

Comparative studies on antifungal, anti-oxidant and phytochemical potential of *Momordica charantia* and *Moringa oleifera*

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Abstract: The Phytochemical, antifungal and anti-oxidant activities of *Moringa oleifera* and *Momordica charantia* leaves were evaluated in this study using the methanol, ethanol and acetone extracts. The phytochemicals identified in of the extract of these plants include; steroids, *tannins*, alkaloids, anthraquinones, flavonoids and terpenoids. The antioxidants levels determined in the extracts of *Momordica charantia* and *Moringa olifera* are; total reducing power, total flavonoid content, total phenol content and DPPH radical scavenging activity. The fungal agents used are; *Aspergillus flavus*, *Aspergillus niger*, *Candida albican* and *Rhizopus*. The anti fungal activities of each of the plant extract were determined using the filter paper disk methods. The extracts possessed an antifungal activity against *Aspergillus flavus*, *Aspergillus niger* and *Candida albican*, while there was no antifungal activity against *Rhizopus sp.* These results were discussed in relation to medicinal potentials of these plants.

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1.0 Introduction

Medicinal plants also known as alternative medicine or unconventional medicine, are therapeutic practises and beliefs outside the realm of mainstream western health care. It emphasizes on therapies that improves quality of life, prevents disease and addresses some disease conditions such as chronic back pain and cancers. Medicinal plants are part and parcel of human society to combat diseases for the dawn of civilization (Ajayi *et al.*, 2008; Amos-Tautua *et al.*, 2011; Morenikeji *et al.*, 2011). *Moringa oleifera* is now the most widely cultivated species of a monogeneric family, the Moringaceae, which has become naturalized in many locations in the tropics (Fashey, 2005). It is a native to the southern foothills of the highlayas in the North-west India and grown mainly in semi-arid, tropical and subtropical areas of North Eastern and South Western Africa particularly in Nigeria (Fahey, 2005). It is a perennial softwood tree which grows best in dry sandy soil with deep roots and timber of low quality, but which for centuries has been advocated for traditional medicinal and industrial uses. All parts of the *Moringa* tree are edible and have long been consumed by humans. *M. Olifera* can easily adapt to varied ecosystems and farming systems and so known for its resistance to drought and diseases (Odee, 1998). The many uses for *Moringa* include; alley cropping (biomass production), animal forage (leaves and treated seed-cake), biogas (from leaves), domestic cleaning agent (crushed leaves), blue dye (wood), fencing (living trees),

fertilizer (seed-cake), foliar nutrient (juice expressed from the leaves), green manure (from leaves), gum (from tree trunks), honey- and sugar cane juice-clarifier (powdered seeds), honey (flower nectar), medicine (all plant parts), ornamental plantings, biopesticide (soil incorporation of leaves to prevent seedling damping off), pulp (wood), rope (bark), tannin for tanning hides (bark and gum), water purification (powdered seeds). *Moringa* seed oil (30-40% yield), also known as Ben oil, is a sweet non-sticking, non-drying oil that resists rancidity (Verma *et al.*, 1976; Fuglie, 2000; Abdulkarim *et al.*, 2005; Joel, 2005; Goel, 2011).

Momordica charantia (Family: Cucurbitaceae), also known as bitter melon plant is widely distributed in many parts of the tropical and subtropical region of the world. It is widely cultivated in Asia, Africa, and Carribean for its edible fruits which is among the most bitter of all fruits (Sofowora, 2006). Nutritionally, the fruit of the plant is widely used and cooked as vegetable. The fruits and leaves of *M. charantia* have medicinal potential in treatment of many diseases such as; piles, leprosy, jaundice, diabetes, snake-bite and have also been found to have vermifuge and antioxidant properties (Arunachalam *et al.*, 2007). It also contains cytotoxic (ribosome-inactivating) proteins such as momorcharin and momordin (Sathish, 2010). In addition to these properties, leaf extracts of bitter melon have clinically demonstrated broad spectrum antimicrobial activity. Various water, ethanol, and methanol extracts of the

leaves have demonstrated in vitro antibacterial activities against *E.coli*, *Staphylococcus*, *Pseudomonas*, *Salmonella*, *Streptobacillus* and *Streptococcus*; an extract of the entire plant was shown to have antiprotozoal activity against *Entamoeba histolytica* (Singh *et al.*, 2006). The fruit and fruit juice has demonstrated the same type of antibacterial properties and, in another study, a fruit extract has demonstrated activity against the stomach ulcer-causing bacteria *Helicobacter pylori*. Phytochemicals are non-nutritive, naturally occurring plant chemical compounds that have protective or disease preventive properties. They are responsible for the colour and organoleptic properties of plants such as deep purple colour of blueberries and the smell of garlic (Brown *et al.*, 2001; Gbolagade and Fasidi, 2005; Jonathan *et al.*, 2011a). They are found in fruits, vegetables, grains and other plants. Some of the commonly known phytochemicals are beta carotene; ascorbic acid, folic acid, and vitamin E. Phytochemicals have been promoted for the prevention and treatment of many health conditions such as cancer, diabetics, heart diseases, and high blood pressure (Craig, 1999; Jonathan and Fasidi, 2003). Antioxidants are the phytochemicals with antioxidant activity which protect the cells against oxidative damages and reduce the risk of developing certain types of cancer. Examples in this group are allyl sulphides (onion, garlic), carotenoids (fruits, carrots), flavonoids and polyphenols. Hormonal actions like isoflavones found in soy, stimulation of enzymes such as indoles, terpenes, and those that interfere with DNA replication such as saponin found in beans interfere with replication of DNA thereby preventing the multiplication of cancer cells (Christopher, 2008).

Antioxidants are our first line of defence against free radical damages and are critical for maintaining optimum health and well being. The need for antioxidants becomes even more critical with increased exposure to free radicals, which are electrically charged molecules with unpaired electrons. Pollution, cigarette smoke, drugs, illness, stress, and even exercise can increase free radical exposure (Mark, 1998; Aina *et al.*, 2012). The widely studied antioxidants include; vitamin A, vitamin E and beta carotene. Vitamin C is considered the most important water-soluble antioxidant in extracellular fluids. It is capable of neutralizing ROS (Reactive Oxygen Species) in the aqueous phase before lipid peroxidation is initiated. Vitamin E, a major lipid-soluble antioxidant, is the most effective chain-breaking antioxidant within the cell membrane where it protects membrane fatty acids from lipid peroxidation. Vitamin C has been cited as being capable of regenerating vitamin E. Beta carotene and

other carotenoids are also believed to provide antioxidant protection to lipid-rich tissues (Mark, 1998).

Antioxidants act as free radical scavengers and hence prevent and repair damage done by these free radicals which can start chain reactions. When the chain reaction occurs in a cell, it can cause damage or death to the cell. Antioxidants terminate these chain reactions by removing free radical intermediates, and inhibit other oxidation reactions (Mark, 1998). Health problems such as heart disease, macular degeneration, diabetes, cancer are caused by oxidative damage. Antioxidants may also enhance immune defences and therefore lower the risk of cancer and infection, coronary heart disease and even altitude sickness (Mark, 1998). In addition to these uses of natural antioxidants in medicine, these compounds have many industrial uses, such as preservatives in food, cosmetics and preventing the degradation of rubber and gasoline. *Aspergillus flavus* is a fungus which is common mold in the environment that causes storage problems in stored grains. It belongs to phylum Ascomycota, Eurotiomycetes class of the order Eurotiales. It can also be a human pathogen, associated with aspergillosis of the lungs. *A. flavus* also produces a toxin, aflatoxin, which is one of the aetiological agents for hepatocellular carcinoma (Klich, 2007). *A. flavus* grows as a yellow-green mold in culture. Like other *Aspergillus* species, it produces a distinctive conidiophore composed of a long stalk supporting an inflated vesicle. Conidiogenous cells on the vesicle produce the conidia. Many strains of *A. flavus* exhibit a greenish fluorescence under UV light that is correlated with levels of aflatoxin production. *Aspergillus niger* is a haploid filamentous fungi commonly found in mesophilic environments such as decaying vegetation, soil, plants and highly essential microorganism producing citric acid as well as extracellular enzymes such as amylases, proteases, and lipases. The use of these enzymes is essential because of its importance for transformation to food enzymes (Jonathan *et al.*, 2011b). *A. niger* is used for waste management and biotransformation (Schuster *et al.*, 2002). This species is also pathogen that cause the spoilage of food and production of secondary metabolites, such as aflatoxin, that are toxic (Schuster *et al.*, 2002; Olawuyi *et al.*, 2010; Jonathan *et al.* 2012). *Candida albicans* is a diploid fungus that grows both as yeast and filamentous cells. It is a causal agent of opportunistic oral and genital infections in humans. Systemic fungal infections (fungemias) including those by *C. albicans* have emerged as important causes of morbidity and mortality in immunocompromised patients (e.g., AIDS, cancer chemotherapy, organ or bone marrow transplantation). *C. albicans* biofilms may form on the surface of

implantable medical devices. In addition, hospital-acquired infections by *C. albicans* have become a cause of major health concerns (Ryan and Ray, 2004).

In recent times, the increasing rate of cancer, diabetics and their causes have become areas of interest. Several naturally occurring plants and foods have been found to provide protection and treatment to cancer, diabetics and bacterial infections (Jonathan *et al.*, 2008; Oluranti *et al.*, 2012). Extracts from plants have been reported to be highly protective against cancer and diabetics and are therefore anticarcinogenic, antidiabetic and antifungal (Sofowora, 2006). This study therefore investigated the antifungal, phytochemical and antioxidant properties of the leaf extracts of *Momordica Charantia* and *Moringa olifera*.

2.0. Materials and methods

2.1. Sources of plant materials

Momordica charantia and *Moringa olifera* leaves used for this study were collected from Oshogbo, Nigeria. They were authenticated using standard Herbarium references. The leaves were air dried at 40°C for 15 days and milled into powder using an electric blender.

2.2. Preparation of plant extract

Methanol, ethanol and acetone were the solvents used for the extraction. 50g of the milled plant was weighed out using a top loading balance into 500ml of the solvents i.e ethanol, methanol and acetone respectively and extracted for 72hrs. The crude extracts were decanted, filtered and concentrated using rotary evaporator until the solvents were recovered completely and dried in a vacuum oven (Jonathan and Awotona, 2010). The residue obtained from the two plant extracts were greenish black solid after which the extract were transferred into a pre-weighed container i.e a vial and stored in a refrigerator at 4°C. Portions were taken from the stored extract prior to experiment.

2.3. Test fungi

The isolates of the test organism were obtained from stored stock culture of *Aspergillus flavus*, *Aspergillus niger*, *Candida albican* and *Rhizopus* species collected from Department of Medical Microbiology, Olabisi Onabanjo University Teaching Hospital using Sabouraud dextrose agar and were incubated at room temperature for 6 days.

2.4. Determination of antifungal activities using filter paper disk method

Whatman filter paper No. 1 was cut into disc using 5.0mm cork borer and sterile blade (Buwa and Staden, 2006). These cut filter papers were sterilised in the oven at 85°C for 60minutes. The dried sterile papers were dipped into 4mls of the various extracts. Sterile Sabouraud dextrose agar was poured into the petri dishes and allowed to set. The plates were seeded

with 24hr old organisms of *Aspergillus flavus*, *Aspergillus niger*, *Candida albican* and *Rhizopus*. The plates were incubated at room temperature for the organisms to grow. The filter papers containing the extracts were placed on the seeded plates. The plates were kept in the refrigerator for proper diffusion of the extracts into the media before incubating at 37°C for 24hrs (Jonathan and Fasidi, 2003). The zones of inhibition were measured.

2.5. Phytochemical evaluation

Phytochemical examinations were carried out for all the extracts according to the standard procedure (Brain and Turner, 1975; Evans 1996; Sofowora, 1993). The extracts were dissolved respectively in distilled water. The filtrates were used for the phytochemical examinations.

2.5.1 Detection of tannins: 1ml of the filtrates was treated with 0.5ml of ferric chloride. A blue-black, green or blue precipitate indicates the presence of tannins.

2.5.2. Detection of alkaloids: 2ml of chloroform and a few drops of Wagner's reagent (iodine to potassium iodide) to 1ml of the filtrate. A reddish brown precipitate indicates the presence of alkaloids.

2.5.3. Detection of phlobatannins: Few drops of 1% of Hydrochloric acid was added to 1ml the filtrate. A reddish brown precipitate indicates the presence of phlobatannins.

2.5.4. Detection of steroids: 0.5ml of acetic anhydride and a few drops of concentrated H₂SO₄ were added to 1ml of filtrate. A bluish-green precipitate indicates the presence of steroids.

2.5.5 Detection of flavonoids: 2ml of NaOH was added to 2ml of the filtrate. The presence of flavonoids is indicated by yellow colour which becomes colourless on addition of dilute acid indicates the presence of flavonoids.

2.5.6. Detection of anthraquinones: 10ml of chloroform was added to 1ml of the filtrate. A greenish yellow precipitate indicates the presence of anthraquinone.

2.5.7. Detection of terpenoids: 5ml of the filtrate was mixed with 2ml of chloroform and carefully added 3ml of concentrated H₂SO₄ to form a layer. A reddish brown colour at the interface indicates the presence of terpenoids.

2.5.8. Detection of saponins: Small amount of extract was shaken with little quantity of water. If foam produced persists for ten minutes, it indicates the presence of saponins.

2.6. Determination of the presence of antioxidant

2.6.1. The total phenolic content

The Total phenolic content (TPC) was determined using the Folin-Ciocalteu colorimeter method according to the procedure of Singleton and Rossi (1965). 1ml of the dissolved extract was mixed

with 0.5ml of Folin-Colcalteu reagent and decolourised with 7.5ml of distilled water. The mixture was kept at room temperature for 5minutes. 10ml of 7% sodium carbonate was added to the mixture and incubated at room temperature for 90minutes. This was done for all samples in triplicate. After incubation, the absorbance against the reagent blank was determined at 760nm. The TPC of the plant was expressed as Gallic acid equivalent (g/100g dry wet) and all the extracts were analysed in triplicates.

2.6.2. Total flavonoid content

The Total flavonoid content (TFC) was analysed following a spectrophotometric method of Dewanto *et al.* (2002). 1ml containing 100 μ g/ml each extracts of the plant material were diluted with 4ml of distilled water in a 10ml volumetric flask. 0.3ml of 5% of NaNO₂ solution was added to each of the volumetric flask. At 5minutes, 0.3ml of 10% of AlCl₃ was added, while at 6minutes, 2ml of 1.0M of NaOH was added. 2.4ml of distilled water was added to the reaction flask and mixed well. Absorbance of the reaction mixture was read at 510nm. TFC were determined as quercetin equivalents (g/100g of dry weight). Three readings were taken for each of the extract and the results were averaged.

The determination of the 2, 2-diphenyl-2-picrylhyrazyl (DPPH) radical scavenging activity was done according to the procedure described by Mensor *et al.*, (2001). Different concentrations of each extract of the plant material were made. The different concentrations used were 2 μ g, 4 μ g and 8 μ g. 2.5ml of each of the extracts were put into different test tubes and 1ml of 0.3Mm DPPH methanol solutions was added to the extract and allowed to react at room temperature for 30 minutes. The absorbance of the resulting mixture was measured at 518nm. The DPPH solution was used as the blank and the absorbance was also measured. The Antioxidant activity (AA %) was determined using the formula:

$$AA\% = \left[\frac{\text{Absorbance Blank} - \text{Absorbance Sample}}{\text{Absorbance Sample}} \right] \times 100$$

2.6.4. Determination of total reducing power

The determination of the reducing power of the plant materials was performed as described by Yen and Duh. The different concentrations (0.1-0.4mg/ml) of the extract were mixed with phosphate buffer (500 μ L 20Mm, Ph 6.6) 500L of 1% potassium ferric cyanide. This was incubated at 50^oC for 20minutes; 0.5ml of 10% trichloroacetic acid were added and the mixture was centrifuged at 2500 revolution per minute for 10minutes. The supernatant was collected and mixed with 1.5ml of distilled water and 300 μ L of 0.1% ferric chloride. The absorbance was read at 700nm.

The experiment was repeated thrice and increase in the absorbance for the reactions mixture indicated increase in the reducing power.

2.7. Statistical analysis

The data were presented as means and standard deviations (SD) from three independent analyzes. Results were evaluated by independent T-Test. The difference was considered significant when P-value was 0.05.

3. Results and Discussion

3.1 Phytochemical evaluation of the extracts of *Momordica charantia* and *Moringa olifera*

Table 1 showed the results of phytochemical evaluation of methanol, ethanol and acetone extracts of *Momordica charantia*. The methanol extract of *Momordica charantia* showed the presence of tannins, alkaloids, steroids, phlobatannins, flavonoids, anthraquinones, terpenoids and saponins. The ethanol extracts also showed the presence of alkaloid, steroids, flavonoid and anthraquinones. In the acetone extract tannins, steroids and saponins were detected. The result of the phytochemical analysis of *Moringa olifera* was presented in Table 2. The methanol and the acetone extracts contained steroids, flavonoid, anthraquinones, terpenoids and saponins with the absence of tannins and alkaloids in the acetone extract but present in the methanol extract of *Moringa olifera*. The ethanol extracts of the two plants showed the presence of the same phytochemicals as contained in the methanol extract.

The phytochemical screening of the methanol, ethanol and acetone extracts of *Momordica charantia* and *Moringa olifera* showed the presence of the following phytochemicals generally; steroids, tannins, anthraquinones, flavonoids and terpenoids. This result is also in line with the report of Bakare *et al.*, (2010) and Kasolo *et al.*, (2011) on the leaves of *Momordica charantia* and *Moringa olifera* respectively. The presence of secondary metabolites such as alkaloids, saponins, tannins, glycosides and cardiac glycosides in the leaf *M. charantia* may contribute to its medicinal value (Bakare *et al.*, 2010). The types of phytochemicals in both plants are not equal indicating variation in the type and quantity of phytochemicals and this was explained by Waller and Nowacki (1978) who suggested that phytochemicals are produced in response to perceived threats by the plants, therefore variation exist in the production of these phytochemicals depending on the type and amount of threat encountered by the plant. Steroids were found to be present in all most extract of both plants. Tannins and saponins were detected in *Moringa* seed ethanol extract in agreement with report by Napoleon *et al.*, (2009). Alkaloids were reported in the present study which was not determined by Napoleon *et al.*, (2009). *Moringa* seed chloroform was shown to

possess only saponins while *Moringa* leaves chloroform extract contained alkaloids, tannins and saponins, which were not reported by Napolean *et al.*, (2009). Farooq *et al.*, (2007) reported that plants occur in varying habitats, a great magnitude of variation in the concentration and composition of phytochemical ingredients in the different parts of such plant is expected. It should be noted that steroidal compound are of importance in pharmacy due to their relationship with such sex hormones. This may be the reason why the leaves of *Momordica charantia* and *Moringa olifera* are used as vegetable for expectant mothers or nursing mother to ensure their hormonal balance since steroidal structure could serve as serve as potent starting material in the synthesis of hormones.

3.3 Antifungal activities of *Momordica charantia* and *Moringa olifera*

Figure 1 showed a graphical representation of the measurement of the zone of inhibition of *Momordica charantia* against the test fungi. The methanol, ethanol and acetone extracts of the plant possessed antifungal activity against *Aspergillus flavus* and *Candida albican*. The methanol extracts produced an inhibitory zone on *Aspergillus flavus* and *Candida albicans* as 50mm and 15mm respectively, that of the ethanol extract on *Aspergillus flavus* and *Candida albican* is 30mm and 20mm respectively. The acetone extract produced the highest zone of inhibition on *Aspergillus flavus* and *Candida albicans* as 40mm and 65mm respectively. The figure 2 shows that the methanol, ethanol and acetone extracts of *Moringa olifera* also possess antifungal activity against all the test fungi with the measurement of the inhibitory zone as 20mm, 15mm, and 30mm respectively. There was no zone of inhibition on the *Rhizopus* sp from any of the plant extracts and this implies that the plants do not affect this particular fungus. This study showed that the acetone extract of *Momordica charantia* had the highest zone of inhibition on *Candida albicans*, while the methanol extract of *Moringa olifera* possessed the lowest zone of inhibition on the fungi. These fungi are known to cause infections in human except for *Rhizopus*. The zones of inhibition observed indicated that they are susceptible to the extract, and these extracts could be used in the treatment of urinary tract infections associated with *Aspergillus flavus* and *Candida albican*.

3.2 Anti-oxidant activity of *Momordica charantia* and *Moringa olifera*

Results presented on Table 3 showed that the mean and standard deviation values of the total phenol content of the methanol, ethanol and acetone extracts of *Momordica charantia*. The extract of acetone was the highest with 0.214 ± 0.014 , followed by methanol

0.193 ± 0.049 , while ethanol had the least value of 0.151 ± 0.016 . On the other hand, the result on Table 4 for *Moringa olifera* showed that the total phenol content of the methanolic extract was the highest (0.255 ± 0.017), followed by ethanolic extract (0.203 ± 0.008), while the extract of acetone had the least value of 0.198 ± 0.041 .

Table 5 showed comparison between the total phenol content of the methanol, ethanol and acetone extract of *Momordica charantia* and *Moringa olifera* using the T-test analysis. Only the ethanol extract of *Momordica charantia* is significant as its p value is 0.042 which is less than 0.05. Table 6 and 7 shows the values when each extract of each plant were compared with the control using the T-test analysis. It was observed that there was no significant difference between any of the plant extract and control.

The total flavonoid content of the methanol, ethanol and acetone extracts of *Momordica charantia* are; 0.801 ± 0.833 , 0.150 ± 0.087 , 0.186 ± 0.534 respectively (Table 8), while the result of table 9 shows values of the total flavonoid content of the methanol, ethanol, and acetone extracts of *Moringa olifera* as 0.125 ± 0.007 , 0.124 ± 0.030 , 0.104 ± 0.001 respectively. On Table 10, there was no significant difference in the total flavonoid content of methanol, ethanol and acetone extracts for both plants compared with the control. Also, there were no significant difference between each of the extracts and control in tables 11 and 12. Results of tables 13 and 14 shows the DPPH radical scavenging activity of both plants. There were no significant differences between the P value and T-test of DPPH for methanol, ethanol and acetone activity of *Momordica charantia* and *Moringa olifera* (Table 15), while significant differences in the activity of the methanol, ethanol and acetone extracts of *Momordica charantia* were observed compared with the control. Similar result was also observed when each of the extracts of *Moringa olifera* was compared with the control in table 17. The results of the total reducing power of the extracts of both plants was presented in tables 18 and 19, while table 20 compared the extracts of both plants. The result shows that only 0.1mg/ml and 0.4mg/ml concentrations showed a significant difference for the methanol extracts of both plants. For the ethanolic extracts of both plants, 0.3mg/ml and 0.4mg/ml showed significant difference, while only 0.4mg/ml concentration produced significant difference for acetone extracts. In table 21, the methanolic extract of *Momordica charantia* showed significant difference compared with the control at 0.1mg/ml, 0.3mg/ml, 0.4mg/ml concentrations whereas, all the concentrations of ethanol extract and control showed significant difference. while when compared between acetone and the control, only 0.1mg/ml concentration

had a significant difference. Result from table 22 showed that the methanol and acetone extracts of *Moringa olifera* showed significance difference compared with control for all the concentrations, while the ethanol extract showed significant difference compared with the control at 0.2mg/ml and 0.3mg/ml concentrations.

Free radicals are known to be major cause of various chronic and degenerative diseases including aging, coronary heart disease, inflammation, stroke, diabetes mellitus, and other cancer as reported by Cheng *et al.*, (2003) and Slater (1984). Free radicals generated as a result of metabolic activities are involved in the ageing processes of tissues and pathologies such as cancer or cardiovascular disease (Namiki, 1990). Antioxidants are substances which can either directly scavenge reactive oxygen species (ROS) or prevent the generation of ROS (Habtemariam, 2006). Perry *et al.*, (1999) also reported that the moringa leaf was the main source of nutrition as it contains numerous nutritional antioxidant compounds in substantial amount, such as ascorbic acid, carotenoids, α -tocopherol, and phenols. Due to high concentration of these beneficial compounds, they are regarded as the best, nutritionally rich plant that can be consumed for a long time over a period of growth and development where food crops are scarce. Leaves can be further processed into powder final products and used as one of the ingredient in improving traditional foods or to develop new innovative food products by considering socio-cultural factors in the regions. Leaves can also be eaten fresh, cooked, or stored as freeze-dried powder for many months without refrigeration, and reportedly without significant loss of nutritional value. The consumption of moringa leaf powder is also crucial in situations where starvation is imminent Mahmood *et al.*, (2010). It has been reported that the antioxidants activity of plant material was well correlated with the phenol compounds (Veliogu *et al.*, 1998). The total phenol content obtained from all the extracts of *Momordica charantia* and *Moringa olifera* ranges between 0.151 to 0.255mg/ml of extract. The highest total phenol was recorded for the methanol extract of *Moringa olifera* as 0.255mg/ml contrary to what was reported by Shu-Jing and Lean-teik (2007) reported the total phenol content of wild *Momordica charantia* as 68.8mg/ml and this concentration level is higher than level of the total phenol content found in this study and this may be as a result of the wild nature of the bitter melon. The results of total flavonoid content of the methanol, ethanol and acetone extract of *Momordica charantia* were 0.801,0.150, and 0.186mg/ml which appears lower than the total flavonoid content in wild bitter melon. When the total flavonoid and phenol contents are compared it can be

observed that the flavonoid content is higher than the phenol content of the *Momordica charantia* and this is in correlation with the finding of Shu-Jing and Lean-teik (2007) on wild bitter melon. Plant tissues contain a wide variety of compounds with antioxidant activity. Phenol compounds (flavonoids and phenolic acids), nitrogen compounds (alkaloids, chlorophyll derivatives, amino acids and amines), carotenoids, lignans and terpenes were reported to possess antioxidative activity in suppressing the initiation or propagation of the chain reactions (Hall and Cuppett, 1997). Flavonoids and phenol compounds are the main antioxidative compounds of fruits and vegetables as earlier observed by Huang *et al.*, (1998). The DPPH result of the methanol, ethanol and acetone extracts of *Momordica charantia* and *Moringa olifera* did not result in the change of colour which indicates the effect of the plant extracts in reducing the DPPH to the stable radical yellow colour diphenylpicrylhydrazine, indicating that these extracts are active in DPPH radical scavenging as reported by Shu-Jing and Lean-teik (2007) who worked on wild *Momordica charantia*. The DPPH radical expressed by the methanol, ethanol and acetone extracts of *Momordica charantia* are 164.250 μ g/ml, 166.850 μ g/ml and 156.850 μ g/ml respectively while those of *Moringa olifera* showed 168.450 μ g/ml, 167.250 μ g/ml and 168.300 μ g/ml. The DPPH radical scavenging activity of the extracts of *Moringa olifera* is higher than that of the *Momordica charantia* as seen in this study. Also these plant extracts showed a stronger scavenging activity than Ascorbic acid which is the control. The scavenging action of plant constituents has been found to relate to polyphenolic compounds (Hatono *et al.*, 1989); (Kimura *et al.*, 1985). In this study, *Momordica charantia* and *Moringa olifera* showed antioxidant activity however, the magnitude of antioxidative potency varied with the type of extracts. This could be due to the difference in concentrations and type of antioxidative compounds present in these extracts.

Table 1. Phytochemical evaluation of *Momordica charantia*

PARAMETER	METHANOL	ETHANOL	ACETONE
Tannins	+	-	-
Alkaloids	+	+	+
Steroids	-	+	+
Phlobatannins	-	-	-
Flavonoids	+	+	-
Anthraquinones	+	+	-
Terpenoids	-	-	-
Saponins	+	-	+

Table 2. Phytochemical evaluation of *Moringa olifera*

PARAMETERS	METHANOL	ETHANOL	ACETONE
Tannins	+	+	-
Alkaloids	+	+	-
Steroids	+	+	+
Phlobatannins	-	-	-
Flavonoids	+	+	+
Anthraquinones	+	+	+
Terpenoids	+	+	+
Saponins	+	+	+

Table 3. The total phenol content of the methanol, ethanol and acetone extracts of *Momordica charantia*.

Plant extract	Mean±standard deviation
Methanol	0.193± 0.049
Ethanol	0.151± 0.016
Acetone	0.214± 0.014
Control	0.283± 0.134

Table 4. The total phenol content of methanol, ethanol and acetone extracts of *Moringa olifera*

Plant extract	Mean±standard deviation
Methanol	0.255± 0.017
Ethanol	0.203± 0.008
Acetone	0.198± 0.041
Control	0.283± 0.134

Table 5: The total phenol content of methanol, acetone and ethanol extracts of *Momordica charantia* and *Moringa olifera* when compared using t-test.

<i>Momordica charantia</i> (T-Test, P-value)	<i>Moringa olifera</i> (T-Test, P-value)
Methanol extract (2.059, 0.109 ^{Ns})	Methanol extract (2.059, 0.150 ^{Ns})
Ethanol extract (2.956, 0.042*)	Ethanol extract (2.956, 0.053 ^{Ns})
Acetone extract (0.626, 0.565 ^{Ns})	Acetone extract (0.626, 0.584 ^{Ns})

KEY: P < 0.05 is significant (*)

Ns = Not significant.

Table 6. The total phenol content of methanol, ethanol and acetone extracts in *Momordica charantia* and control (vitamin C) are compared using t-test.

Extract and Control	T-Test	P-Value
Methanol extract & Control	1.097	0.334 ^{Ns}
Ethanol extract & Control	1.701	0.228 ^{Ns}
Acetone extract & Control	0.901	0.461 ^{Ns}

KEY:

P < 0.05 is significant

Ns = Not significant.

Table 7. T-Test and P-Value analysis of total phenol content of methanol, ethanol and acetone extracts in *Moringa olifera* with control.

Extract and Control	T-Test	P-Value
Methanol extract & Control	0.364	0.735 ^{Ns}
Ethanol extract & Control	1.029	0.405 ^{Ns}
Acetone extract & Control	1.060	0.385 ^{Ns}

KEY: P < 0.05 is significant, Ns = Not significant.

Table 8. The total flavonoid content in methanol, ethanol and acetone extracts of *Momordica charantia*.

Plant extract	Mean, Standard deviation
Methanol extract	0.801± 0.633
Ethanol extract	0.150± 0.487
Acetone extract	0.186± 0.534
Control	0.554± 0.596

Table 9. The total flavonoid content of the methanol, ethanol and acetone in *Moringa olifera*

Plant extract	Mean, Standard deviation
Methanol extract	0.125± 0.007
Ethanol extract	0.124± 0.030
Acetone extract	0.104± 0.001

Table 10. T-Test and P-Value analysis of the total flavonoid content of *Momordica charantia* plant extracts and *Moringa olifera* plant extracts.

Plant extracts	T-Test	P-Value
A	1.848	0.206 ^{Ns}
B	0.786	0.484 ^{Ns}
C	2.656	0.117 ^{Ns}

A= Methanol extracts of *Momordica charantia* and *Moringa olifera* B= Ethanol extracts of *Momordica charantia* and *Moringa olifera*, C= Acetone extracts of *Momordica charantia* and *Moringa olifera*. P < 0.05 is significant, Ns = Not significant.Table 11. T-Test and P-Value of the total flavonoid content of the methanol, ethanol and acetone extracts in *Momordica charantia* with controls.

Extract and Control	T-Test	P-Value
Methanol extract & Control	0.671	0.562 ^{Ns}
Ethanol extract & Control	3.365	0.067 ^{Ns}
Acetone extract & Control	3.045	0.078 ^{Ns}

KEY: P < 0.05 is significant, Ns = Not significant.

Table 12 T-Test and P-value of the Total flavonoid content of the methanol, ethanol and acetone extracts in *Moringa olifera* with control.

Extract and Control	T-Test	P-Value
Methanol extract & Control	3.500	0.068 ^{Ns}
Ethanol extract & Control	3.652	0.063 ^{Ns}
Acetone extract & Control	3.880	0.060 ^{Ns}

P < 0.05 is significant, Ns = Not significant.

Table 13. Mean and standard deviation values of the DPPH free radical scavenging activity of the methanol, ethanol and acetone extracts of *Momordica charantia*.

Plant extract	Mean, Standard deviation
Methanol extract	164.250± 4.171
Ethanol extract	166.850± 3.741
Acetone extract	156.850± 3.040

Table 14. Mean and standard deviation values of the DPPH free radical scavenging activity of the methanol, ethanol and acetone extracts of *Moringa olifera*.

Plant extract	Mean, Standard deviation
Methanol extract	168.450± 12.791
Ethanol extract	167.250± 0.150
Acetone extract	168.300± 0.424
Control	0.397± 0.656

Table 15 T-Test and P-value analysis of DPPH radical scavenging activity of the methanol, ethanol and acetone extracts of *Momordica charantia* with *Moringa olifera*.

Plant extracts	T-Test	P-Value
A	0.441	0.725 ^{Ns}
B	0.151	0.894 ^{Ns}
C	0.274	0.534 ^{Ns}

Table 18. The total reducing power of the methanol, ethanol and acetone extracts at various concentrations of the *Momordica charantia*

Concentration (mg/ml)	Methanol (mean±s.d)	Ethanol (mean±s.d)	Acetone (mean±s.d)	Control (mean±s.d)
0.1mg/ml	0.257± 0.031	0.468± 0.203	1.076± 0.417	0.561± 0.054
0.2mg/ml	0.252± 0.139	0.302± 0.353	0.431± 0.044	0.543± 0.010
0.3mg/ml	0.288± 0.028	0.305± 0.014	0.427± 0.021	0.568± 0.047
0.4mg/ml	0.362± 0.027	0.124± 0.025	0.506± 0.011	0.554± 0.047

mean±s.d: Mean and Standard deviation

Table 19: The total reducing power of the methanol, ethanol and acetone extracts at various concentrations of the *Moringa olifera*.

Concentration (mg/ml)	Methanol (mean±s.d)	Ethanol (mean±s.d)	Acetone (mean±s.d)	Control (mean±s.d)
0.1mg/ml	0.392± 0.046	0.314± 0.032	0.328± 0.008	0.561± 0.054
0.2mg/ml	0.292± 0.046	0.302± 0.047	0.398± 0.047	0.543± 0.543
0.3mg/ml	0.268± 0.010	0.416± 0.046	0.396± 0.002	0.568± 0.047
0.4mg/ml	0.294± 0.010	0.469± 0.015	0.334± 0.017	0.554± 0.047

mean±s.d: Mean and Standard deviation

A= Methanol extracts of *Momordica charantia* and *Moringa olifera*, B= Ethanol extracts of *Momordica charantia* and *Moringa olifera*, C= Acetone extracts of *Momordica charantia* and *Moringa olifera*. P < 0.05 is significant, Ns = Not significant

Table 16. T-Test and P-value of the DPPH free radical scavenging activity of the methanol, ethanol and acetone extracts of *Momordica charantia* with control.

Extract and Control	T-Test	P-Value
Methanol extract & Control	74.501	0.011*
Ethanol extract & Control	87.600	0.010*
Acetone extract & Control	72.758	0.009*

P < 0.05 is significant, Ns = Not significant.

Table 17 T-Test and P-value of the DPPH free radical scavenging activity of the methanol, ethanol and acetone extracts of *Moringa olifera* with control.

Extract and Control	T-Test	P-Value
Methanol extract & Control	24.913	0.034*
Ethanol extract & Control	108.260	0.001*
Acetone extract & Control	553.237	0.001*

P < 0.05 is significant, Ns = Not significant.

Table 20. T-Test and P-value of total reducing power in methanol, ethanol and acetone extracts of *Momordica charantia* and *Moringa olifera*.

Plant extracts	Concentrations			
	0.1mg/ml (T-test, P-value)	0.2mg/ml (T-test, P-value)	0.3mg/ml (T-test, P-value)	0.4mg/ml (T-test, P-value)
A	4.163, 0.019*	0.471, 0.677 ^{ns}	1.137, 0.352 ^{ns}	4.102, 0.036*
B	1.291, 0.320 ^{ns}	1.611, 0.196 ^{ns}	3.397, 0.042*	38.519, 0.000*
C	3.101, 0.090 ^{ns}	0.897, 0.421 ^{ns}	2.527, 0.125 ^{ns}	14.317, 0.000*

A= Methanol extracts of *Momordica charantia* and *Moringa olifera*, B= Ethanol extracts of *Momordica charantia* and *Moringa olifera*, C= Acetone extracts of *Momordica charantia* and *Moringa olifera*. P < 0.05 is significant, Ns = Not significant.

Table 21. T-test and P-value result of the various concentrations of methanol, ethanol and acetone extracts of *Momordica charantia* with the control (Vitamin C).

Extract& Control	0.1mg/ml (T-test, P-value)	0.2mg/ml (T-test, P-value)	0.3mg/ml (T-test, P-value)	0.4mg/ml (T-test, P-value)
Methanol & control	8.205, 0.002*	3.608, 0.068 ^{Ns}	8.797, 0.002*	6.100, 0.007*
Ethanol & control	4.106, 0.015*	9.078, 0.009*	9.099, 0.005*	9.313, 0.009*
Acetone & control	0.772, 0.512 ^{Ns}	13.412, 0.002*	3.978, 0.016*	15.690, 0.004*

P-Value < 0.05 is significant, Ns = Not significant.

Table 22. T-test and P-value result of the various concentrations of methanol, ethanol and acetone extracts of *Moringa olifera* with the control (Vitamin C).

Extract& Control	0.1mg/ml (T-test, P-value)	0.2mg/ml (T-test, P-value)	0.3mg/ml (T-test, P-value)	0.4mg/ml (T-test, P-value)
Methanol & control	6.804, 0.005*	6.828, 0.017*	4.602, 0.022*	2.965, 0.078*
Ethanol & control	2.116, 0.164 ^{Ns}	4.292, 0.042*	4.122, 0.020*	1.730, 0.213 ^{Ns}
Acetone & control	7.395, 0.016*	5.230, 0.029*	0.230, 0.007*	7.553, 0.008*

P-Value < 0.05 is significant, Ns = Not significant.

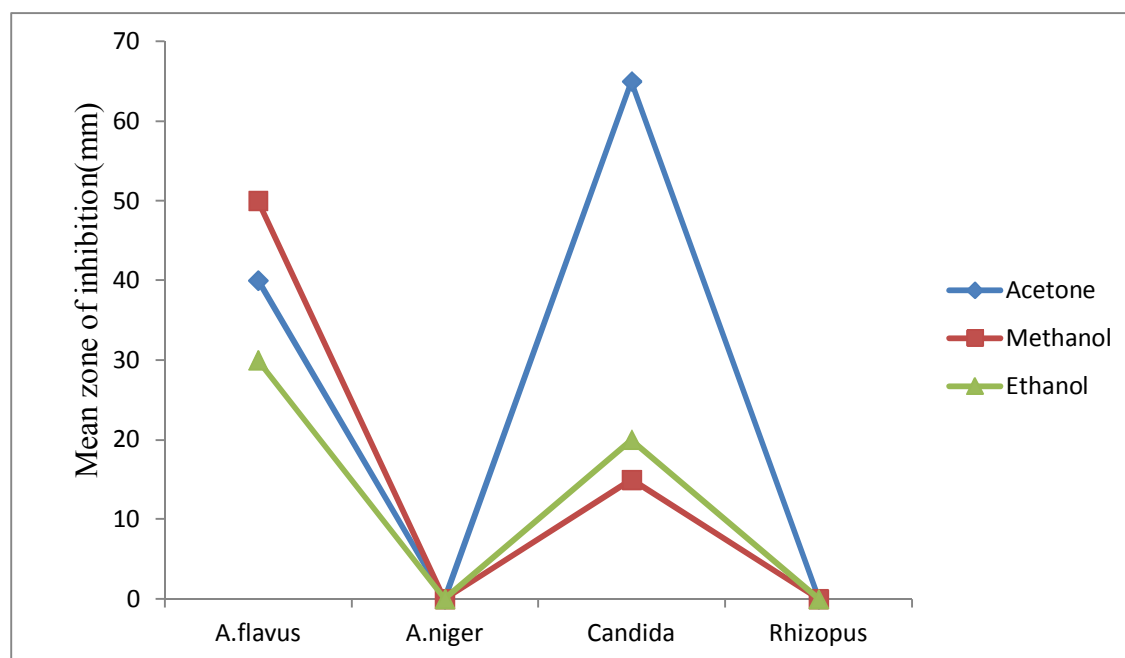


Figure 1. Mean zone of inhibition of *Momordica charantia* on the test fungi.

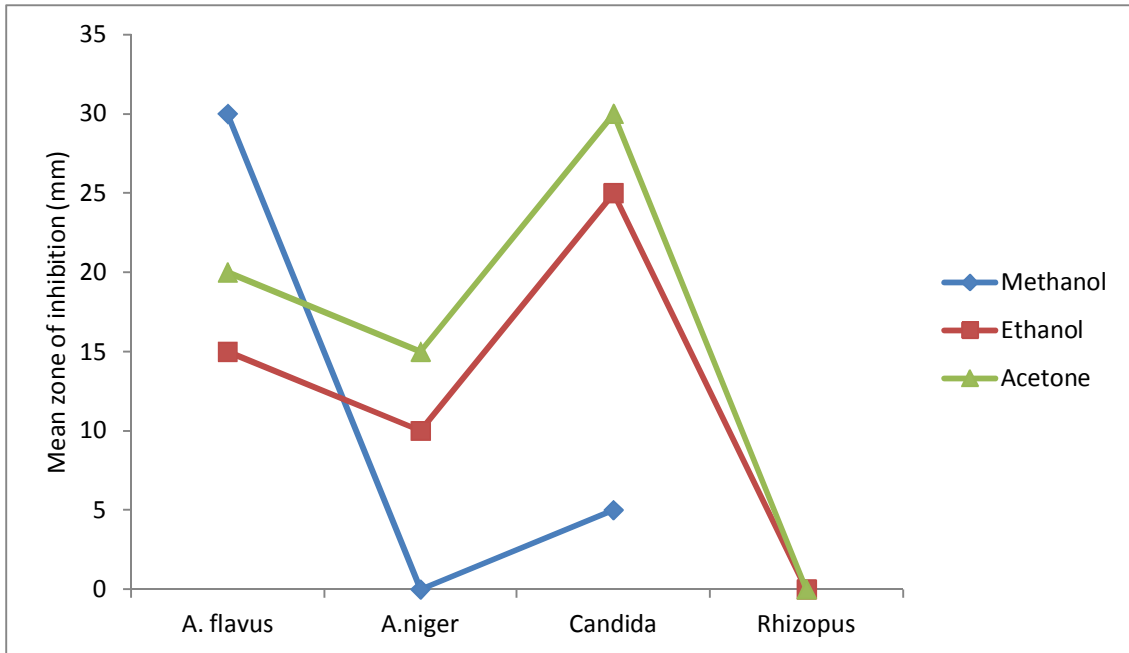


Figure 2. Mean zone of inhibition of *Moringa olifera* on the test fungi.

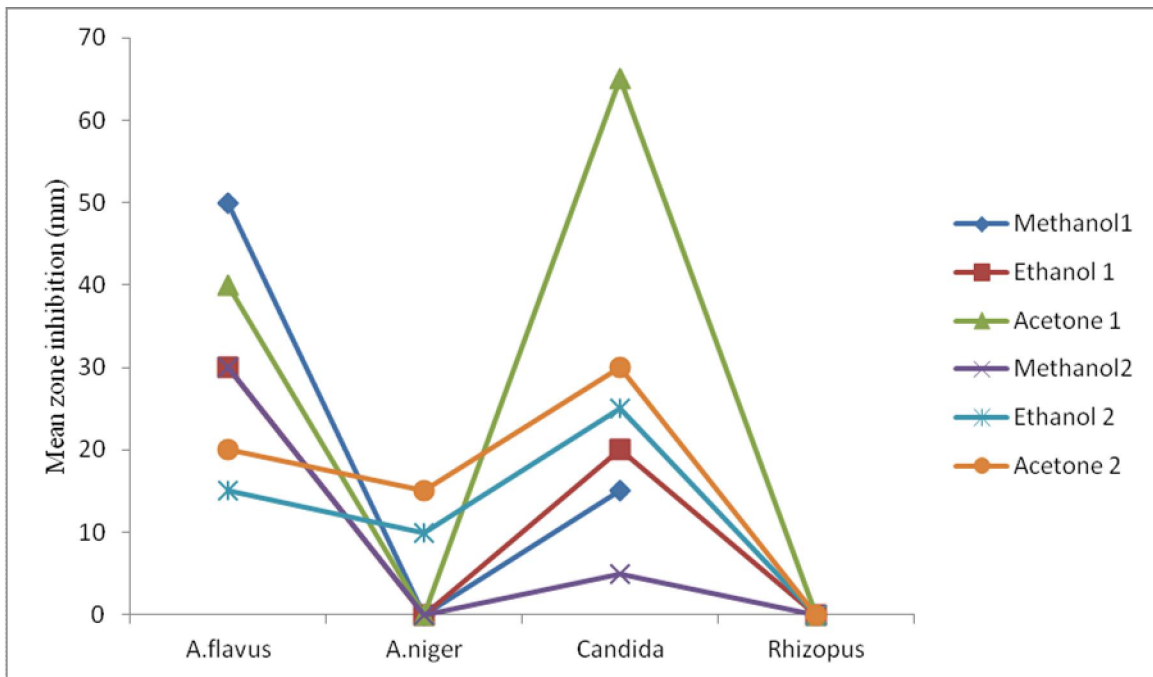


Figure 3. Mean zone of inhibition of *Momordica charantia* and *Moringa olifera* on the test fungi.

Methanol 1 = Methanol extract of *Momordica charantia*, Ethanol 1 = Ethanol extract of *Momordica charantia*, Acetone 1 = Acetone extract of *Momordica charantia*, Methanol 2 = Methanol extract of *Moringa olifera*, Ethanol 2 = Ethanol extract of *Moringa olifera*, Acetone 2 = Acetone extract of *Moringa olifera*

Conclusion.

It was obvious from these studies that all extracts of *Momordica charantia* and *Moringa oleifera* possessed varying degree of antifungal and antioxidant potentials. It was observed that among the two plants, *Moringa oleifera* was better than *Momordica charantia*

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