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Chemical composition and biological activities of essential oil from the leaves of *Sesuvium portulacastrum*

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Abstract

Sesuvium portulacastrum has long been used as a remedy for fever and scurvy. Hydrodistillation was used to extract the essential oil from the fresh leaves of *Sesuvium portulacastrum*. The essential oil yield obtained was 0.15%. Using GC–MS analysis, α-pinene, camphene, β-pinene, α-terpinene, *O*-cymene, limonene, 1,8-cineole, α-terpinene, bornyl acetate, tridecane, *trans*-caryophyllene and α-humulene were identified. The hole plate diffusion method was used for antibacterial testing. The essential oil exhibited antibacterial activity against *Acetobacter calcoacetica*, *Bacillus subtillis*, *Clostridium sporogenes*, *Clostridium perfringens*, *Escherichia coli*, *Salmonella typhii*, *Staphylococcus aureus* and *Yersinia enterocolitica.* The mycelium growth inhibition method was used for the antifungal testing. The oil exhibited antifungal activity against *Candida albicans*, *Aspergillus niger*, *Aspergillus flavus* and *Penicillium notatum*. Using the β-carotene, acetone and linoleic acid method for the antioxidant testing, the essential oil showed antioxidant activity threshold of 15.9 mm mean zone of color retention.

Keywords: *Sesuvium portulacastrum*; Essential oil; Antibacterial; Antifungal; Antioxidant; GC–MS; Chemical components

1. Introduction

Some plant species have demonstrated antibacterial, antifungal and antioxidant activity (Graven et al., 1992; Gundidza, 1993; Rabe and van Staden, 1997; Burits et al., 2001; Lee et al., 2003). The volatile compounds have great application and demand in food, perfumery, cosmetics, pharmaceutical and winery industries. In our search for commercially useful essential oils, *Sesuvium portulacastrum* was selected as one of the plants for study.

This plant is distributed throughout the world since it is used as an ornamental plant. It is found in the northern, western and central parts of Zimbabwe (Espejel, 1986; Taylor, 1992). The thick, fleshy leaves are borne on succulent, reddish-green stems that branch regularly forming dense stands close to the ground. Small, showy pink flowers are borne more or less continually throughout the year.

Sesuvium portulacastrum has a long history of use in folk medicine. Traditional healers in Zimbabwe and South Africa use the plant to treat various infections and kidney problems. It has been used in traditional medicine as a remedy for fever, kidney disorders and scurvy (Rojas et al., 1992). Recent studies have shown that the major phytoconstituents of *Sesuvium portulacastrum* are *trans*-4-hydroxyprolinebetaine, praline and 3,5,4′-trihydroxy-6,7-dimethoxyflavone 3-glucoside (Khajuria et al., 1982; Adrian-Romero et al., 1998). The occurrence of these compounds is associated with a possible role in osmoregulation (Wyn Jones, 1980; Adrian-Romero et al., 1998).

It has been recorded that *Sesuvium portulacastrum* and other similar plant species, such as *Carpobrotus edulis* adapted either to dry habitats or saline environments, have medicinal properties (Conco, 1991; van der Watt and Pretorius, 2001; Burits et al., 2001). These plants have been used as traditional medicines by the indigenous people in Africa, Latin America and in Asian countries, such as India, China, Pakistan and Japan (Conco, 1991; Rabe and van Staden, 1997; Hoareau and DaSilva, 1999). The secondary metabolites from these plant species have been

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shown to have a great potential as substitutes for some synthetic raw materials in food, perfumery, cosmetics and pharmaceutical industries (Lis-Balchin and Deans, 1997). As a result, there is a great effort to screen plants for phytoconstituents and biological activities worldwide in the bid to produce more cost-effective remedies that are affordable to the poor populations of the world (Lis-Balchin and Deans, 1997; Webber et al., 1999).

The essential oil from this plant was analyzed for the chemical components. It was also tested for its antibacterial activities against a wide spectrum of Gram-positive and Gram-negative bacteria. In addition, the essential oil was subjected to antifungal and antioxidant testing. The major chemical components obtained were α-pinene, camphene, β-pinene, α-terpinene, *O*-cymene, limonene, 1,8-cineole, α-terpinene, bornyl acetate, tridecane, *trans*-carryophyllene and α-humulene. The essential oil exhibited notable antibacterial activity against both Gram-positive and Gram-negative bacteria as well as significant antifungal and antioxidant activity.

2. Materials and methods

2.1. Plant material

The plant material was collected in Zimbabwe with the authorization of the Zimbabwean Government and in agreement with the United Nation Convention on Biodiversity. The plant was identified by botanists at the National Herbarium and Botanic Gardens in Harare, Zimbabwe. Three voucher specimens were deposited at the National Herbarium and Botanic Gardens in Harare, Zimbabwe and one at the Herbarium of the Department of Pharmacy at the University of Zimbabwe.

2.2. Essential oil extraction

The leaves (1000 g) were subjected to hydrodistillation for approximately 3 h using a Clavenger-type apparatus. The essential oil yield obtained was 0.18% (v/w). It was dried over anhydrous sodium sulphate. After filtration, it was stored at approximately 4 °C until tested and chemically analyzed. The essential oil was subjected to GC/MS analysis, antibacterial, antifungal and antioxidant testing.

2.3. Gas chromatography and mass spectroscopy analysis

2.3.1. Method of analysis

A wet needle of the essential oil was inserted directly into the inlet (spitless mode) of a Hewlett Packard 6890 Gas Chromatograph. The temperature of the injection port was set at 220 °C, while the pressure at the inlet was maintained at 3.96 psi. A HP-5MS (cross linked 5% phenyl methyl siloxane) column (30 m × 0.25 mm × 0.25 μm film thickness) was temperature programmed from 60 to 150 °C at 3 °C/min after a 3.5 min delay. Helium was used as a carrier gas at 0.7 ml/min. Mass spectra were recorded by a HP 5937 series mass selective detector (MSD).

2.4. Antibacterial testing

2.4.1. Test organisms

The test organisms were selected on the basis that they cause a lot of infections in humans. Both Gram-positive and Gram-negative bacterial species were selected as test organisms. The organisms were obtained from the Department of Pharmacy at the University of Zimbabwe, who in turn obtained them from Scotland as pure characterized strains.

2.4.2. Inoculation procedure

The isosensitest broth was inoculated aseptically with the appropriate microorganism 24 h before testing. This was to ensure that the bacteria fully adapted to the broth. The procedure was repeated for each bacterial species. The inoculated bacterial strains were incubated at 25 °C for 24 h. The procedure was repeated for each bacterial species.

2.4.3. Determination of antibacterial activity of essential oils

Essential oils were diluted with absolute alcohol to produce the following concentrations: 10, 20, 50 and 100% (v/v). Agar was melted in a steam bath set at 30 °C to prevent solidification. Four Petri dishes were pre-inoculated with the appropriate bacteria in the following manner. One millilitre of the bacterial suspension was pipetted into the appropriately labeled Petri dish to which 25 ml of molten agar was then added followed by thorough mixing of the bacteria and molten agar. The agar was allowed to set for 1 h. Four 4 mm wide holes were then made in the agar using a borer. Oil (25 μl) of a specific concentration was introduced into each of the holes in an appropriately labeled Petri dish using a sterile micropipette. Gentamicin (10 μg/ml) was used as a positive control and absolute alcohol as a negative control. The dishes were then incubated at 25 °C for 24 h after which zones of inhibition were measured and recorded. The zone of inhibition was taken to be the diameter of the zone visibly showing the absence of growth including the 4 mm hole. Where there was no inhibition, the value of 0 mm was assigned to the test sample.

2.5. Antifungal testing

2.5.1. Test organisms

The fungal species were chosen on the basis that they cause serious systemic and skin infections in humans, especially in people living with HIV/AIDS. Four fungal species were used for the antifungal testing, namely: *Candida albicans*, *Aspergillus niger*, *Aspergillus flavus* and *Penicillium notatum*. These were obtained from the Department of Pharmacy at the University of Zimbabwe. The fungal species were pure characterized strains from Scotland.

2.5.2. Determination of antifungal activity of essential oils

The medium was inoculated with 10 ml of the fungal suspension and shaken thoroughly to mix. The essential oil was then added into the flasks and three flasks were used for each oil concentration. The volumes of the oils used were 25, 50 and 100 μl