The combination of gas chromatography–olfactometry and multidimensional gas chromatography for the characterisation of essential oils

Graham Eyres\textsuperscript{a}, Philip J. Marriott\textsuperscript{b}, Jean-Pierre Dufour\textsuperscript{a,*}

\textsuperscript{a} Department of Food Science, University of Otago, P.O. Box 56, Dunedin 9054, New Zealand
\textsuperscript{b} Australian Centre for Research on Separation Science, Department of Applied Chemistry, RMIT University, G.P.O. Box 2476V, Melbourne, Vic. 3001, Australia

Available online 1 August 2006

Abstract

A research area of great interest to the flavour industry is the analysis of odour active compounds in essential oils. In this paper, a methodology is presented for the identification of character-impact odorants in essential oil samples using (a) gas chromatography–olfactometry (GC–O); (b) comprehensive two-dimensional gas chromatography (GC × GC) combined to time-of-flight mass spectrometry (TOFMS) and (c) heart-cut multidimensional gas chromatography–olfactometry (MDGC–O). The specific advantages and limitations of each technique are discussed. The advantage of combining these techniques in a strategy to identify character-impact odorants is demonstrated using examples from coriander leaf (\textit{Coriandrum sativum}) and hop (\textit{Humulus lupulus}) essential oils. In particular, resolution of co-eluting regions of compounds and evaluation of their individual odour activity is discussed. In coriander leaf, only \textit{E}-2-dodecenal was found to contribute to a co-eluting odour region, \textit{E}-2-dodecen-1-ol and 1-dodecanol being present below detection threshold. Using MDGC on a hop essential oil sample, eight significant peaks were resolved from an 18 s heart-cut where a potent odorant was perceived during GC–O.

© 2006 Elsevier B.V. All rights reserved.

Keywords: Methodology; Gas chromatography–olfactometry; Multidimensional gas chromatography; GC × GC–TOFMS; Deans switch; Character-impact odorants

1. Introduction

The identification of compounds responsible for ‘character-impact odorants’ in essential oils is of particular interest to the perfume and flavour industry. These compounds may be defined as odour active compounds that are responsible for (or significantly contribute to) the distinctive odour profile of a sample. It is often the case that a single compound is predominantly responsible for the odour of a sample, such as eugenol in clove essential oil (\textit{Syzygium aromaticum} or \textit{Eugenia caryophyllata}). In other samples, such as hop essential oil (\textit{H. lupulus}), the characteristic odour is due to the perception of complex mixtures of volatile compounds [1,2].

A majority of research on characterising essential oils is based on chemical composition using a chemical detector. However, it is well established that the odour thresholds of volatile compounds can differ by many orders of magnitude (e.g., parts per trillion up to odourless compounds) [3,4]. The relationship between concentration and odour intensity may also vary considerably between compounds. Because of the large variation in these two properties, the response of a chemical GC detector (e.g., FID or MS) is not representative of odour activity. For example, the most abundant compound in a chromatogram may not be the most important odorant [5]. Consequently, the impact of a compound on the odour of a sample must be evaluated using human assessors. A valuable tool for identifying character-impact odorants is gas chromatography–olfactometry (GC–O), where human ‘sniffers’ are used to detect and evaluate volatile compounds as they elute from a GC column [3].

Several different GC–O methodologies have been developed to evaluate the relative importance of odour active compounds in a sample [3,6,7]. In CharmAnalysistm, a dilution series is prepared and each dilution is assessed by GC–O until no odours are perceived [8–10]. The results quantify the odour potency of a compound, which is based upon the ratio of its concentration to its odour threshold in air [8].
A limitation of conventional GC–O is that it is performed using a single column where co-elution of peaks is likely to occur, making identification of the compound responsible for a perceived odour difficult or uncertain. For example, co-elution may result in multiple possibilities for which compound is responsible for an odour. There is also the possibility to make an incorrect identification if a trace odorant is masked by a large odourless compound. Alternatively, co-elution of more than one odor active compound during GC–O analysis may result in a mixed odour perception; this situation is known as an ‘odour cluster’ [5]. One possible solution for identifying character-impact odorants where co-elution occurs, is to use comprehensive two-dimensional gas chromatography (GC × GC). Hyphenating GC × GC to time-of-flight mass spectrometry (TOFMS) presents researchers with a very powerful identification tool. For more information on the development and operation of GC × GC and TOFMS, the reader is directed to several comprehensive reviews [11–13].

The specific objective of this research is to develop a methodology to identify the compounds responsible for character-impact odorants. This will be demonstrated using two examples of essential oils: coriander leaf (C. sativum), also known as cilantro; and hops (H. lupulus), which are used to impart flavour and aroma to beer.

2. Experimental

2.1. Materials

The sample of coriander leaf essential oil was provided by the Department of Chemistry, University of the South Pacific, Suva, Republic of the Fiji Islands. Sample preparation has been described previously [5]. Commercial samples of the ‘spicy’ fraction of hop essential oil from Target and Cascade varieties were obtained from Botanix Ltd. (Paddock Wood, Kent, UK). The production process involves extraction of hop pellets using liquid CO2 followed by isolation of the whole essential oil using molecular distillation under high vacuum. Fractionation is then achieved using a combination of distillation and chromatographic methods [14]. The resulting spicy fraction is rich in monoterpenes and sesquiterpenes alcohols and has previously been described as having a sandalwood or oakmoss character [15].

Dilutions were prepared using cyclohexane (pesticide analysis grade, 99.7%; BDH Laboratory Supplies, Poole, Dorset, UK) for GC–O, GC × GC–FID and heart-cut multidimensional gas chromatography–olfactometry (MDGC–O) analysis or n-hexane (Pestana™, ≥95%; Riedel-de Haën, Sigma–Aldrich Co., St. Louis, MO, USA) for GC × GC–TOFMS analysis.

Reference compounds of E-2-dodecenal (≥97%) and E-2-dodecen-1-ol (≥97%) were purchased from Fluka Chemie GmbH (Sigma–Aldrich Co., Buchs, Switzerland); 1-heptanol and 1-dodecanol (≥99%) were purchased from Nu-Chek-Prep Inc. (Elysian, MN, USA); and methane gas (UHP grade, ≥99.9%) was supplied by BOC Gases (Auckland, New Zealand).

2.2. Gas chromatography–olfactometry and CharmAnalysis™

GC–olfactometry analyses were performed using a HP5890 Series II plus gas chromatograph (Hewlett Packard, Avondale, PA, USA) connected to a GC–O port (Datu Technology, Geneva, NY, USA).

The separation was carried out on a 25 m × 0.32 mm I.D. × 0.5 μm dI BPX5 capillary column (SGE International, Ringwood, Australia). Helium was used as the carrier gas with a constant flow of 2 mL min⁻¹ with an initial column head pressure of 59 kPa at 60 °C. Sample volumes of 1 μL were manually injected using a 5 μL syringe (SGE International). The injector was operated with a split ratio of 50:1 at 220 °C. The GC oven was temperature programmed from 60 to 210 °C at a rate of 6 °C min⁻¹ then increased to 290 °C at 10 °C min⁻¹ and held for 20 min to ensure the column was completely clean. Each GC–O sniff run had a maximum duration of 25 min to ensure the assessors did not suffer fatigue [10].

The olfactory detector was maintained at 300 °C for GC–O analysis. As compounds eluted from the column they were presented to the assessor in a stream of humidified air at 50 °C to be evaluated. The assessor recorded the duration and the description of an odour and the data was collected using Charmware™ software (version 1.08, Datu Technology). A series of alkanes (C8–C26) was run using a FID to establish retention indices for olfactometry analysis.

GC–O analyses were performed by two experienced assessors on a series of dilutions starting with a 10% (v/v) sample of essential oil in cyclohexane. Serial dilutions (factor of two) were sequentially assessed until no odours were detected. CharmAnalysis™ was performed by integrating the GC–O results of each dilution using the Charm calculations described elsewhere [9,10].

2.3. Comprehensive two-dimensional gas chromatography (GC × GC)

GC × GC separations were performed using two Agilent 6890A gas chromatographs, the first (Agilent Technologies, Little Falls, DE, USA) equipped with a FID and the second (Agilent Technologies, Palo Alto, CA, USA) coupled to a Pegasus III time-of-flight mass spectrometer (TOFMS; LECO Corp., St. Joseph, MI, USA). Both instruments were retrofitted with an Everest model Longitudinally Modulated Cryogenic System (LMCS; Chromatography Concepts, Doncaster, Australia). The operation of the LMCS has been described elsewhere [16].

A series of alkanes (C8–C22) were analysed to establish first dimension retention indices (tI) for GC × GC in both systems. These GC × GC retention indices were matched to GC–O retention indices in order to locate odour active regions of interest and identify potential compounds responsible.

2.3.1. GC × GC-FID

The column set consisted of a 30 m × 0.25 mm I.D. × 0.25 μm dI BPX5 (SGE International) primary column (1D) coupled in series to a 1.1 m × 0.1 mm I.D. × 0.1 μm dI BP20 (SGE
International) second dimension column (2D). Unless otherwise stated the experimental conditions were as follows: the oven was programmed from 60 to 210 °C at 3 °C min⁻¹ followed by an increase to 240 °C at 10 °C min⁻¹ and held for 10 min to ensure all compounds eluted. Sample injections of 1 μL, at 10% and 1% (v/v) essential oil in cyclohexane, were made using an Agilent 7863 Series auto-sampler. The injector was operated at 220 °C with a split ratio of 50:1. Hydrogen was used as a carrier gas at a constant flow of 2 mL min⁻¹ with an initial column head pressure of 178.4 kPa at 60 °C. Average linear velocity of the carrier gas was determined using methane and was found to be 34.4 cm s⁻¹ at 60 °C. The FID was operated at 260 °C at an acquisition rate of 100 Hz. The modulation period was set at 5 s and the cryogenic trap was maintained at −15 °C for the duration of each analysis. Conventional 1DGC-FID chromatograms were generated using the same system and experimental conditions except with the LMCS modulator turned off.

2.3.2. GC × GC–TOFMS

The column set consisted of a 30 m × 0.25 mm I.D. × 0.25 μm d₁ BPX5 (SGE International) primary column (1D) coupled in series to a 0.8 m × 0.1 mm I.D. × 0.1 μm d₂ BP20 (SGE International) second dimension column (2D). The oven was held at an initial temperature of 60 °C for 0.2 min before increasing the temperature to 186 °C at 3 °C min⁻¹ followed by an increase to 240 °C at 10 °C min⁻¹ and held for 5 min to ensure all compounds eluted. Sample injections of 1 μL, at 10% and 1% (v/v) essential oil in n-hexane, were made using an Agilent 7683 Series auto-sampler. The injector was operated at 250 °C with a split ratio of 50:1. Hydrogen was used as a carrier gas at a constant flow of 2 mL min⁻¹ with an initial column head pressure of 288.2 kPa at 60 °C. Average linear velocity was determined using butane gas and was found to be 36.3 cm s⁻¹ at 60 °C. The modulation period was set at 5 s and the cryogenic trap was maintained at −15 °C for the duration of each analysis.

The 2D column outlet was coupled to the TOFMS via a 26 cm uncoated transfer tube maintained at 250 °C. The TOFMS filament was switch on to acquire data after a solvent delay of 2 min. The TOFMS had a source temperature of 200 °C and was set to acquire ions from m/z 41 to 415 at a rate of 100 spectra s⁻¹ with a detector voltage of 1560 V.

2.4. Heart-cut multidimensional gas chromatography–olfactometry (MDGC–O)

The MDGC separations were performed on two capillary columns with different stationary phases (1D; 2D) in a single Agilent 6890N GC oven (Agilent Technologies, Shanghai, China) (Fig. 1). An Agilent G2855A Deans switch interface (DS) controlled whether the flow from the 1D column was directed to either: (i) FID 1 via a deactivated fused silica transfer line (TL), or (ii) the 2D column. Additional carrier gas was supplied to the Deans switch to provide the switching flow using an auxiliary electronic pressure control (EPC) module. Using the Deans switch, discrete regions of eluate can be selectively ‘heart-cut’ from the first to the second column to resolve co-eluting regions.

The design of the Deans switch interface and principle of operation are published elsewhere [17–19].

The outlet of the 2D column was either connected to FID 2 or hyphenated to a sniff port (ODO II; SGE International) in order to evaluate the odour activity of resolved compounds. Alternatively, the column eluate was split (1:1) using a press-fit Y-connector (Phenomenex, Torrance, CA, USA). An LMCS cryotrap (CT) (Chromatography Concepts) was also placed at the start of the 2D column to provide cryofocussing and added flexibility. This will be discussed further in Section 3.

Initial experimental conditions were selected using the Deans switch calculator software provided (version A.01.01; Agilent Technologies). The inlet and auxiliary EPC pressures were then fine-tuned using methane and 1-heptanol injections according to the original method proposed by Deans [17]. Fine tuning ensures that pneumatic switching is performed efficiently, i.e. 100% of the 1D eluate is switched to either FID 1 or the 2D column with no losses. The dimensions of the transfer line to FID 1 are calculated so that the pneumatic resistance matches that of the 2D column. This also ensures efficient operation of the Deans switch. The MDGC system was operated under constant pressure mode to maintain the pressure balance between the two columns throughout the oven temperature program.

2.4.1. MDGC conditions for coriander leaf essential oil

The first column (1D) was a 30 m × 0.32 mm I.D. × 0.25 μm d₁ HP5 (J&W Scientific, Agilent Technologies, Palo Alto, CA, USA) and the second column (2D) was a 30 m × 0.32 mm I.D. × 0.5 μm d₂ Solgel Wax (SGE International). The transfer line to FID 1 was 1.48 m × 0.15 mm I.D. deactivated fused silica (SGE International). The oven was programmed from 60 to 210 °C at 6 °C min⁻¹ followed by an increase to 280 °C at 10 °C min⁻¹ and held for 10 min to ensure all compounds eluted. Sample injections of 1 μL, at 1% (v/v) essential oil in cyclohexane, were made using an Agilent 7683 Series auto-sampler. The injector was operated at 240 °C with a split ratio of 20:1. Hydrogen was used as a carrier gas with a constant inlet pressure of 91.3 kPa and a constant auxiliary pressure of 56.5 kPa. These conditions gave a 1D flow rate of 2.5 mL min⁻¹ (38.4 cm s⁻¹) and a 2D flow rate of 3.75 mL min⁻¹ (56.5 cm s⁻¹) at 60 °C. The 2D column was connected directly to either FID 2 or the sniff port without splitting the eluate. Both detectors (FID 1; FID 2) were operated at 290 °C with an acquisition rate of 20 Hz.
2.4.2. MDGC conditions for the spicy fraction of Target hops

The first column (1D) was a 30 m × 0.32 mm I.D. × 0.25 μm d<sub>f</sub> HP5 (J&W Scientific) and the second column (2D) was a 30 m × 0.25 mm I.D. × 0.25 μm d<sub>f</sub> Solgel Wax (SGE International). The transfer line to FID 1 was 0.92 m × 0.1 mm I.D. deactivated fused silica (SGE International). The oven was programmed from 60 to 210 °C at 3 °C min<sup>−1</sup> followed by an increase to 280 °C at 10 °C min<sup>−1</sup> and held for 10 min to ensure all compounds eluted. Sample injections of 1 μL, at 10% (v/v) essential oil in cyclohexane, were made using an Agilent 7683 Series autoSampler. The injector was operated at 250 °C with a split ratio of 50:1. Hydrogen was used as a carrier gas with a constant inlet pressure of 163.7 kPa and a constant auxiliary pressure of 135.8 kPa. These conditions gave a 1D flow rate of 2.4 mL min<sup>−1</sup> (31.1 cm s<sup>−1</sup>) and a 2D flow rate of 3.6 mL min<sup>−1</sup> (51.7 cm s<sup>−1</sup>) at 60 °C. The eluate of the 2D column was split (1:1) between FID 2 and the sniff port. Both detectors (FID 1; FID 2) were operated at 290 °C with an acquisition rate of 20 Hz.

3. Results and discussion

The requirement for two-dimensional separations is well documented [12, 20–22], as the theoretical peak capacity of a single 50 m column is only 260 peaks [23]. In addition, peaks are neither evenly nor randomly distributed in a chromatogram because compounds often demonstrate related chemical properties [12]. For example, sesquiterpenoid compounds are notoriously difficult to resolve because they have the same structural formulae and therefore exhibit very similar interactions with the column stationary phase. Consequently, the peak capacity required for the system needs to be much greater than the actual number of compounds in the sample [23]. The result of these factors is that considerable co-elution of peaks is inevitable in single column (1D) GC analysis, particularly for complex samples.

For hop essential oil, 485 compounds have been identified and reported in the literature [24, 25], and recent research suggests that it may actually contain up to 1000 compounds [24]. Fig. 2A shows a section of a 1DGC separation of the spicy fraction of Cascade hops that exhibits considerable co-elution in a complex region of oxygenated sesquiterpenoid compounds. Fig. 2B demonstrates the superior resolution obtained using GC × GC and better reveals the true complexity of this essential oil sample.

In a recent study on coriander leaf essential oil, 81 compounds were identified with GC × GC–TOFMS representing 99.4% of the total ion count (TIC) [5]. In comparison, only 41 compounds could be identified using 1DGC on two different stationary phases with a quadrupole mass spectrometer. Despite the fact that coriander has a comparatively simple composition and is relatively well characterised, GC × GC–TOFMS resulted in the identification of 41 compounds not previously reported in the literature for coriander leaf [5].

It is indisputable that GC × GC–TOFMS is a powerful analytical technique that allows the detection, resolution and identification of many more peaks than conventional 1DGC. But the question remains as to whether it can be used in isolation to effectively identify compounds responsible for character-impact odorants. The problem is that the detector is not specifically measuring the vital property of odour activity. Unless the detection threshold and function between concentration and odour intensity are known for each compound, a conclusion about their odour impact in the sample cannot be made. It is also impossible to find new odorants using threshold data alone and subsequently important trace odorants, that are present below the analytical detection limit but above sensory threshold, may be overlooked.

So from one perspective, GC × GC–TOFMS actually increases the complexity of the analysis by generating a greater number of possibilities for character-impact odorants that need to be investigated. This actually increases the amount of data processing and research time required.

To solve this problem of too much data, the methodology presented proposes to use GC–O to selectively determine the location of odour active regions in a chromatogram in order to direct and prioritise research efforts (Fig. 3). GC × GC–TOFMS can then be used to resolve and identify compounds eluting in these odour active regions in order to identify the compounds potentially responsible. In the coriander leaf example, β-ionone (0.02% TIC) was not initially identified using GC × GC–TOFMS because it was present below the sensitivity level set for data processing. It was not until a potent odour was perceived during GC–O (12.0% total odour units) that it was identified by specifically searching in the corresponding region [5].

The limitation of conventional GC–O is that it only uses a single GC column and so a significant degree of co-elution is expected to occur due to lack of resolution. Co-eluting compounds in odour active regions generate multiple possibilities that need to be evaluated to determine which compound is...
responsible for an odour perceived during GC–O. This may be achieved by assessing pure reference compounds, but if a standard cannot be obtained, then it must be either synthesised or extracted and purified. This is potentially a very time-consuming process and if the peak responsible for the odour cannot be located or the compounds cannot be identified, then this process is made even more difficult, if not impossible.

During GC–O of the spicy fraction of Target hops, an intense woody, cedarwood odour was perceived (Fig. 4). This coincided with a complex region of the chromatogram where a large number of oxygenated sesquiterpenoid compounds co-elute. Separation with GC × GC demonstrates this complexity and the improved resolution obtained (Fig. 5). At least six compounds were resolved where the woody odour was known to elute. In addition, spectral de-convolution with TOFMS also indicated further co-elution was occurring (data not shown). The oxygenated sesquiterpenoid compounds eluting in this region exhibit very similar mass spectra, making unambiguous identification challenging without reference standards. It is also difficult to source or to synthesise sesquiterpenoid reference compounds in order to test the odour activity of each individually. Thus, it is not known which peak is (or peaks are) responsible for the woody odour.

The problem is that the eluate from GC × GC is not ideal for evaluation using olfactometry. This is because the human nose is a slow ‘detector’ with a breathing cycle of only 3–4 s [3], which is too slow to reliably assess the narrow, rapid eluting peaks produced by GC × GC. For example, the peaks shown in Fig. 5 elute within a two second period with peak widths between 100 and 400 ms. Regular modulation in GC × GC also creates multiple slices for each peak. Therefore, while GC × GC may be able to resolve the co-eluting compounds, it cannot be used to determine which compound is responsible for a perceived odour.

A potential solution is to use a traditional, heart-cut multidimensional GC–olfactometry (MDGC–O) apparatus (Fig. 1). In comparison to comprehensive GC × GC, only a proportion of the column eluate is separated in two dimensions. However, it is able to resolve a number of selected co-eluting odour regions and maintain one discrete peak per compound with a broader peak width, making it very suitable for olfactory assessment [26].

This system was initially used to investigate a co-eluting odour region in the coriander leaf sample. A potent floral, coriander-like odour was perceived in GC–O coinciding with a co-eluting cluster of compounds (Fig. 6). GC × GC achieved baseline resolution of these compounds and TOFMS identified them as E-2-dodecenal, E-2-dodecen-1-ol and 1-dodecanol. But the question of interest is which compound is responsible for the odour perceived. These co-eluting peaks were heart-cut from the first column and cryotrapped before subsequent release and separation on the 2D column in the MDGC–O system. The three compounds were completely resolved on the 2D
column with enough separation to allow evaluation of their odour activity individually in a 1.5 min time-frame (Fig. 7). Only E-2-docenenal was found to contribute to the odour of the cluster, the other compounds being present below detection threshold for the assessor. Fig. 7 also shows that the total analysis time for resolution of these peaks (I∼1480) was only 22 min.

Fig. 8A shows the first column (1D) separation on the MDGC instrument of the same region where the woody odour was perceived in conventional GC–O of the spicy fraction of Target hops (Fig. 4). The shaded area shows the 18 s region that was selectively heart-cut to the 2D column. Fig. 8B demonstrates the resolution achieved for the heart-cut region on the 2D column showing eight significant peaks and a number of minor peaks (inset) co-eluting in this region. A human assessor can comfortably sniff each compound over a 4 min time-frame in order to evaluate the odour activity of each peak individually. The relative ease of this task means that the information can be gathered for every heart-cut peak in a single analytical run. The analysis can then be repeated for confirmation or statistical validation. The peak widths and separation also make the MDGC–O system compatible with more complex olfactometry data acquisition such as odour intensity measurements. In addition, the peak widths are also compatible with simultaneous detection with a quadruple or an ion trap mass spectrometer, potentially a very powerful technique [26–28].

Where several odour active compounds co-elute and contribute to a mixed odour perception in conventional GC–O, the MDGC–O system could be used to determine the odour quality, relative potency (e.g., CharmAnalysis™), or odour intensity of each compound in order to quantify the odour contribution of each compound to a sample.

Regarding the interpretation of olfactometry results, caution should be taken when making conclusions about the importance of an odorant [29], particularly when using only a few assessors. This is because of large variation in sensitivity between individuals in a population. Specific anosmia or hyperosmia may also have a serious impact on results, by either completely miss-
ing or overestimating the importance of an odorant respectively. Therefore, when making an inference about the importance of an odorant to a population, a panel of assessors is required [3,30].

An advantage of MDGC–O is that compounds can be resolved and have their odor activity evaluated without the requirement for identification. Locating the odor active peaks of interest minimizes the required amount of extraction, purification and traditional structure elucidation (e.g., preparative GC, NMR, IR) or organic synthesis. An added advantage of the Deans switch system is the use of the back-flush feature to reverse the carrier gas flow to elute heavy compounds out the injector in order to shorten the analysis time [31,32].

The LMCS at the start of the 2D column provides cryofocussing for heart-cut regions to improve sensitivity and resolution. Although this effect is minimal when using a 30 m 2D column, it is expected that this would have an impact for shorter columns in an optimised system [33]. The LMCS also provides added flexibility allowing heart-cut regions to be withheld in the trap while a prior heart-cut elutes, thus preventing any possible interference. Alternatively, the oven temperature could be reduced to obtain optimal separation conditions for the second column, avoiding the need for a two oven system. An example of the latter application is for MDGC–O analysis of enantiomers, providing a powerful tool for assessment of the odor activity of chiral compounds in a complex mixture without the requirement for prior sample fractionation [26,34].

It could be argued that given the advantages of the MDGC–O system, there is no need for GC × GC–TOFMS. However, the authors contend that these are complementary rather than competing techniques. The primary advantage of GC × GC in this case is the comprehensive separation of the entire sample in two dimensions in a single analysis. The sensitivity of GC × GC is also greater than MDGC due to the greater cryofocussing effect observed after the short, efficient 2D column (e.g., 1 m × 0.1 mm I.D.). Spectral de-convolution using fast TOFMS detection also provides superior identification power. Therefore, GC × GC–TOFMS is invaluable for the methodology by providing a superior analysis of the overall chemical composition of a sample.

4. Conclusions

Complete characterization of complex samples is an inefficient use of research time when only interested in odor active compounds. The current methodology (Fig. 3) proposes to use conventional GC–olfactometry to locate the odor active regions in the chromatogram and generate an odor profile for the whole sample. Quantifying odor potency using CharmAnalysis™ provides a relative order of importance that may be used to prioritise identification efforts. The GC–O results direct the resolution and identification of compounds responsible using GC × GC–TOFMS, minimising the research time required for data processing and identification.

Where co-eluting peaks coincide with odor regions and give multiple possibilities, heart-cut MDGC–O is used to resolve the co-eluting regions and evaluate the odor activity of each compound individually. These results are then used to direct further identification efforts using GC × GC–TOFMS. Final confirmation of the identity of the compound responsible for an odorant can then be achieved using reference compounds.

Acknowledgements

G.E. would like to acknowledge the Tertiary Education Commission, New Zealand, for providing scholarship funding and the New Zealand Vice-Chancellors’ Committee for providing funds from the Claude McCarthy fellowship. G.E. also thanks the ISCC organising committee for sponsoring conference attendance. The authors gratefully acknowledge the ongoing support of LECO Australia.

References


