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ORIGINAL ARTICLE

Volatile constituents and biological activities of the leaf and root of *Echinacea* species from South Africa



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Abstract *Echinacea* is used ethnomedicinally for the treatment of various diseases such as cough, respiratory infections, and bronchitis among other uses in Eastern Cape region of South Africa. This study evaluated the volatile components of the essential oil of the plant, its toxicity, anti-inflammatory and analgesic activities in rodents.

Dried leaf and root of the plant were separately processed by hydrodistillation for 4 h and their essential oils (EOs) were collected. Extracted oils were subjected to GC/GC–MS analysis. The essential oil was further evaluated for acute toxicity, anti-inflammatory and analgesic activities. The toxicity profile of the essential oil was evaluated in mice through the oral route (p.o.), and anti-inflammatory activity was evaluated on the carrageenan-induced edema model in rats at the doses of 100–200 mg/kg, while its analgesic effect was evaluated on the acetic acid-induced writhings model in mice at doses of 100–200 mg/kg.

GC/GC–MS analysis of EOs showed that a number of compounds identified in the leaf and root oils were 25 and 31 respectively. The chemical compositions of the oils varied and the major compounds identified in the oils include germacrene D, naphthalene, caryophyllene oxide, α -phellandrene and α -cadinol. The essential root oil did not cause mortality at the highest dose of 5000 mg/kg; hence, its LD₅₀ was estimated to be ≥ 5000 mg/kg, p.o. The anti-inflammatory test

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results showed that the essential root oil caused significant ($p < 0.05$ – 0.01) reduction in edema size compared to the negative control group on the carrageenan-induced edema and the results for the analgesic test showed that the essential root oil caused significant ($p < 0.05$) reduction in number of writhings at 1000 mg/kg compared to the negative control group.

It is concluded that root and leaf of this *Echinacea* species contain volatile oils which varied in their yield and chemical compositions. The essential root oil is non-toxic orally and it demonstrated significant anti-inflammatory and analgesic activities in laboratory animals.

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1. Introduction

Medicinal plants are plants that are assumed to have healing properties and often used to treat some illnesses. These medicinal plants have been identified and used throughout the human history, making an impact on both world health and international trade for treating various diseases such as diarrhea, wounds, and dry cough (Van Wyk et al., 1997).

Echinacea also known as purple coneflower is a medicinal plant that belongs to the Asteraceae family. The plant consists of more than 19 species with three species of medicinal interest known as *Echinacea purpurea*, *Echinacea angustifolia* and *Echinacea pallida*. *Echinacea* has been used for centuries by Native Americans for the treatment of various diseases (Turner et al., 2000). Currently, *Echinacea* is being used to combat bacterial, viral, protozoan and fungal infections, and as an anti-inflammatory agent and as a possible chemo preventative agent (Barnes et al., 2005).

All the three species of medicinal interest of *Echinacea* contain varying amounts of essential oils in root, leaf, flower and aerial parts (Mazza et al., 1999). Essential oil of *E. purpurea* contains caryophyllene, caryophyllene epoxide, germacrene D and borneol while that of *E. angustifolia* and *E. pallida* contains ketoalkynes and ketoalkenes (Mazza et al., 1999; Hudaib et al., 2002).

Essential oils have been recognized as therapeutic agents and widely utilized as potent natural medicinal components of plants (Djilani and Dicko, 2012). Essential oils are complex mixtures of volatile constituents frequently containing 20–60 or more individual compounds (Miguel, 2010). Chemical composition of a particular species can vary greatly and some factors that have been identified to be responsible include genetic, geographical location, climatic changes, growing conditions, seasonal variation, time of collection etc. among other factors (Andrade et al., 2011). Common classes of compounds found in essential oils include hydrocarbons, esters, oxides, lactones, alcohols, phenols, aldehydes and ketones. These components have been reported to be responsible for several bio-activities ascribed to essential oils (De Sousa et al., 2011). In this study the essential oils of root and leaf of *Echinacea* were evaluated for chemical composition and biological activities in order to determine its contribution to the reported bioactivities of the plant and to validate its traditional uses in treating some diseases including infections, respiratory diseases and pain-related ailments.

In the Eastern Cape region of South Africa, *Echinacea* is a popular herbal medicine used by the traditional healers to manage various diseases including wound healing, respiratory infections, and pain among several others. The dried leaf and

root of the *E. purpurea* species used in this study was supplied by the Indigenous Knowledge System holder (Mr. Reuben Matewu) and was among the several medicinal plants submitted for chemical and biological studies for the purpose of verifying their medicinal efficacies in laboratory animals by our research group.

2. Materials and methods

2.1. Drugs, chemicals and reagents

Acetic acid (BDH Chemical Ltd., Poole, England), diclofenac potassium (Diclogesic® Supreme), aspirin. All chemicals and reagents were obtained from Sigma–Aldrich Chemical Co. (St Louis, MO, USA). All the chemicals used including the solvents, were of analytical grade.

2.2. Animals

Mice and rats were obtained from the South African Vaccine Initiative, Johannesburg, and kept at the Animal Holding Facility, Zoology Department, WSU. Male and female Wistar rats (200–300 g) were randomly selected ($n = 6$), and were used for the anti-inflammatory test. Male Swiss mice (25–35 g; $n = 6$) were also used for the acute toxicity and the analgesic tests. The animals were kept under standard conditions of temperature and humidity and had free access to rat chow and water. Food was however withheld overnight prior to experiments while water was provided ad libitum. This study was approved by the Department of Higher Education, WSU, and Ethical Clearance Approval obtained, Walter Sisulu University Ethics Committee Reference No. DVC (AA&R) DRD/SREC: Reference No: 31.

2.3. Plant collection

Dried *Echinacea* root and leaf was supplied by the Herbal Practitioner (IKS holder) at King William's town, Eastern Cape on the 3rd of July, 2014.

2.4. Extraction of the essential oil

Essential oil of the dried leaf (342 g) and root (368 g) of the plant were obtained by hydrodistillation using the Clevenger-type apparatus for 4 h (British Pharmacopoeia, 1980). The oils were separately collected in airtight glass vial containers, and stored at 4 °C (Oyedeji et al., 2006) before analysis. The yields

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