Claims

1. A method of self-calibrating a ~~high resolution~~ mass spectrum, ~~optionally with preliminary calibration,~~ comprising:

generating a ~~comprehensive~~ reference set of theoretical mass to charge ratios corresponding to all possible elemental compositions consistent with one or more specified rules over a restricted mass range or ranges, wherein the reference set has a maximum mass or mass to charge ratio selected from the group consisting of: (i) < 100; (ii) 100-200; (iii) 200-300; (iv) 300-400; and (v) 400-500; and

matching preliminary mass to charge ratios of the mass spectrum with ~~or against a~~ the reference set of elemental compositions so that a maximum number of mass spectrum peaks are fitted with expected precision to some elemental compositions from the set when one or more calibration parameters are adjusted.

2. A method as claimed in claim 1, wherein the step of matching mass to charge ratios of the mass spectrum with ~~or against a~~ the reference set of elemental compositions comprises:

finding all the differences between every inaccurate mass to charge value of the mass spectrum and mass to charge values of the reference set of elemental compositions;

grouping found differences according to expected mass accuracy;

selecting ~~the~~ a correct group, where the selection of the correct group is based on: (i) the maximum number of entities in the group; and/or (ii) comparison of natural abundances of the sets of elemental compositions in groups; and

adjusting one or more calibration parameters of a calibration routine so as to reduce the mass error between mass to charge ratios of the spectrum and the corresponding known precise mass to charge ratios of elemental compositions in the selected group.

~~3. A method as claimed in claim 2, where the selection of the correct group is based on: (i) the maximum number of entities in the group; and/or (ii) comparison of natural abundances of the sets of elemental compositions in groups.~~

~~4. A method of self-calibrating a mass spectrometer comprising:~~

~~ionising a sample to generate analyte ions;~~

~~mass analysing at least some of the analyte ions to determine first observed mass to charge ratios of the analyte ions;~~

~~matching at least some of the first observed mass to charge ratios with or against a reference set of possible or predicted elemental compositions having known precise mass to charge ratios; and~~

~~adjusting one or more calibration parameters of a calibration routine so as to optimise the match between one or more of the first observed mass to charge ratios and the corresponding known precise mass to charge ratios of one or more possible or predicted elemental compositions contained within the reference set.~~

~~5. A method as claimed in claim 4, wherein the step of adjusting one or more calibration parameters of a calibration routine further comprises adjusting one or more calibration parameters of a calibration routine so as to reduce the mass error between one or more of the first observed mass to charge ratios and the corresponding known precise mass to charge ratios of one or more possible or predicted elemental compositions contained within the reference set.~~

~~6. A method as claimed in any preceding claim, wherein the reference set of possible or predicted elemental compositions has a maximum mass or mass to charge ratio selected from the group consisting of: (i) < 100; (ii) 100-200; (iii) 200-300; (iv) 300-400; and (v) 400-500.~~

3~~7~~. A method as claimed in any preceding claim, wherein the reference set ~~of possible or predicted elemental compositions~~ comprises a reference set of possible or predicted organic molecules.

4~~8~~. A method as claimed in claim ~~7~~ 3, wherein the reference set of possible or predicted organic molecules have a composition in the form $C_{n1}H_{n2}N_{n3}O_{n4}$.

5~~9~~. A method as claimed in claim ~~8~~ 4, wherein $n1 \leq n2 \leq 2n1 + 2$.

6~~10~~. A method as claimed in claim ~~7~~ 3 or ~~8~~ 4, wherein $n3+n4 \leq 4N$, wherein N is 4, 5, 6, 7, 8, 9, 10, 11 or 12.

7~~11~~. A method as claimed in any ~~of claims 4-10~~ preceding claim, wherein the step of adjusting one or more calibration parameters is made without reference to adding one or more intrinsic or internal calibrants to the sample.

8~~12~~. A method as claimed in any ~~of claims 4-11~~ preceding claim, wherein the step of adjusting one or more calibration parameters is made without reference to mass analysing one or more extrinsic or external calibrants or lock masses.

9~~13~~. A method as claimed in any preceding claim, further comprising removing from the reference set of ~~possible or predicted elemental compositions~~ one or more elemental compositions which are determined either: (i) to have or are likely to have a low or relatively low abundance; or (ii) to be relatively unlikely to be present in the sample.

10~~14~~. A method as claimed in any ~~of claims 4-13~~ preceding claim, wherein the step of mass analysing the sample comprises using a multi-reflecting Time of Flight mass analyser optionally utilising Encoded Frequent Pushing ("EFP").

11~~15~~. A method of mass spectrometry comprising a method as claimed in any preceding claim.

12~~16~~. A mass spectrometer comprising:

an ion source for ionising a sample to generate analyte ions;

a mass analyser for mass analysing at least some of the analyte ions to generate a mass spectrum ~~determine first observed mass to charge ratios of the analyte ions~~; and

a control system arranged and adapted:

(i) to generate a reference set of theoretical mass to charge ratios corresponding to all possible elemental compositions consistent with one or more specified rules over a mass range or ranges, wherein the reference set of possible or predicted elemental compositions has a maximum mass or mass to charge ratio selected from the group consisting of: (i) < 100; (ii) 100-200; (iii) 200-300; (iv) 300-400; and (v) 400-500 ~~to match at least some of the first observed mass to charge ratios with or against a reference set of possible or predicted elemental compositions having known precise mass to charge ratios~~; and

(ii) to match preliminary mass to charge ratios of the mass spectrum with or against the reference set of elemental compositions so that a maximum number of mass spectrum peaks are fitted with expected precision to some elemental compositions from the set when one or more calibration parameters are adjusted ~~to adjust one or more calibration parameters of a calibration routine so as to optimise the match between one or more of the first observed mass to charge ratios and the corresponding known precise~~

~~mass to charge ratios of one or more possible or predicted elemental compositions contained within the reference set~~.

13~~17~~. A mass spectrometer as claimed in claim ~~16~~ 12, wherein the mass spectrometer comprises a multi-reflecting Time of Flight mass analyser ~~optionally utilising Encoded Frequent Pushing ("EFP")~~.

14. A method of *de novo* calibration of mass spectrometry data comprising:

(i) obtaining at least one peak detected mass spectrum containing no *a priori* known species;

(ii) generating theoretical m/z values for all elemental compositions consistent with any specified rules over a restricted m/z range or ranges;

(iii) determining possible matches between the theoretical m/z values and peaks in the mass spectrum;

(iv) adjusting a set of calibration parameters based on the matches; and

(v) choosing calibration parameters which optimise the likelihood of the mass spectral data given the calibration parameters and the set of possible matches, or choosing calibration parameters which optimise the posterior probability of the calibration parameters given the data and the set of possible matches.

15. A mass spectrometer comprising:

an ion source for ionising a sample to generate analyte ions;

a mass analyser for mass analysing at least some of the analyte ions to obtain at least one peak detected mass spectrum containing no *a priori* known species; and

a control system arranged and adapted to perform *de novo* calibration by:

(i) generating theoretical m/z values for all elemental compositions consistent with any specified rules over a restricted m/z range or ranges;

(ii) determining possible matches between the theoretical m/z values and peaks in the mass spectrum:

(iii) adjusting a set of calibration parameters based on the matches; and

(iv) choosing calibration parameters which optimise the likelihood of the mass spectral data given the calibration parameters and the set of possible matches, or choosing calibration parameters which optimise the posterior probability of the calibration parameters given the data and the set of possible matches.