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Full Length Research Paper

Isolation of volatile compounds of *Aloe excelsa* (Berger)

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Industrial and pharmacological applications of volatile and non-volatile compounds isolated from plants have been dominating the commercial sector over the recent two decades. Attempts in isolation of volatile compounds of aloes have impact on the medicinal as well as the cosmetic industries. Volatile compound isolation and extraction was performed on leaf exudates of *Aloe excelsa.* Three commercially important compounds, namely: phenylacetonitrile, carvone and limonene were identified using gas chromatography mass spectroscopy (GC-MS).

Key words: *Aloe excelsa*, phenylacetonitrile, carvone, limonene, volatile compounds.

INTRODUCTION

Aloe excelsa being one of the larger species of Aloe belongs to the family Aloaceae (Glen et al., 1997), and it occurs in abundance in Zimbabwe. It has been used extensively as a traditional remedy by indigenous people. Extensive studies of extraction and isolation has been carried out on *Aloe ferox*, *Aloe vera* as well as *A. excelsa* (Speranza et al., 1986, 1990; Koyama et al., 1994; Eloff, 1998; Amabeoku et al., 1998; Dagne, 2000; Coopoosamy and Magwa 2007). This investigation deals with the isolation of volatile compounds present in the leaf exudates of *A. excelsa*.

MATERIALS AND METHODS

Isolation of volatile compounds in *Aloe excelsa*

Collection of plant materials

The plant materials were obtained from Zimbabwe, with the assistance of Prof. K.Dzama, Professor, University of Cape Town and Ratidzayi Takawira-Nyenya, Principal research Officer, National Herbarium and Botanical Garden, Zimbabwe.

Abbreviation: GC-MS, Gas chromatography mass spectroscopy analysis.

Steam distillation and volatile compound isolation

Approximately 1 kg of *A. excelsa* leaves was put in a 5 L flask which contained approximately 2 L of distilled water and placed on a steam distiller (heating mantle) at a boiling temperature for approximately 3 h until 1 ml oil was recovered. Steam was generated from the boiling water in the flask and the oil mixture was extracted from the plant material. The volatile extracts (oils) were collected in a graduated arm of Clevenger apparatus and the oil samples were then subjected to gas chromatography mass spectroscopy analysis (GC-MS). All tests were done in triplicate.

Gas chromatography technique (GC)

The exudates obtained from steam distillation were then analyzed on a Hewlett Packard (HP) 6890 Gas Chromatograph with a wet needle of the sample material being directly inserted into the inlet (spotless mode). The column consisted of a cross-linked 5% pH ME Siloxane on 30 x 0.25 mm x 0.25 µm thick film with a column head pressure of 55 Kpa. The carrier gas used was helium and the flow was 35 cm/s-split flow 30 – 40:1. The temperature of the injection port was set at 220°C. The oven temperature was programmed from 60 – 150°C at 3°C/mm after a 3 min delay. A HP 5973 Series Mass Selective Detector (MSD) recorded the mass spectra. Essential oil constituents were identified on the basis of retention time, co- injection with authentic compounds and GC-MS.

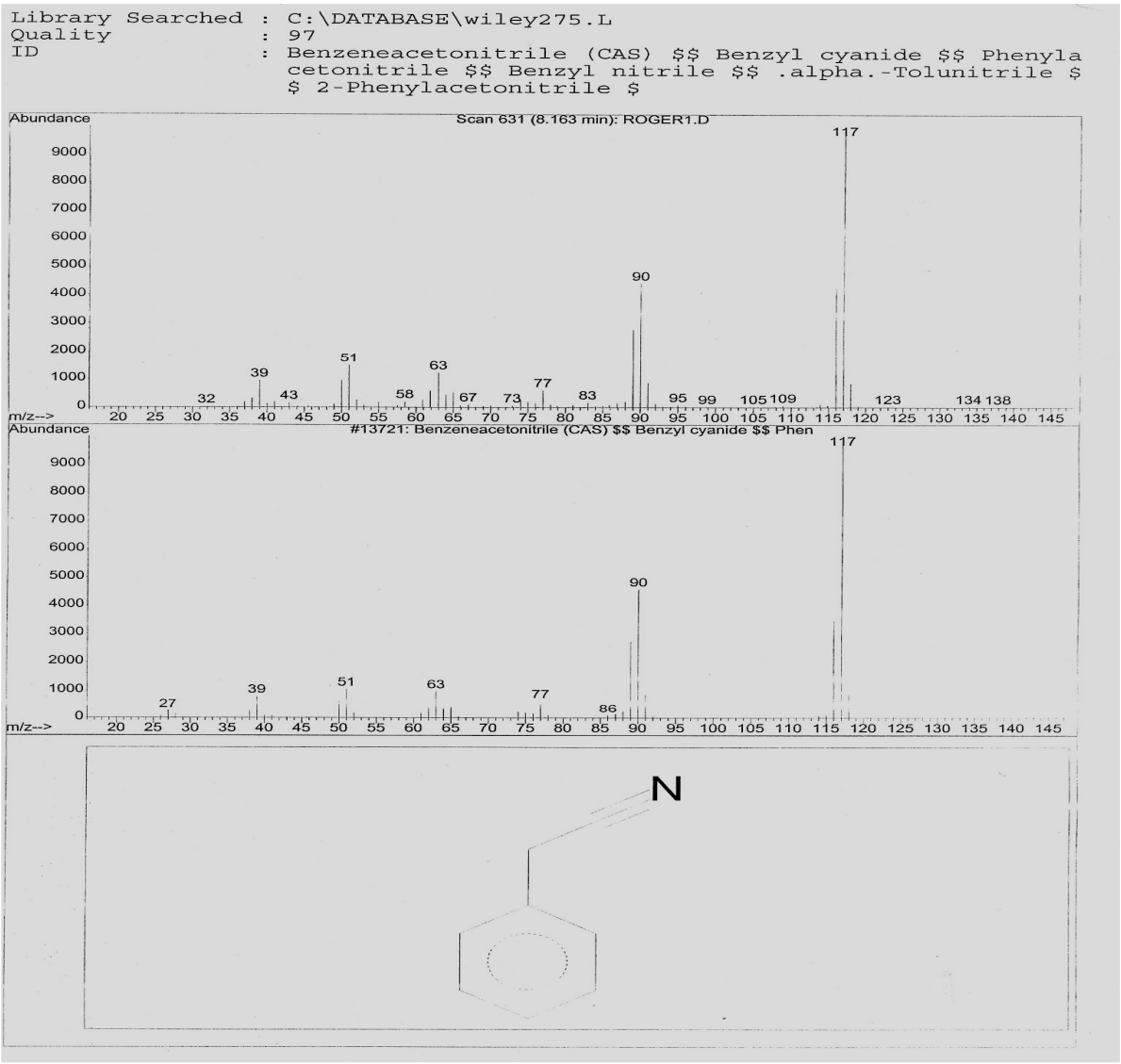

Figure 1. GCMS analysis of phenylacetonitrile.

RESULTS AND DISCUSSION

Isolation of the volatile compounds: limonene, carvone and 2-phenylacetonitrile were for the first time obtained from *A. excelsa.* The GC-MS results indicate a high percentage level of all three compounds (Limonene, Carvone and 2-phenylacetonitrile) (Figures 1-4). However, the most abundant compound was limonene at 99% followed by carvone and 2-phenylacetonitrile both at 97%.

A. excelsa volatile extract was characterized by limonene (99%), carvone (97%) and 2-phenylacetonitrile (97%). This is in contrast with *A. ferox* which also showed a series of volatile compounds (Magwa et al., 2006). The significance of these volatile compounds has been associated with the well-being of the species (Magiatis et al., 1999; Tzakou et al., 2001; Filipowicz et al., 2003, Venkatsen et al., 2005; Magwa et al., 2006).

Limonene is well known for its antimicrobial and antiseptic activities (Magiatis et al., 1999; Martins et al., 2000; Tzakou et al., 2001; Filipowicz et al., 2003, Magwa et al., 2006). However, there is a higher percentage (99%) of limonene in *A. excelsa* than in *Sesuvium portulacastrum* which was investigated by Filipowicz et al. (2003) and Magwa et al. (2006). It is also assumed that the same phenomenon is applicable to *A. excelsa* as it also demonstrated high antimicrobial activity. With regards to cosmetics, nutrition or health application, limonene is well-known for flavour and fragrance additives in perfumes, soaps, foods, chewing gums and beverages (Venkatsen et al., 2005; Chen et al., 2006; Erazo et al., 2006). Limonene has been known to be beneficial in the

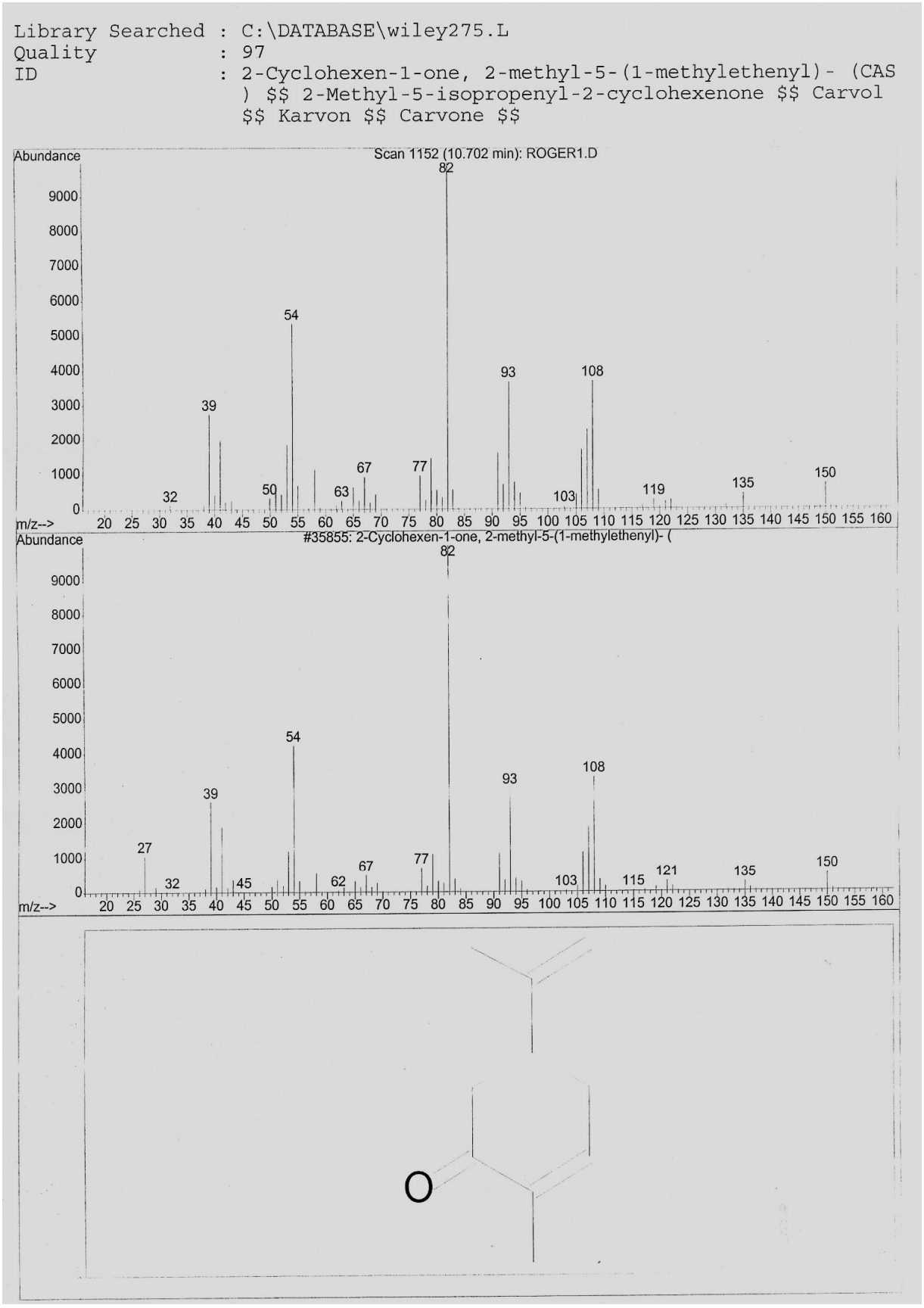

Figure 2. GCMS analysis of carvone.

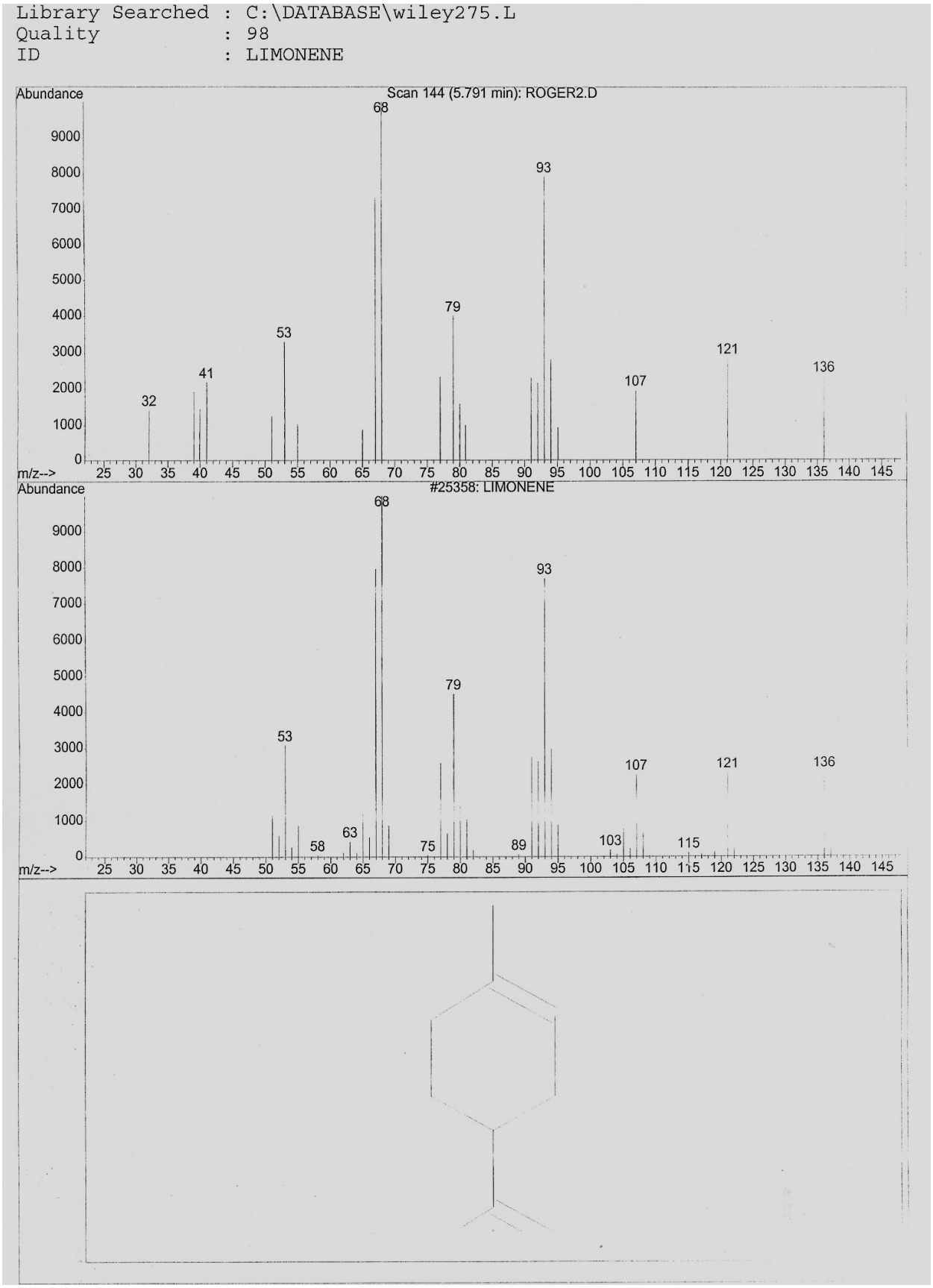

Figure 3. GCMS analysis of limonene at 98%.

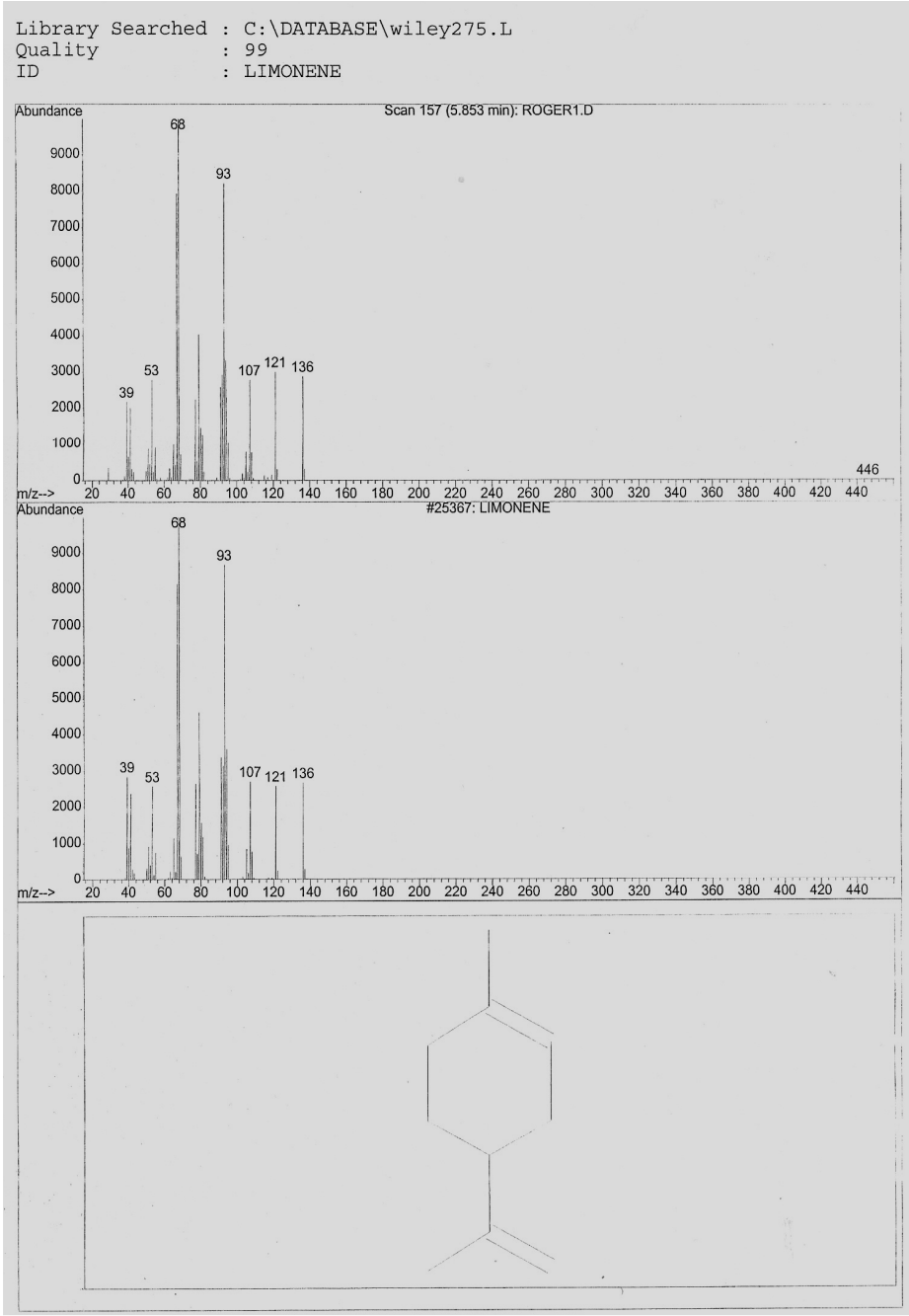

Figure 4. GCMS analysis of limonene at 99%.

nutritional sphere. D-limonene is often present in fruit juices, ice cream, sweets and chewing gum. The dietary intake of limonene varies depending on the type of food consumed. In health-related applications, limonene is used for oral administration (Jain and Filipps, 1991). During oral administration d-limonene is rapidly absorbed in the gasterointestinal tract in humans (Magiatis et al., 1999; Martins et al., 2000; Tzakou et al., 2001; Filipowicz et al., 2003). It is assumed that once absorbed d-limonene is rapidly distributed to the different tissues of the body and is readily metabolized (Martins et al., 2000; Tzakou et al., 2001).

Carvone and phenylacetonitrile are regarded as highly volatile compounds (De Carvalho et al., 2006). They also seem to be predominant in *A. excels,* however, their physiological significance in *A. excelsa* is not yet known. It is assumed that phenylacetonitrile is responsible for defense strategy (Venkatsen et al., 2005; Chen et al., 2006; Erazo et al., 2006). This evidence is supported by the fact that many plant products which contain these

compounds constitute the ingredients of pesticides used against pests and herbivores (De Carvalho et al., 2006). However, the carvone and phenylacetonitrile in *A. excelsa* gel is assumed to enhance the skin defense mechanisms. This contention has been demonstrated by the Aloe gel from *A. ferox* and *A. vera* which have been consistently used as a topical treatment for skin problems and burn wounds. It could also be speculated that as these compounds constituted the terpene series, they might be involved in many biological activities. The isolation of carvone from *A. excelsa* further substantiates its importance as a pharmacological important species. The isolation of carvones also confirms the use of *A. excelsa* in traditional medicines.

The physiological significance of carvones could be attributed to the regulation of plant signals which lead to the attraction of pollinators during the flowering season as well as defense strategies (De Carvalho et al., 2006). It is assumed that this compound is associated with defense mechanisms in *A. excelsa*. In this regard, an interaction between carvone and phenyacetonitrile is not excluded in *A. excelsa*. However, it is also quite likely that these compounds support many other important mechanisms which deal with the primary metabolism of plants (Chen et al., 2006; Erazo et al., 2006; De Carvalho et al., 2006).

In addition to human, plants produce volatile compounds known to influence insect preferences for oviposition and feeding (Floate et al., 1993; Seidelmann et al., 2003). 2-Phenylacetonitrile is a highly volatile compound found in most plant species (Julkunen-Titto, 1986). Its primary function is assumed to be stimulus for pollination, chemical ecology, as a sex-attractant and to influence insect behaviour (Floate et al., 1993; Seidelmann et al., 2003). It is also assumed that phenylacetonitrile from *A. excelsa* has a similar biological function which appears to influence insect preference as an entomological pollinating agent.

The phenylacetonitrile, in addition, appears to provide some stimuli which could enhance sexual behaviours of courting insects, which could then be attracted to the *A. excelsa* plant (Floate et al., 1993; Seidelmann et al., 2003).

Conclusion

The volatile constituent has for the first time been established in *A. excelsa* and holds interesting economic value. Phenylacetonitrile, carvone and limonene are important in current medical cures. These results could assist in ascertaining the corrective use of the aloe products in the commercial environment.

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