

propionic acid solution was delivered to ES probe tip 4 at a flow rate of 500 ul/min. The calibration solution was Electro-sprayed from probe tip 4 with pneumatic nebulization assist. Mass spectra 64 acquired while Electro-spraying the calibration solution from ES probe tip 4 is shown in FIG. 4b. Peaks 65, 66 and 67 with mass to charge values of 118, 508 and 998 respectively were formed from the singly charged protonated molecular ions of the calibration components of known molecular weight. Other peaks present were from contamination compounds present in solution. The abundance of peak 65 (118 m/z) is approximately 4,300. Mass spectrum 68 in FIG. 4c was acquired while simultaneously spraying sample and calibration solutions from ES tips 3 (50 ul/min) and 4 (500 ul/min) respectively. Sample or gramicidin S peak 71 abundance of approximately 2,600 has been reduced by less than 15% when compared to the gramicidin S peak 61 acquired when independently sprayed. The calibration peak heights have changed less than 15% comparing mass spectra 64 and 68 acquired with single and simultaneous solution spraying.

[0043] The nebulization gas flow and the calibration solution flow through ES tip 4 was turned off during the acquisition of mass spectrum 60 shown in FIG. 4a. Conversely, the nebulization gas flow and the sample solution flow through ES tip 3 was turned off during the acquisition of mass spectrum 64 shown in FIG. 4b. Both calibration and sample solution flows and nebulization gas flows to ES tips 3 and 4 were turned on during acquisition of mass spectrum 68 shown in FIG. 4c. Ions formed from the two independent simultaneous Electro-sprays mixed in the gas phase prior mass analysis allowing acquisition of a mass spectrum with an internal standard. A quadrupole mass analyzer was used to acquire the data shown in FIGS. 4a through 4c. Alternatively, other types of mass analyzers could be used such as Time-Of-Flight, three dimensional quadrupole ion traps, magnetic sector, Fourier Transform Mass Spectrometers and triple quadrupoles. Internal standards within a mass spectrum can be used to improve the accuracy of mass to charge assignments of sample peaks, particularly for mass spectra acquired with higher resolution. The sequence of mass spectra shown in FIGS. 4a through 4c can be acquired in under one minute limited only by the mass spectrum accumulation time and the speed with which individual liquid flow rates can be turned on or off. The invention allows the efficient mixing of gas phase ions produced from multiple solutions Electro-sprayed simultaneously over a wide range of liquid flow rates. Sample and calibration solutions can be introduced through multiple ES probe tips with no need to adjust probe tip position after initial optimization. The invention increases the versatility of an analytical mass analysis system that can accept multiple solution inputs with unattended operation. An Electro-spray ion source comprising multiple inlet probes, configured for independent or simultaneous spraying, minimizes system downtime, maximizes sample throughput, allows selective acquisition of mass spectra with internal standards without contaminating sample solutions. As will be described below, a multiple inlet probe API source can also be used to study ion-ion gas phase interactions at atmospheric pressure.

[0044] In the example shown in FIGS. 4a through 4c, the solution flow to ES tips 3 and 4 was supplied through delivery lines 9 and 10 respectively by liquid pumps which could be turned on or off independently with or without nebulization gas flow. Alternatively, solution 44 can be

supplied to ES tip 7 from solution reservoir 45 as shown in FIG. 2. Solution 45 is drawn to ES tip 7 through delivery line 15 by the venturi force induced from the nebulization gas supplied to ES tip 7 through line gas delivery line 13. With solution reservoir 45 positioned below ES probe tip 7, solution flow to ES tip 7 stops when the nebulization gas is turned off. If no nebulization assist is used when Electro-spraying from ES tip 7, a gas pressure head can be applied to solution 45 in reservoir 44 to aid in initially forcing liquid to ES tip 7. The electrostatic forces from the electric field applied during unassisted Electro-spraying can also maintain solution flow through ES tip 7. Liquid flow to ES tip 7 can then be turned off by removing the gas pressure head on solution 45 in reservoir 45 and reducing the electric field at ES tip 7. Unassisted Electro-spray can be turned on or off by applying the appropriate relative potentials to an individual ES tip and then removing the potential from the tip. For example if two independent ES probes are configured in an ES source and 6,000 volts is applied to each probe independently during ES operation then the spraying from a given probe can be switched on or off by applying kilovolt potentials to the ES probe or lowering the probe voltage to stop the Electro-spray. Each ES tip 3, 4, 5 and 7 can be individually configured to optimize performance for a specific set of applications with a range of liquid flow rates and solution chemistries. ES tips can be configured with single, double and triple tube layers to accommodate various gas and liquid layers at the ES tip connected to specific solution and gas delivery lines. Single layer tips such as replaceable microtips which allow low ES flow rates may be pre-loaded prior to installation in an ES source and do not require solution delivery lines. Multiple microtips can be configured to spray simultaneously if is desirable to acquire mass spectra with an internal standard while Electro-spraying at liquid flow rates in the 25 to 500 nanoliter per second range. For higher liquid flow rates, layered ES tip configurations are typically used.

[0045] FIG. 12 is a diagram of a two layer Electro-spray tip. With a two layered ES tip configuration, nebulization gas 74 can be supplied through annulus 71 between a second layer tube 70 surrounding liquid sample introduction tube 72 to assist the in the formation of charged liquid droplets during Electro-spray operation. Sample bearing solution is delivered to exit end 73 of inner tube 72 through bore 75. A second liquid layer can be delivered through annulus 71 replacing the gas flow if liquid layering is desired during operation at the ES probe tip. Alternatively, ES probe tips may be configured with three concentric layers as diagrammed in FIG. 13. Typically with a three layer ES probe, sample solution is introduced through bore 88 of inner tube 80, a second solution can be introduced through annulus 84 between tubes 80 and 81 and, if required, a gas flow 85 can be delivered through annulus 83 between tubes 81 and 82. The solutions delivered through bore 88 and annulus 84 mix at the first layer tube exit 86 in region 87 during ES operation. The second solution delivered through annulus 84 may contain known calibration compounds which mix with the sample solution delivered through bore 88 in region 87 during ES operation. Conventionally, calibration compounds are mixed with sample bearing solution prior to the solution being delivered through bore 88.

[0046] One ES probe tip or combinations of ES probe tips 3, 4, 6 and 7 can be configured as two or three layer assemblies similar to that shown in FIGS. 12 and 13.