Chemical Composition of Some Novel Aromatic Oils from the Australian Flora

I.A. Southwell, M.F. Russell and R.L Smith
Wollongbar Agricultural Institute,
Wollongbar, NSW 2477, Australia

Keywords: essential oils, *Backhousia citriodora*, lemon myrtle, *B. anisata*, aniseed myrtle, *Darwinia citriodora*, lemon-scented myrtle, Myrtaceae, *Acacia nuperrima* ssp. *cassitera*, Table Top Acacia, Mimosaceae, *Prostanthera staurophylla*, New England mintbush, Lamiaceae.

Abstract

The leaves of *Backhousia citriodora, B. anisata, Darwinia citriodora, Acacia nuperrima* ssp. *cassitera* and *Prostanthera staurophylla* were steam distilled and their essential oils isolated and examined by GC and GCMS to determine their chemical composition. *Backhousia citriodora* and *B. anisata* were confirmed as being rich in citral and anethole respectively with rare chemical varieties of each rich in citronellal and methyl chavicol. The chemical and physical characteristics of these oils indicated that these species were ideal alternative sources of citral, citronellal, anethole and methyl chavicol type oils. Similar analysis of the previously unexamined *Darwinia citriodora, Acacia nuperrima* ssp. *cassitera and Prostanthera staurophylla* showed that these species were potential sources of methyl myrtenate and methyl geranate, kessane and the rare α-phellandren-8-ol respectively.

INTRODUCTION

The indigenous Australian flora contains an abundance of essential oil bearing taxa with commercially useful and structurally interesting constituents (Lassak and Southwell, 1977; Southwell, 1987; Southwell and Brophy, 2000). Species recently re-investigated include *Backhousia citriodora* (Southwell et al., 2000) and *Backhousia anisata* (Blewitt and Southwell, 2000) along with the newly investigated *Darwinia citriodora* (Southwell et al., 2001a), *Acacia nuperrima* ssp. *cassitera* (Southwell, 2000) and *Prostanthera staurophylla* (Southwell et al., 2001b).

Backhousia citriodora (Fig. 1; Family:Myrtaceae; Common Name:Lemon Myrtle) found in the rainforests of coastal Queensland from around Brisbane to Mackay, is a popular plant in cultivation reaching up to 20 metres in height with white flowers appearing in summer through to autumn and occurring in clusters at the ends of the branches. Although the flowers are attractive, the plant is grown more for the lemon fragrance of the foliage. When crushed, the leaves emit a very strong aroma which would rival any member of the citrus family. *Backhousia anisata* (Fig. 4; Family:Myrtaceae; Common Name:Aniseed Myrtle) is native to the Bellingen Valley region of the NSW mid-north coast and grows in medium to heavy soils in semi-shaded riparian positions to about a height of 20 m with a spread of 4 m. *Darwinia citriodora* (Fig. 7; Family:Myrtaceae; Common Name:Lemon-scented Myrtle) is an evergreen shrub native to Western Australia growing in light, moist soils and open semi-shaded positions growing to 1.2 m with a spread of 1 m. *Acacia nuperrima* ssp. *cassitera* (Fig. 10; Family:Mimosaceae; Common Name:Table Top Acacia) is a dwarf shrub with showy small yellow wattle flowers growing in stoney range regions of north Queensland. *Prostanthera* is a genus of about 90 species which occur only in Australia. They are known generally as "mint bushes" because of the aromatic foliage of many species. As a genus of the Lamiaceae, *Prostanthera* is related to a number of culinary herbs such as mint, thyme, oregano and sage. *Prostanthera teretifolia* (Fig. 13) and *P. staurophylla* both grow on granite outcrops in a restricted location in the New England area of New South Wales, and are usually small shrubs up to 1-2 metres high. Leaves are linear and rounded in cross-section and up to 15mm long. They are grey-green in colour and highly aromatic.

Proc. Int. Conf. on MAP
Eds. J. Bernáth et al.
Acta Hort. 597, ISHS 2003

The spring flowers are usually deep purple (sometimes mauve). Some botanists consider that *P. teretifolia* and *P. staurophylla* should be regarded as a single species.

This paper describes the chemical composition of the steam volatile oil of the leaves and terminal branchlets of each species and compares each oil with similar commercial oils or indicates the potential as a source of a novel isolate.

MATERIALS AND METHODS

Plant Material

The *Backhousia* samples were provided as leaf or oil from established or nearly established commercial plantations. *Darwinia citriodora* leaf material was collected from specimens cultivated on the east coast of Australia (Southwell et al., 2001a), *Acacia nuperrima* ssp. *cassitera* was collected from the Dimbulah region of north Queensland (Southwell, 2000) and *Prostanthera staurophylla* from Torrington in the New England Tablelands of New South Wales (Southwell et al., 2001b).

Oil Extraction

Oil samples were steam distilled by Clevenger-type laboratory-scale (500-1000g fresh weight, all species), pilot-scale (15 kg, *Backhousia citriodora*) and commercial-scale (1 tonne, *Backhousia* only) procedures.

Oil Analysis

The leaf oils were analysed and constituents quantified using a Hewlett Packard 5890 gas chromatograph, 3393A integrator, 7673A autosampler and an Alltech AT 35 60 m x 0.25 mm, 0.2 µm film thickness, mid polarity FSOT column with hydrogen (45 cms/sec) as carrier gas, injection port (split 1:50) at 250°C, flame ionisation detector at 300°C and temperature programming from 60°C (1 minute) to 250°C at 10°C/min. Integration percentages were determined by area normalisation of total FID response from the injection of a 1% solution of oil in ethanol. GC/MS investigations were performed similarly using a Hewlett Packard 6890 instrument fitted with an HP5-MS 30.3 m x 0.25 mm, 0.25 µm film thickness, FSOT column with helium (36 cm/sec) as carrier gas, injection port (split 1:50) at 250°C, mass selective detector (HP 5973) at 250°C (source) and 150°C (quad) with transfer line 280°C and ion source filament voltage of 69.9eV. Retention indices were measured with respect to n-alkane standards. Component identification was made on the basis of mass spectral fragmentation, retention time comparison with authentic constituents and mass spectral and retention matching with commercial (NIST, Wiley and Adams) libraries. Assignments were often confirmed by concentration enhancement by elution from an alumina column with diethyl ether in hexane and by measuring the infrared absorption of the liquid film on a Perkin Elmer 16PC FTIR. Proton and carbon magnetic resonance investigations were based on either (A)1D ^{1}H and J-Modulated ^{13}C spectra and 2D COSY-45 and HMBC correlations using a $CDCl_3$ solution at 28°C on a Bruker Avance DRX-500 Spectrometer or (B) DEPT, gCOSYDQF, gHMBC and gHMQC techniques on a Bruker Avance 300 Spectrometer using an inverse 1H/BB 5mm probe with z-gradient.

RESULTS

Backhousia citriodora, Lemon Myrtle

The leaf essential oil of *Backhousia citriodora* (Fig. 1) was found to contain myrcene (0.1-0.7%), 6-methyl-5-hepten-2-one (0.1-2.5%), linalool (0.3-1.0%), citronellal (0.1-0.9%), *iso*-neral (0.6-2.7%), *iso*-geranial (1.0-4.2%), neral (32.0-40.9%) and geranial (46.1-60.7%) (Southwell et al., 2000) (Fig. 2). Small quantities of this oil, which is richer in citral than both lemongrass *(Cymbopogon flexuosus* and *C. citratus)* and *Litsea cubeba*, are now in commercial production. The citronellal chemotype of *B. citriodora* (Fig. 3) is quite rare.

***Backhousia anisata,* Aniseed Myrtle**

The leaf oil from *Backhousia anisata* plantings (Fig. 4) was found to be rich in either E-anethole (Fig. 5) or methyl chavicol (Fig. 6) providing an excellent alternate source to anise seed, star anise and fennel for E-anethole and to basil for methyl chavicol (Blewitt and Southwell, 2000). Sample quantities are being produced.

***Darwinia citriodora,* Lemon-Scented Myrtle**

The leaf essential oil of *Darwinia citriodora* (Fig. 7) indicated two chemical forms yielding from 0.5 to 1.1% oil, respectively, rich in either methyl myrtenate (56-76%) (Fig. 8) or an approximate 50/50 mix of methyl myrtenate and methyl geranate (49-75%) (Fig. 9) (Southwell et al., 2001a). Some samples of this oil also contain small amounts of the pungent isovaleraldehyde detracting from its value as a fragrance.

***Acacia nuperrima* ssp. *cassitera,* Tabletop Acacia**

The leaf oil of *Acacia nuperrima* ssp. *cassitera* (Mimosaceae) (Fig. 10) also indicated two chemotypes yielding 0.6% and 0.3%, rich in kessane (88.8%) (Fig. 11) and α-pinene (16.2%) (Fig. 12), respectively (Southwell, 2000).

***Prostanthera staurophylla,* New England Mintbush**

The leaf of *Prostanthera staurophylla* (Lamiaceae, closely related to *P. teretifolia*, Fig. 13) yielded 2.1% (fresh weight) of an oil rich in terpenoids including the rare α-phellandren-8-ol (5.7%). Other significant contributors to the mint-like aroma were found to be limonene (26.9%), α-pinene (10.7%), verbenone (8.4%), camphor (3.9%) and chrysanthenone (2.1%) (Fig. 14) (Southwell et al., 2001b).

DISCUSSION

As the market demand for dried leaves of *B. anisata* and *B. citriodora* can not keep up with the production of these culinary herbs, producers are exploring the market potential of the essential oils. These investigations have shown that both oils are equivalent or even better than the traditional sources of anethole and citral respectively. *B. citriodora* oil analysis, for example, indicated a consistently higher citral content than lemon grass and *Litsea cubeba*. This was reflected in the fragrance which could be described as having a finer citral note than lemon grass and *Litsea cubeba* possibly due to the higher proportion of *exo*-, *cis*-, and *trans*-isocitrals (Fig. 2). Similarly, *B.anisata* provides an excellent alternative source for aniseed flavours.

One chemical variety of *Darwinia citriodora* provides the world's best source of natural methyl myrtenate while the other variety is a source of the methyl myrtenate/methyl geranate ester mixture. With the removal of any traces of isovaleraldehyde, these oils have the potential to provide a good source of these fragrant esters. *Acacia nuperrima* ssp. *cassitera* is shown, by this investigation, to be the best source of kessane, surpassing *Prostanthera* sp. aff. *ovalifoli*a, the best source previously known (Southwell and Tucker, 1993). The oil from *Prostanthera staurophylla*, rich in the ketones verbenone, camphor and chrysanthenone has a typically strong ketonic-mint aroma with potential for use as a flavour, fragrance or aromatherapy oil.

ACKNOWLEDGEMENTS

We thank Peter G. Waterman, Centre for Phytochemistry, Southern Cross University, Lismore and David J. Tucker, Dept. Chemistry, University of New England, Armidale for NMR spectra, Joseph J. Brophy, School of Chemistry, University of New South Wales, Sydney, for mass spectra, Maree Blewitt, PO Box 24, Bellingen, Dennis W. Archer, Toona Essential Oils, Gympie and Athol Durre, Dimbulah, for leaf and oil samples and Robert Lowe and Warwick Press, Wollongbar Agricultural Institute, Wollongbar for preparation of this paper/poster. We are also indebted to the Rural Industries Research and Development Corporation (RIRDC), Canberra and Southern Cross Botanicals Pty Ltd, Lennox Head for funding the presentation.

Literature Cited

Blewitt, M. and Southwell, I.A. 2000. *Backhousia anisata* Vickery, an alternative source of (E)-anethole. J. Essential Oil Research 12:445-454.

Lassak, E.V. and Southwell, I.A. 1977. Essential oil isolates from the Australian flora. Part I. Int. Flav. Food Add. 1977: 126-132.

Southwell I.A. and Brophy J.J., 2000. Essential oil isolates from the Australian Flora. Part III. J. Essential Oil Research 12:267-278.

Southwell, I.A. 1987. Essential oil isolates from the Australian flora. Part II. Flav. Fragrance J. 2:21-27.

Southwell, I.A. and Tucker, D.J. 1993. *cis*-Dihydroagarofuran from *Prostanthera* sp. aff. *ovalifolia.* Phytochemistry 33:857-862.

Southwell, I.A., 2000. *Acacia nuperrima* ssp. *cassitera*, a new source of kessane. J. of Essential Oil Research 12:566-568.

Southwell I.A., Russell M., Smith R.L. and Archer D.W., 2000. *Backhousia citriodora* F. Muell. (Myrtaceae), a superior source of citral. J. Essential Oil Research 12: 735-741.

Southwell, I.A., Brophy, J.J. and Tucker, D.J. 2001a. *Darwinia citriodora* (Myrtaceae), a new source of methyl myrtenate and methyl geranate. J. of Essential Oil Research 13:58-60.

Southwell, I.A., Russell, M. and Waterman, P.G. 2001b. α-Phellandren-8-ol, a rare monoterpene alcohol from the essential oil of *Prostanthera staurophylla* F. Muell. J. of Essential Oil Research 13: in press.

Figures

Fig. 1. *Backhousia citriodora* flowering stem

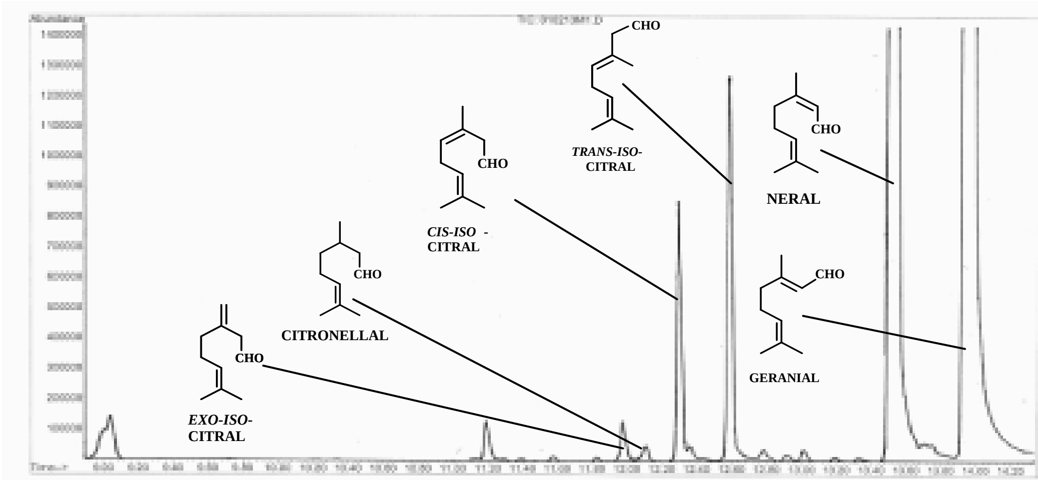

Fig. 2. Gas chromatogram of oil from the citral chemotype of *Backhousia citriodora*.

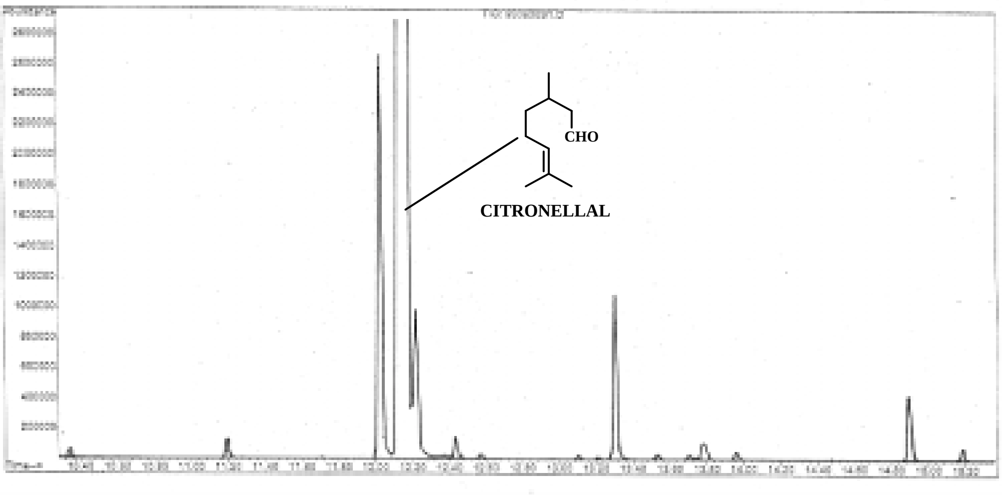

Fig. 3. Gas chromatogram of oil from the citronellal chemotype of *Backhousia citriodora*.

Fig. 4. *Backhousia anisata* plantation.

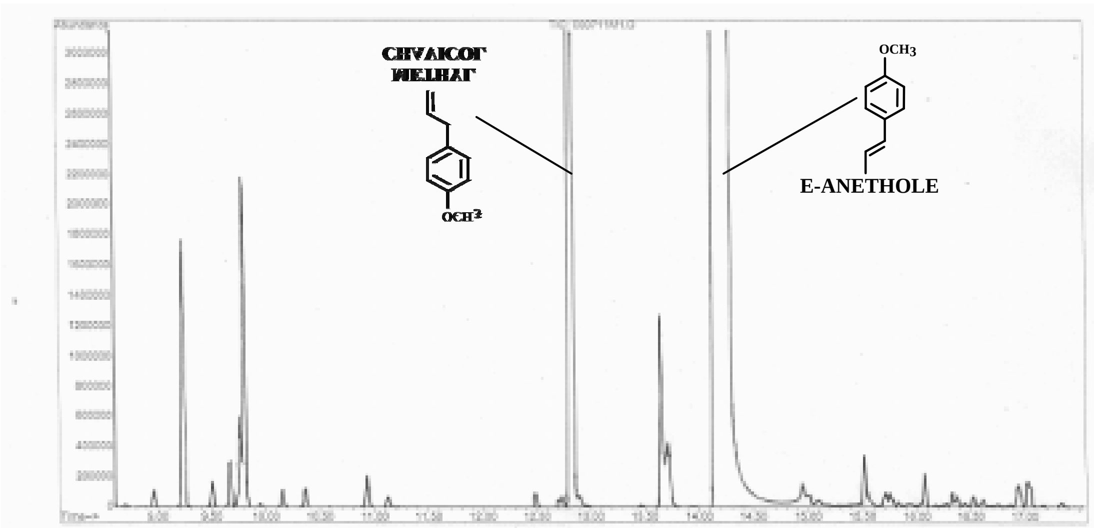

Fig. 5. Gas chromatogram of oil from the anethole chemotype of *Backhousia anisata*.

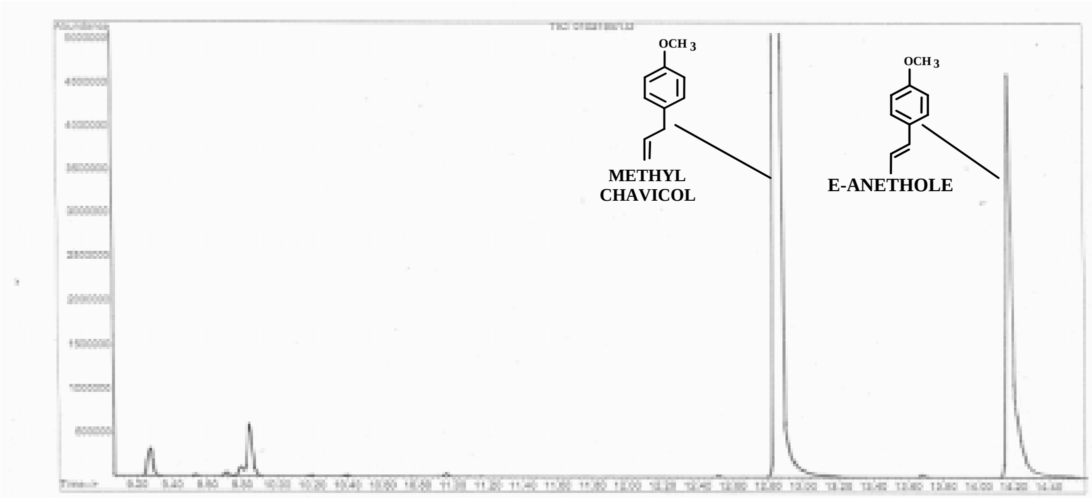

Fig. 6. Gas chromatogram of oil from the methyl chavicol chemotype of *Backhousia anisata*.

Fig. 7. *Darwinia citriodora* flowering shrub

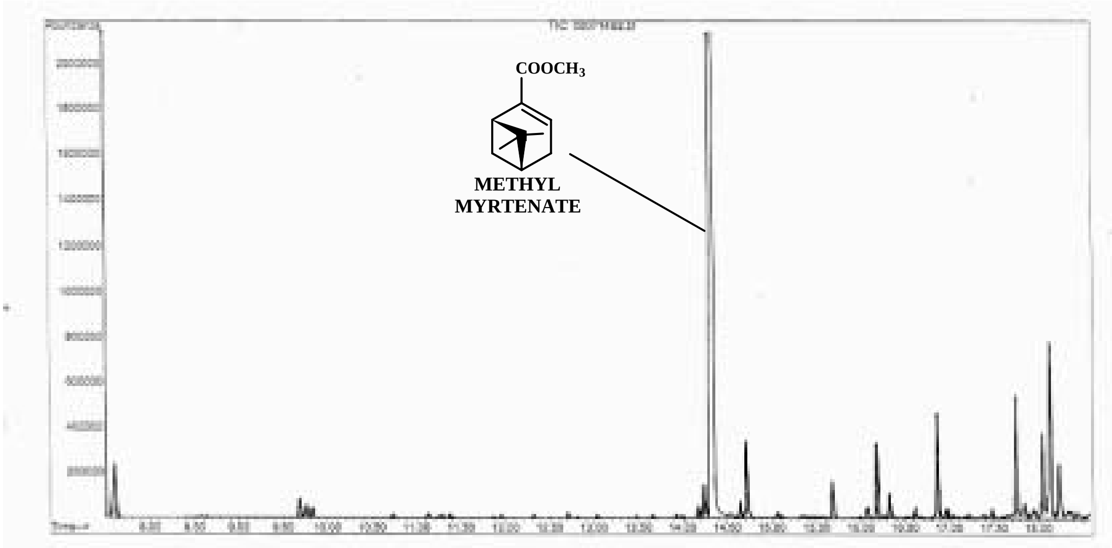

Fig. 8. Gas chromatogram of oil from the methyl myrtenate chemotype of *Darwinia citriodora*.

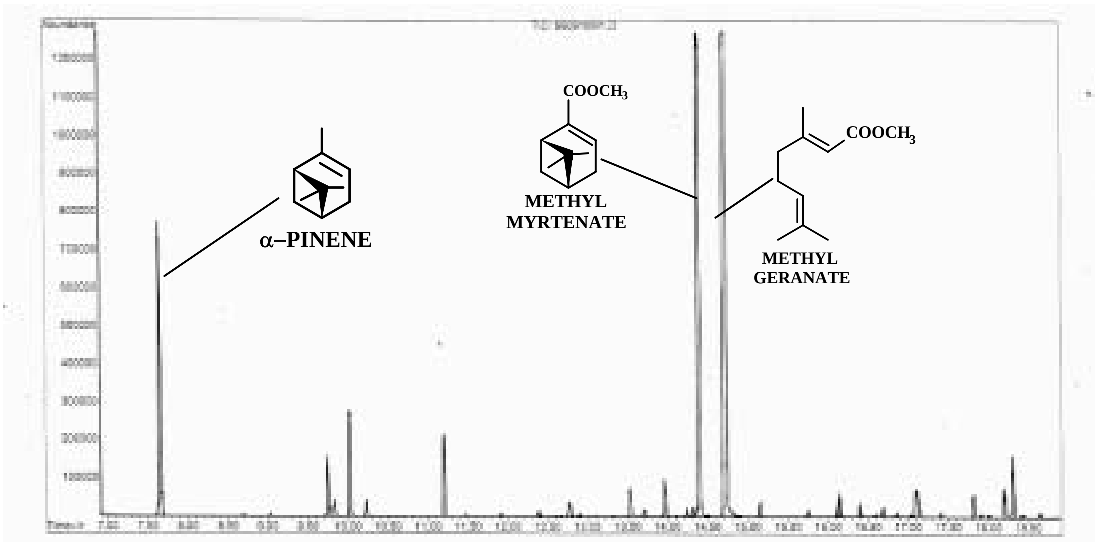

Fig. 9. Gas chromatogram of oil from the methyl myrtenate/methyl geranate chemotype of *Darwinia citriodora*.

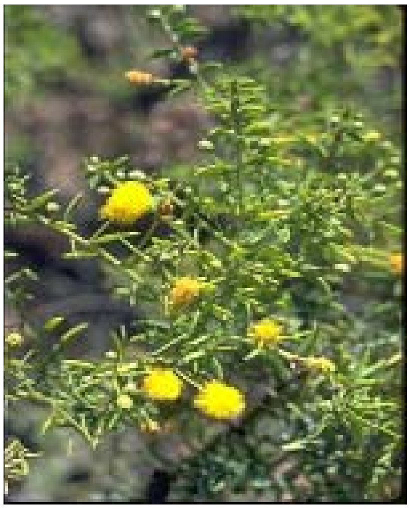

Fig. 10. *Acacia nuperrima* ssp. *cassitera* flowering shrub

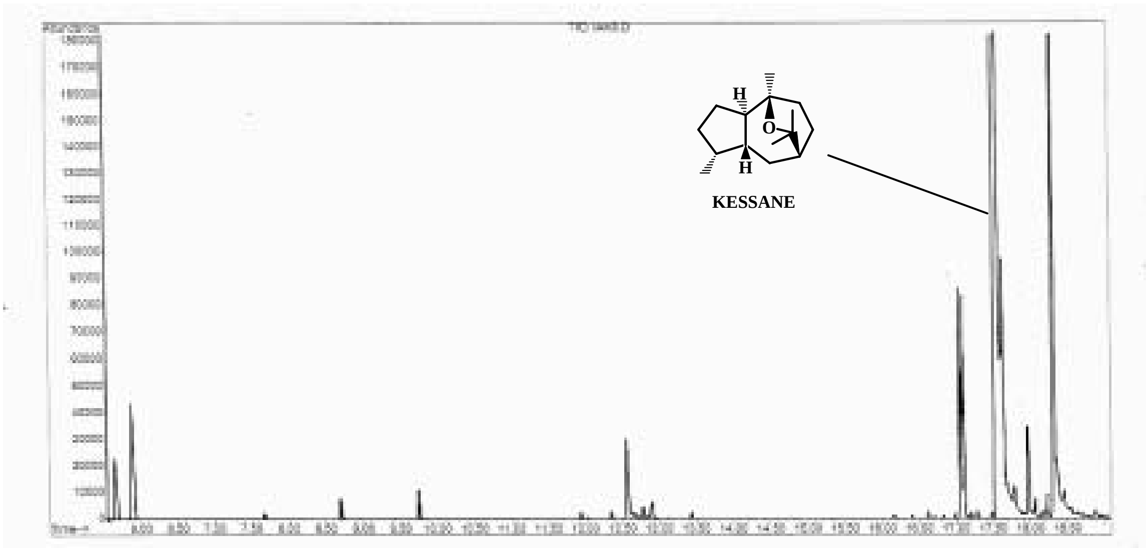

Fig. 11. Gas chromatogram of oil from the kessane chemotype of *Acacia nuperrima* ssp. *cassitera*.

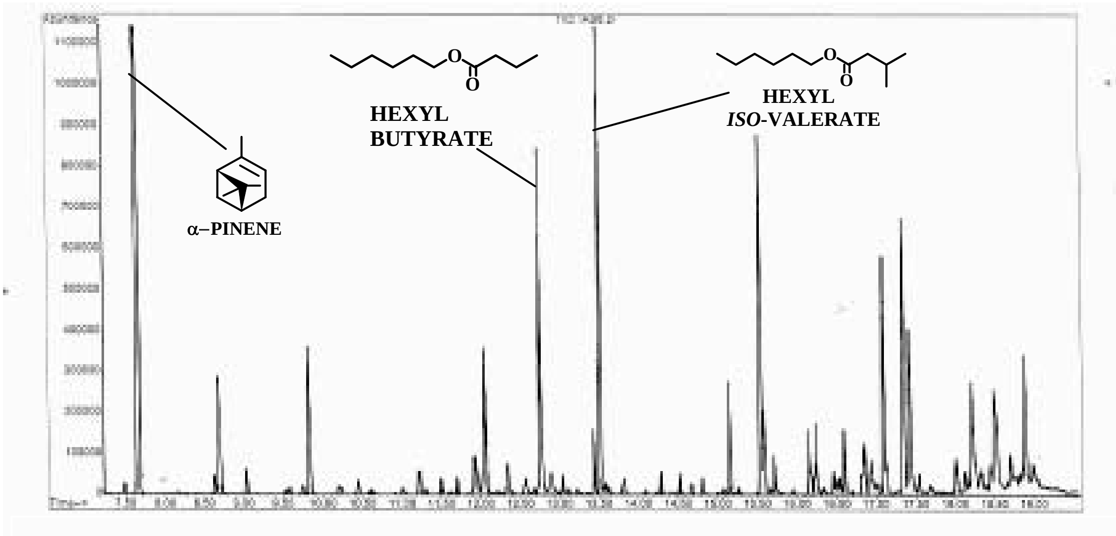

Fig. 12. Gas chromatogram of oil from the α-pinene chemotype of *Acacia nuperrima* ssp. *cassitera*.

Fig. 13. *Prostanthera teretifolia* flowering shrub

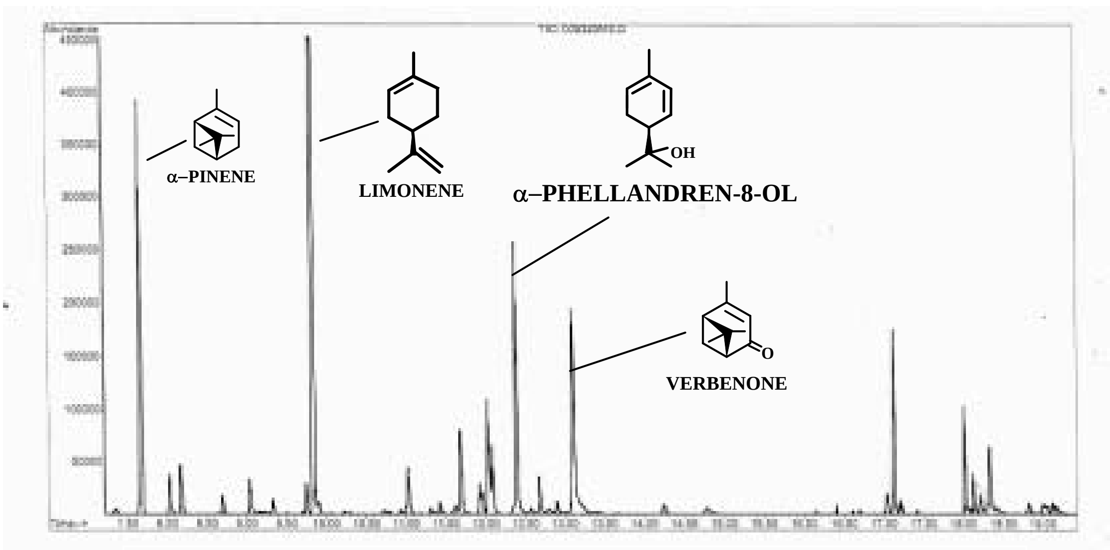

Fig. 14. Gas chromatogram of oil from *Prostanthera staurophylla*