Journal of Medicinal Plants Research Vol. 5(6), pp. 1031-1033, 18 March, 2011
Available online at http://www.academicjournals.org/JMPR
ISSN 1996-0875 ©2011 Academic Journals

Short Communication

Bioactivity of rhizome essential oils from two varieties of *Cyperus articulatus* (L.) grown in Nigeria, using brine shrimp (*Artemia salina*) lethality tests

O. M. Ameen[1]*, L. A. Usman[1], I. A. Oladosu[2], N. O. Olawore[3] and I. A. Ogunwande[4]

[1]Department of Chemistry, University of Ilorin, Ilorin, Kwara State, Nigeria.
[2]Department of Chemistry, University of Ibadan, Ibadan, Nigeria.
[3]Department of Chemistry, Ladoke Akintola University, Ogbomosho, Nigeria.
[4]Department of Chemistry, Lagos State University, Ojo, Lagos State, Nigeria.

Accepted 20 October, 2010

Pulverized rhizomes (500 g) of red and black varieties of *Cyperus articulatus* were separately hydrodistilled to obtain rhizome essential oils. The red and black varieties afforded oil with yields of 0.62 and 0.60% w/w, respectively. Both were tested for bioactivity using brine shrimp lethality test, the two oils samples showed significant activity with LC_{50} of 2.84 µg/ml and 3.34 µg/ml for red and black varieties, respectively.

Key words: Bioactivity, brine shrimp lethality test, *Cyperus articulatus*, essential oils.

INTRODUCTION

Essential oils are known for their biological activities. These attributes are reflected in their antimicrobial, antifeedants, insecticidal, larvicidal and molluscidal properties (Laurent et al., 1997; Isman et al., 1990; Koul et al., 1990; Lahlou et al., 2001; Ogunwade et al., 2001). The aforementioned properties are due to separate and synergetic actions of the oil constituents on the organisms (Lahlou, 2004). Constituents of essential oil include monoterpenoids, sesquiterpenoids and aromatic compounds. However, monoterpenoids and sesquiter-penoids constitute a large proportion of essential oils. Many sesquiterpenes of caryophyllane, eudesmane, patchoulane and rotundane types are present as hydrocarbon and oxygenated compounds in many essential oils. For example, mustakone and cryophyllene oxide have been identified in rhizome essential oil of Brazilian grown *Cyperus articulatus* by Zoghbi et al. (2006). Sonwa and Konig (2001) had also isolated (-) – norotundane, (-) – isorotundone, cypera – 2, 4(15) – diene and (+) – cyperadone from *Cyperus rotundus*.

Inouye et al. (2001), had reported that bacteria are more susceptible to oxygenated compounds than hydrocarbon, for instance in the antimicrobial investigation of essential oil constituents they found that the following oxygenated monoterpenoids; cinnamaldehyde, citral, penalladehyde, octanol, and nonanol were active against *Escherichia coli*, *Staphylococcus aureus*, *Streptococcus pyogen*, *Staphylococcus pneumonia* and *H. influenza*. Similarly, Magiatis et al. (2002) reported the antibacterial properties of camphor and 1, 8 – cineole found in the essential oils of *Achillae frasil* and *Achillae toygetea*. The properties of these two compounds were earlier reported by Prudent et al. (1993) and Aligiannis et al. (2000).

The antifeedants and cytotoxic properties of volatile oils of *Echiochilon fruiticosum* (Desf.) against adult of *Tribolium confusum* was reported by Zardi – Bergaoui *et al* (2007). The cytotoxic activities of the essential oils of *Aeollanthus pubescens* (Benth.) and *Ocimum gratissimum* on the human epidermic cell line HaCat have also been evaluated (Koba et al., 2008), while the

*Corresponding author. E-mail: moameen@unilorin.edu.ng, aommohd@yahoo.com. Tel: +2348035019199.

essential oil of *Cryptomeria japonica* was found to displayed significant brine shrimp larvae lethality (Cheng et al., 2003). *C. articulatus* (L.) is a tropical sedge widely grown in Nigeria, where it is commonly known as “Kajiji” by the Hausa, “Woire” by the Fulani and “Ifin” by the Yoruba (Lowe and Stanfield, 1974). *C. articulatus* is used in traditional medicine for the treatment of several ailments like epilepsy, malaria and dysentery in different parts of the world. For instance, its rhizomes are used in the treatment of migraine in Gabon (Melching et al., 1997). The non-volatile extracts of the rhizomes of *C. articulatus* are known to posses anti-convulsant, anti-malaria and anti-microbial properties (Inglis, 1994; Mongeill et al., 1995; Etkin., 1997; Bum et al., 2001). Repellant and antifeedant properties of these extracts have also been reported (Abubakar et al., 2000). Essential oils from *Cyperus* species are generally constituted by sesquiterpenoids and traces of monoterpenoids, with cyperene being the main hydrocarbon (Triveds, 1964). The rhizome essential oils of red and black varieties of *C. articulatus* growing in Nigeria had earlier been characterized (Olawore et al., 2006). Like other *Cyperus* species both oils were richer in sesquiterpenoids than monoterpenoids. However, the oils showed substantial quantitative and qualitative variations in their constituents. Some of the identified constituents were present in both oils while others showed characteristics of one of the sample. Oil of the red variety was characterized by the abundance of cypertundone, piperitone, α – maaline, germacrone, γ – epicubenol, α – pinene and cyperene oxide, while in the oil of the black variety the main constituents were; cedrol, guai – 5 – en – 11 – ol, cyperotundone, sabinene, α–pinene, trans – pinocarveol, cis – carveol, trans – carveol and α–cardinal (Olawore et al., 2006). With the abundance of oxygenated sesquiterpenoids in both oils, they are expected to be biologically active. These activities have been reported by various workers (Inglis, 1994; Mongeill et al., 1995; Etkin., 1997; Abubakar et al., 2000; Bum et al., 2001). However, the cytotoxicity of the oil is yet to be evaluated as a result we set out to evaluate the cytotoxicity of the two oils using brine shrimp lethality test (BST) and to investigate variation if there is any in the cytotoxicity of the two oils.

MATERIALS AND METHODS

Plant materials

The dried rhizomes of red and black varieties of *C. articulatus* were obtained in Ilorin, Kwara State, Nigeria. Identification was carried out at the herbarium of the Forestry Research Institute of Nigeria, Ibadan, where voucher specimens were deposited.

Oil isolation

Pulverized rhizome of the red and black types of *C. articulatus* (500 g) were separately hydrodistilled in a Clevenger – type apparatus according to the British Pharmacopoeia specification.

Brine shrimp lethality bioassay

The modified method of McLaughlin et al. (1998) was employed in this study. Natural sea water was poured into an improvised hatching chamber made of plastic dish, brine shrimp eggs were added at the closed section of the chamber. The open air section of the chamber was then exposed to fluorescent light for 48 h. Bottles of the same size used were washed and sterilized before use. Different concentration of the oils of the two varieties of *C. articulatus* were prepared using n – hexane in triplicates (1000, 100, 10 μg/ml). After 48 h, a drop of DMSO and 4 ml of sea water were added to each of the sample bottles containing the oil sample. Ten brine shrimp larvae were carefully counted into each of the sample bottles and the volume of the sea water was made up to 5 ml. Tests for each concentration was done in triplicate. A control experiment containing 5 ml of sea water, a drop of DMSO and ten brine shrimp larvae was set along side. The experiment was maintained at room temperature for after 24 h the number of surviving larvae were counted and recorded, the data obtained were subjected to Finney’s Probit analysis to determine the LC_{50} of each oil. The toxicity is expressed by this LC_{50} which is defined as concentration of the oil that kills 50% of the larvae within 24 h. percentage mortality was also calculated as number of dead larvae divided by initial number of larvae (10) multiply by 100.

$$\% \text{ Mortality} = \frac{\text{No. of Dead larvae}}{\text{Initial no. of live larvae}} \times 100$$

RESULTS AND DISCUSSION

A substance is considered to be cytotoxic if it inhibits vital metabolic processes or it causes disorders in living organisms resulting in perversion of behavior or death (Fatope, 1995). Various essential oils have been known to demonstrate cytotoxic activities, (Zardi – Bergaoui et al., 2007; Koba et al., 2007; Cheng et al., 2003). The brine shrimp assay basically detects substances that are cytotoxic enough to kill shrimp’s larvae on exposure to solution of the sample.

The results of the brine shrimp lethality assay (BST) of the essential oils of the two varieties of *C. articulatus* are shown in Table 1. In the table, the cytotoxicity of both oils increase with increase in concentration of the oils. For the oil of the red type none of the larvae survived at 1000 and 100 μg/ml, however, six of the larvae survived at 10 μg/ml which amount to 80% mortality. The cytotoxicity of the oil of the black type followed the same pattern as reported for the oil of the red type. Meanwhile, nine of the larvae survived at 10 μg/ml amounting to 70% mortality. LC_{50} for the oil of the red and black varieties are 2.59 and 3.38 μg/ml respectively. Brine shrimp lethality assay is a rapid inexpensive and simple bioassay for testing plant extracts including essential oils bioactivity, the result of which in most cases correlate with cytotoxic and antitumor properties of the plant It has been established that the oil of the black variety which is richer in terpenols than oil of the red type (Olawore et al., 2006), hence the oil of the

Table 1. Brine shrimp lethality results for the rhizome essential oils of the red and black varieties of *C. articulatus*.

C. articulatus (type)	Dosage (µg/ml)	Survivals			No of mortality	% mortality	LC_{50} (µg/ml)
		1	2	3			
Red	10	3	2	1	24	80	2.59
	100	0	0	0	30	100	
	1000	0	0	0	30	100	
Black	10	0	9	0	21	70	3.38
	100	0	0	0	30	100	
	1000	0	0	0	30	100	

black variety is expected to exhibit higher bioactivity (Inouye et al., 2001). However, the result obtained contradicts this assertion because though, both oils exhibit significant activities against the brine shrimp larvae, but the oil of the red variety has higher lethality than the oil of the black variety as reflected in their LC_{50}, what is responsible for this observation is not yet ascertain since the mechanism of the action has not been investigated. This result corroborates the folk medicinal uses of *C. articulatus* and also the results of an earlier work as repellant and as an antifeedant (Abubakar et al., 2000). This property is attributed to cytogenic and toxic compounds present in the oils. Thus, the oils may possess potent antitumor compounds, which are of importance in pharmaceutical industries.

REFERENCES

Abubakar MS, Abdurahman EN, Haruna AK (2000). The repellant and antifeedant properties of *cyperus* against *Tribolium easteneum* Hbst. Phytotherap. Res., 14: 281–283.

Aligiannis N, Kalpoutzakis E, Chinou IB, Mitakou S, Gikas E, Tsarbopoulos A (2000). Composition and antimicrobial activity of the essential oil of five taxa of siderites from Greece. J. Agric. Food Chem., 49: 811–815.

Bum, EN, Schmutz N, Neyer C, Rakotonirina A, Bopelet N, Portet C, Jeker A, Rokotonirina SV, Olpe, HR, Hercling P (2001). Anticonvulsant properties of methanolic extract of *cyperus articulatus*. J. Ethnopharm., 76: 145–150.

Cheng SS, Chang HT, Chang ST, Tsai KH, Chen WJ (2003). Bioactivity of selected plant essential oils against the yellow fever mosquito *Aedis aegypti* and brine shrimp, Bioresour. Technol., 89(1): 99–102.

Etkin NL (1997). Antimalaria plants used by Hausa in Northern Nigeria. Trop. Doctor, 27: 12–16.

Fatope MO (1995). Phytocompounds: Their bioassay and diversity. Discovery and Innovation, 7(3): 229–236.

Inglis C (1994). Checklist of economically important cyperaceae. Cyperaceae Newsletter, 4: 13.

Inouye S, Takizawa T, Yamaguchi H (2001). Antibacterial activity of essential oils and their major constituents against respiratory tract pathogens by gaseous contact. J. Antimicrob. Chemother., 47: 565–576.

Isman MB, Koul O, Luczynski A, Kaminski J (1990). Insecticidal and antifeedants bioactivities of Neem oils and their relationship to Azadirachtin content. J. Agric. Food. Chem., 38: 1406.

Koba K, Sanda K, Guyon C, Raynaud C, Millet J, Chaumont JP, Nicod, L (2007). Chemical composition and in vitro cytotoxic activity of essential oils from two tropical Lamiacea: *Aeollanthus pubescens* Benth. and *Ocimum gratissimum* L., J. Essent. Oil. Bear. Plants, 10: 60–69.

Koul O, Smira MJ, Isman MB (1990). Asarones from Acorus calamus L. oil their effect on feeding behaviour and dietary utilization in *Peridroma saucia*. J. Chem. Ecol., 16: 1911–1920.

Lahlou M (2004). Methods to study the Phytochemistry and bioactivity of essential oils. Phytother. Res., 18: 435–448.

Lahlou M, Berrada R, Hmamouchi M (2001). Molluscidal activity of thirty essential oils on Bulinus truncates. Thérapie, 56: 71–72.

Laurent O, Vilaseca LA, Chantraine JM (1997). Insecticidal activity of essential oils on triatomo infestans. Phytother. Res., 11: 285–290.

Lowe J, Stanfield DP (1974). The flora of Nigerian sedges (family cyperaceae). Ibadan University Press, pp. XVII–XVIII.

Magiatis P, Skaltsounis A, Chinou I, Haroutounian SA (2002). Chemical composition and in – vitro antimicrobial activity of the essential oils of three Greek Achillea Species. Z. Naturforsch. 57c: 287–290.

McLaughlin JL, Rogers LL, Anderson JE (1998). The use of biological assays to evaluate botanicals. Drug Inf. J., 32: 513–524.

Melching S, Bulow N, Wihstutz K, Jung SA, Konig WA (1997). Natural occurrence of Enatiomers of cadina – 3, 3 – diene and 8 – amorphene. Phytochem., 44: 1291.

Mongeill E, Desmarcheller C, Coussio J, Cicnu G (1995). Antimicrobial activity and DNA interaction in medicinal plants from the Peruvian Amazom. Rev. Argeul., Microbiol., 22: 199–203.

Ogunwade IA, Bello MO, Olawore ON, Muili KA (2001). Phytochemical and antimicrobial studies on *butyrospermum paradoxum*, Fitoterapia, 72: 54–56.

Olawore NO, Usman LA, Ogunwade IA (2006). "Volatile Constituents of Cyperus articuletus. J. Essent. Oil Res., 18(6), 604–606.

Prudent D, Perineau F, Bessiere JM, Michael G, Bravo R (1993). Chemical analysis, bacteriostatic and fungistatic properties of the essential oil of the atoumau from Martinique (*Alphinia speciosa*). J. Essent. Oil Res., 5: 255–264.

Sonwa MM, Konig WA (2001). Chemical study of the essential oil of *Cyperus rotundus*. Phytochem., 58: 799–810.

Triveds B, Motl O, Smolikova J, Sorm F (1964). Structure of the hydrocarbon cyperene. Tetrahedron Lett., Lett., 1197.

Zardi BA, Jannet HB, Harzallah – Skhiri F, Chaeb I, Hammami S, Mighri Z (2008). Chemical composition, toxicity and antifeedant activities of the Tunisian of *Echiochilon fruiticosum* Desf. Aerial part volatile fractions against *Tribolium confusum* du val, JEOBP, 11(1): 112–19.

Zoghbi BM, Adrade AE, Oliveira J, Carreura ML, Giulthou SPN (2006). Yield and chemical composition of the essential oil of the stems and rhizomes of *cyperus articulatus* L. cultivated in the state of Para, Brazil. JEOR, 18(1): 10–12.