Use of complementary ionisation techniques for compound identification in GC-MS

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Background
• GC-MS of methoximated trimethylsilyl derivatives is a robust and reproducible method for ID and quantification of many metabolites
• Success of this method is due to the high resolution and peak capacity of capillary GC, and vast libraries of electron impact spectra

Problem 1: overlapping peaks
• Overlapping peaks are unavoidable
• Detection of overlapping peaks requires mass spectra of co-eluting peaks have substantive differences, but this may not be the case (Fig 1 & 2)

Problem 2: metabolite ID
• Many peaks in a GC-MS chromatogram cannot be identified either because there are no similar spectra, or several spectra that give good matches

Problem 3: EI mass spectra lack structural info
• EI ionisation results in extensive fragmentation such that high m/z fragment ions are rare and spectra are dominated by common low m/z fragment ions
• For many compound classes EI spectra lack structural information and cannot distinguish among sub-classes (e.g. Inositol, methylated inositol, cyclohexanepentol, hexoses: Fig 1 & 2)

Is chemical ionisation the solution?
• CI produces spectra that are partially orthogonal to EI

Methods
• Methoximated trimethylsilyl derivatives of 72 standards and aqueous extracts of 10 plant species were analysed by GC-MS
• Ionisation with an open EI source (at source T=250°C) and positive CI with CH₄ (150°C) or NH₃ (180°C) using a closed CI source

Results
• CH₄-CI spectra had less fragmentation than EI and all metabolites contained several abundant high m/z fragment ions (c.f. Fig 1 & 3). MH+ was at least 5% of base peak for most compounds (Fig 3 & 4). Compared with EI, CH₄-CI spectra were better at distinguishing among sub-classes of compounds because spectra were structurally rich and much of the structural info was in abundant high m/z fragments in “clean” parts of spectrum
• Higher proton affinity of NH₃ led to less fragmentation and simple spectra with little structural info (Fig 4). MH+ or M+18 was base peak for most compounds. The simplicity of NH₃-CI spectra meant they were invaluable for detecting co-eluting peaks (Fig 4)

Conclusions
• CH₄-CI and NH₃-CI spectra are being routinely used for assisting in compound ID
• CH₄-CI is useful for compound ID because of structural info and abundant high m/z fragments
• NH₃-CI is useful for detecting overlapping peaks due to simplicity of spectra
• NH₃-CI reveals MH+ and thus can serve as a quick check on tentative IDs made from EI spectra, or MH+ can be used as a filter to reduce the number of candidates
• Utility of CI for identification would be bolstered by the recent development of robust exact mass GC-MS
• Drawbacks are decreased sensitivity and tailing of late-eluting compounds (Fig 5)

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