



Comparison of the phytochemicals and antimicrobial activity of leaf extracts of *Calotropis procera*, *Momordica charantia* and *Allium ascalonicum*

Jesumirhewe Christiana^{1*}, Utomi Ozioma Lilian² and Ogunlowo Oladejo Peter³

¹Department of Pharmaceutical Microbiology, College of Pharmacy, Igbinedion University Okada Edo State, Nigeria. E-mail: ebarunosen2002@yahoo.co.uk

²Department of Pharmaceutical Microbiology, College of Pharmacy, Igbinedion University Okada Edo State, Nigeria. E-mail: liiyutomi@gmail.com

³Department of Molecular Sciences, Macquarie University, Australia. E-mail: ogunlowooladejo@yahoo.co.uk

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Abstract

The emergence of new infectious diseases, resurgence of several infections that appeared to have been controlled and the increase in bacterial resistance has led to the need for studies directed towards the development of new antimicrobials. Considering the challenge of obtaining new molecules with antimicrobial properties from microorganisms, identifying antimicrobials from other natural sources is of great importance. The phytochemical and antimicrobial activity of ethanolic leaf extracts of *Calotropis procera*, *Momordica charantia* and *Allium ascalonicum* was investigated in an attempt to evaluate their medicinal potential in the treatment of gastroenteritis, a major cause of death especially in children under the age of five in Nigeria. The phytochemical screening revealed the presence of the active medicinal constituents analyzed. The extracts had varying degree of inhibitory effects on the isolates tested. *Allium ascalonicum* extract had the highest antimicrobial activity compared to *Calotropis procera* and *Momordica charantia* extracts. Gentamicin also had good inhibitory effects on the isolates used. Our study showed that the selected plant extracts had good antimicrobial effects on the pathogenic bacteria causing gastroenteritis with *Allium ascalonicum* extract showing the highest activity. These plants can be useful in treating infections caused by these bacteria and suggests further advance investigations.

Keywords: Medicinal plants, Phytochemical screening, Antibacterial activity, Antimicrobials.

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1. Introduction

Herbs and/or plants as medicine have been used by man for therapeutic purpose (Ezekwesili-Ofili and Okaka, 2019). Different plant parts like root, stem, bark, heartwood, leaf, flower, fruits, seeds or plant exudates have medicinal properties (Essiett and Basse, 2013). It is generally estimated that over 6,000 plants in Africa are in use in traditional folk and herbal medicine which represents about 75% of the medicinal needs of the third world countries (Veerachari and Bopaiah, 2011). The promising potentials of medicinal plants used in various traditional, complementary and alternate systems of treatments of human diseases have been

* Corresponding author: Jesumirhewe Christiana, Department of Pharmaceutical Microbiology, College of Pharmacy, Igbinedion University Okada Edo state, Nigeria. E-mail: ebarunosen2002@yahoo.co.uk

demonstrated (Alam, 2009). There are a number of compounds that are possibly natural antimicrobial agents in medicinal plants. These possible antimicrobial agents are safe, cheaper and potent and also play a vital role in the development of novel drugs useful in the treatment of microbial infections (Gilani and Rahman, 2005). Plants like *Calotropis procera*, *Allium ascalonicum* and *Momordica charantia* are examples of medicinal plants used in Nigeria. *Calotropis procera* is of the family Asclepiadaceae (Apocynaceae). The numerous therapeutic effects of *Calotropis procera* are indicated in several reports in the literature. Previous reports show that the whole plant of *Calotropis procera* could be used alone/with other herbs in the treatment of common diseases which include rheumatism, cold and eczema. It can also be used as a purgative and taken in cases of indigestion and diarrhea (Ul-Zaman and Ahmad, 2017; and Asoso et al., 2018). *Momordica charantia* belongs to the family Cucurbitaceae. Previous reports show that among its many uses it is useful in various systems as antidiabetic, antimicrobial, anticarcinogenic, laxatives etc. (Ingle and Kapgate, 2018; Adeyemi and Oluwasegun, 2019; and Saravani et al., 2019). It is also indicated in treating skin diseases. *Allium ascalonicum* belong to the family Alliaceae and it has been reported to be useful as an antioxidant, antihelminthic and antimicrobial (Adeniyi and Anyiam, 2004; Kyung, 2012; and Mnayer et al., 2014). The therapeutic uses of available medicinal plants have been useful in treating various ailments as a result of the reliability and stability in such products for therapeutic purposes (Akharaiyi and Boboye, 2015). This work was carried out to compare the phytochemical constituents and antimicrobial activity of leaf extracts of *Calotropis procera*, *Momordica charantia* and *Allium ascalonicum* used as medicinal plants in the treatment of gastroenteritis in Nigeria.

2. Materials and methods

2.1. Plant collection and authentication

The fresh leaves of *Calotropis procera* and *Momordica charantia* were collected from the old boys hostel, Crown Estate, Igbinedion University Okada (IUO) and the Pharmacognosy Departmental garden College of Pharmacy Igbinedion University Okada Edo state respectively. *Allium ascalonicum* was collected from Forestry Research Institute of Nigeria (FRIN) Botanical Garden, Ibadan, Oyo state, Nigeria. The plants were identified in the College of Pharmacy, IUO and authenticated at FRIN in Ibadan, Nigeria. The leaves of each plant were sorted and thoroughly washed with clean water and room dried. After drying they were pulverized and subjected to organic solvent extraction.

2.2. Organic solvent extraction

The powdered *Calotropis procera* was placed inside a thimble of a soxhlet apparatus and about 500 ml of extracting solvent (ethanol) was added in a distillation flask and the soxhlet extractor placed on the flask. The solvent was heated and vapor produced was cooled by the condenser which dripped back into the chamber housing the powdered plant thus dissolving it. The soxhlet chamber was emptied by a siphon side arm when almost full with the solvent running down the distillation flask. The cycle was repeated many times for three days. Extracts were finally concentrated in the distillation flask. The same procedure was repeated for *Momordica charantia* and *Allium ascalonicum*. Ethanol was removed by using an evaporator. Weight of total pulverized sample of *Calotropis procera*, *Momordica charantia*, *Allium ascalonicum* was 350 g, 300 g and 180 g respectively. Percentage yield of extracts of *Calotropis procera*, *Momordica charantia*, and *Allium ascalonicum* samples were 18.07%, 19.11% and 19.68% respectively.

2.3. Microorganism

Five isolates of *Escherichia coli* obtained from faecal sources were collected from University of Benin Teaching Hospital, Benin City (UBTH) and Igbinedion University Teaching Hospital Okada (IUTH), for the experiment.

2.4. Phytochemical screening

The methods described by Harborne (1973) and Trease and Evans (1989) was used.

2.5. Antibacterial screening of the crude extracts

The antibacterial activity of the leaf extracts was evaluated by agar well diffusion (Asoso et al., 2018). Wells were made (8 mm diameter) on each culture plate of the organisms with the aid of a sterile cork borer. The three extracts were reconstituted to 800 mg/ml with Dimethyl sulfide (DMSO₂). Aseptically, each well was filled up with 0.5 ml of each extract using a Pasteur pipette. Gentamicin 30 µg/ml and Dimethyl sulfide was used as positive and negative control respectively. The plates were incubated at 37 °C for 24-48 h. The zones of inhibition were measured and recorded in millimeters.

3. Results

Calotropis procera, *Momordica charantia*, *Allium ascalonicum* were authenticated as *Calotropis procera* (Ait) Ait.f. (Family: *Asclepiadaceae*), *Momordica charantia* linn (Family- *Cucurbitaceae*) and *Allium ascalonicum* Hort. (Family- *Liliaceae*) respectively. The voucher specimen of the plants had reference No. FHI. 109879, 109880 and 109881 respectively and has been kept in the FRIN herbarium for further reference. Flavonoids, saponins glycosides, alkaloids were found in the ethanolic extracts of *Momordica charantia* and *Allium ascalonicum* leaves. Phenol was also detected in the ethanolic extract of *Momordica charantia* leaves. Saponins and alkaloids were the only constituent detected in the ethanolic extract of *Calotropis procera* leaves (Table 1).

Table 1: Phytochemical screening of *Calotropis procera*, *Momordica chaxrantia*, *Allium ascalonicum*

Phytochemical constituents	<i>Calotropis procera</i>	<i>Allium ascalonicum</i>	<i>Momordica charantia</i>
Tannins	-	-	-
Flavonoids	-	+	+
Saponins	+	+	+
Glycosides	-	+	+
Alkaloids	+	+	+
Phenols	-	-	+

Note: - Negative and + Positive.

The results of the antimicrobial activity of the ethanolic extracts of *Calotropis procera*, *Momordica charantia*, and *Allium ascalonicum* leaves on the *E. coli* isolates showed *Allium ascalonicum* extract had the highest antimicrobial activity, followed by *Calotropis procera* while *Momordica charantia* extracts exhibited the lowest antimicrobial activity. Gentamicin the positive control also had activity on the bacterial isolates (Figure 1).

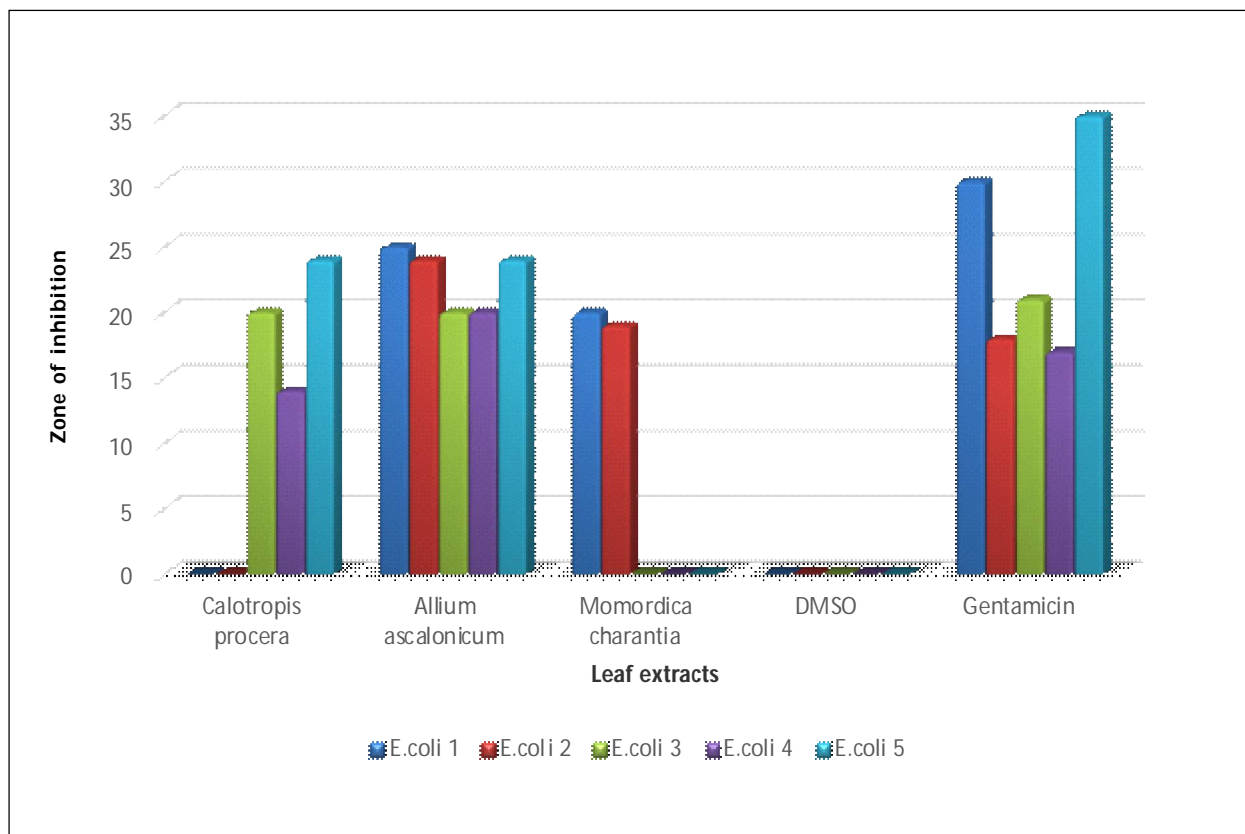


Figure 1: Antimicrobial activity of the extracts

4. Discussion

There has been an increasing interest in alternative therapy globally (Frass et al., 2012; and Welz et al., 2018) and a consistent increase in the use of herbs derived from plants as alternatives or rather complementary to the use of orthodox or synthetic drugs. The awareness of having beneficial effects in the use of nature's products, the side effects of conventional drugs along with the risk involved in the consumption of substandard and fake drugs especially in developing countries has contributed to the increasing demand of alternative therapy (Ezekwesili-Ofili and Okaka, 2019). The phytochemical screening showed that the plant extracts had chemical constituents of pharmacological importance. The phytochemical screening revealed alkaloids and saponins in *Calotropis procera* leaf extracts. Saponins have been reported to be important therapeutically as they are shown to have hypolipidemic and anticancer activity (Akindele et al., 2017). They are also necessary for the activity of cardiac glycosides (Sarker and Nahar, 2007). Plants rich in saponins are also reported to have immune boosting and inflammatory properties (Francis et al., 2002). The antimicrobial activity of alkaloids has been reported by a number of authors (Djeussi et al., 2013; Manandhar et al., 2019; and Okeke and Ezeabara, 2018). Previous reports reveal that phytochemicals show antimicrobial activity against a number of microorganisms (Ingle and Kapgatte, 2018; Asoso et al., 2018; and Akindele et al., 2017). Powdered leaves of *Calotropis procera* have been reported to be useful for the fast healing of wounds as a purgative and to treat indigestion. They are also useful in treating skin disorders and liver problems. Dried leaves of *Calotropis procera* are also useful in promoting sexual health (Tiwari et al., 2014; and Panda et al., 2015). Tannins and phenols were absent in the *Allium ascalonicum* extract while tannin was the only constituent absent in the *Momordica charantia* extract out of the constituents screened in the phytochemical screening. The chemical constituents have one or more therapeutic importance. The antimicrobial screening results of *Calotropis procera*, *Momordica charantia*, and *Allium ascalonicum* against microorganisms causing gastroenteritis especially *E. coli* showed that the *Allium ascalonicum* extract at a concentration 800 mg/ml had the highest antimicrobial activity compared to the other plant extracts. This is in correlation of an earlier report suggesting the therapeutic potential of *Allium ascalonicum* against *Helicobacter pylori* infection which could be explored for patients with gastroduodenal disorders. There are a lot of previous reports showing the antimicrobial efficacy of *Calotropis procera* and *Momordica charantia* leaf extracts with only very few reports on the antimicrobial effect of *Allium ascalonicum* leaf extract. A number of *Allium* species are noted for their antimicrobial activity. They include garlic (*A. sativum* L.) (Kang et al., 2010), onion (*A. cepa* L.) (Mohamed, 2010), shallot (*A. ascalonicum* L.) (Rattanachaikunsopon and Phumkhachorn, 2009a), elephant garlic (*A. ampeloprasum* L. var. *ampeloprasum* auct.) (Rattanachaikunsopon and Phumkhachorn, 2009b), and wild garlic (ramsons; *A. ursinum*) (Ivanova et al., 2009). The antimicrobial activities of allium extracts of bulbs (Mohamed, 2010; and Mahmoudabadi and Nasery, 2009) and of essential oils (Rattanachaikunsopon and Phumkhachorn, 2009a; and Rattanachaikunsopon and Phumkhachorn, 2009b) have been reported. *Allium* leaves and flowers have also been reported for their antimicrobial activities (Ivanova et al., 2009). The antimicrobial activity of *Calotropis procera* and *Momordica charantia* leaf extracts previously demonstrated in past reports correlates with results from this present study (Kyung, 2012; Akindele et al., 2017; and Saravani et al., 2019).

5. Conclusion

Our results confirm earlier reports by various researchers on the potential uses of plants in traditional medicine which include *Calotropis procera*, *Momordica charantia* and *Allium ascalonicum* (Kyung, 2012; Akindele et al., 2017; and Saravani et al., 2019). The significant inhibitory potentials displayed by ethanolic leaf extract of *Calotropis procera*, *Momordica charantia* and *Allium ascalonicum* against susceptible strains of *E. coli* establish the scientific rationale for the use of these plants in folk medicine. Phytochemicals detected in the plants shows that if the plants are properly screened, they could probably yield drugs of pharmaceutical importance. Future work to isolate, purify and characterize these bioactive constituents is therefore recommended.

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