionally, it was found that *J. curcas* and *M. charantia* had higher neutralizing efficiency by significantly raising the pH of artificial gastric acid than their crude peptide counterparts when the artificial gastric acid neutralizing activity of the plant extracts and crude peptide extract was assessed. This study also discovered that the crude peptide extracts of *M. charantia*, *L. cylindrica*, and *J. curcas* have gastroprotective qualities. Acid and pepsin are necessary for the development of peptic ulcer disease. Theoretically, antacid treatment has two advantages: it lowers the overall acid load and increases the pH of the stomach to have an immediate buffering impact. The side effects of effective antacid therapy include altered bowel functions, constipation (due to aluminum salts), diarrhea (due to magnesium salts), and an altered pH system (due to sodium bicarbonate from high sodium levels). As a result, there is a need for natural plant alternatives with fewer side effects [39].

CONCLUSION

The methanol extract of the three plants, *Momordica charantia*, *Luffa cylindrica*, and *Jatropha curcas*, as well as their crude peptide extracts, were investigated in this study and demonstrated varying degrees of antioxidant and antacid activities. The results support the ethnomedicinal claims that the plants can be used in the management of peptic ulcers. Therefore, the plants could serve as leads in the search for antiulcer agents from natural origin.

List of abbreviations: GnRH: Gonadotropin-releasing hormone, AMPs: Anti-microbial peptides, FHI: Forest Herbarium Ibadan, TPC: Total phenolic content, GAE: Gallic acid equivalent, TFC: Total Flavonoid Contents, DPPH: 2,2-diphenyl-1-picryl-hydrazyl-hydrate, FRSA: Free radical scavenging activity.

Competing interests: The authors declare that there are no competing interests.

Authors' contributions: MAS designed the study, supervised the project, and gave technical input, reviewed, and edited the manuscript. IAA performed part of the experiment, reviewed, and edited the manuscript. JTS performed the experiment and wrote the first draft of the manuscript.

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