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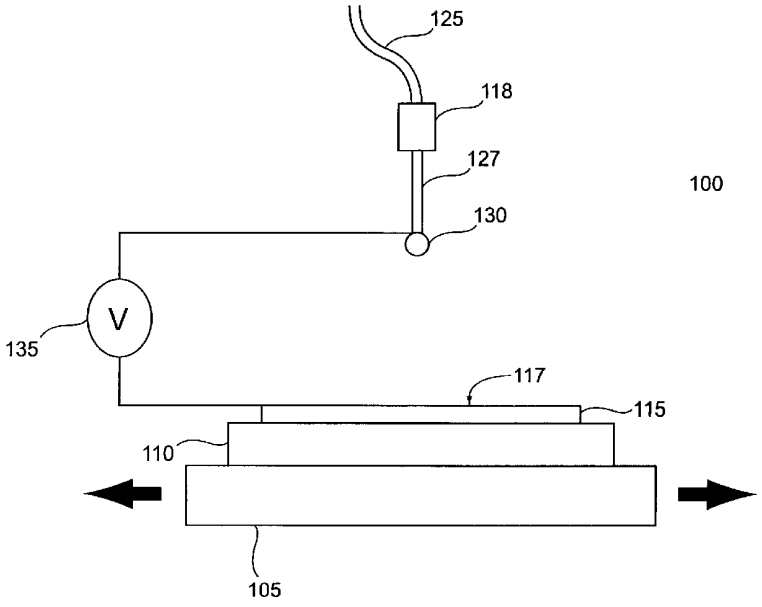
(54) Title: SAMPLE DEPOSITION METHOD AND SYSTEM



(57) Abstract: This invention provides methods and systems for rapidly, accurately and efficiently depositing liquid droplets (130) onto a planar uper surface (117) of sample plate (115) held in sample plate holder (110) which is supported on, or incorporated into, a motion table (105) included in an LC MALDI sample distribution system (100) for MALDI-mass spectrometry sample preparation. System (100) also includes a holding mechanism (118) for holding at least one capillary (125) which terminates at a capillary tip (127) on which droplet (130) is formed. A power supply (135) forms an electric field between sample plate surface (117) and droplet (130) which acts to pull the droplet (130) onto the sample plate (115).

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SAMPLE DEPOSITION METHOD AND SYSTEM

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to US Ser. No. 09/765,207, filed January 17, 2001, which application is incorporated herein by reference.

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BACKGROUND OF THE INVENTION

Field of the Invention

[0003] The present invention relates to mass analysis. More particularly, the present invention relates to sample preparation and handling for mass spectrometry processes.

Background

[0004] Mass spectrometry (MS) is a method of mass analysis in which the chemical composition of a substance is identified by separating gaseous ions derived from the substance according to their mass-to-charge ratio. One type of ionization used in mass spectrometry is known as matrix-assisted laser desorption/ionization (MALDI). In MALDI, a sample and a matrix are co-crystallized from a liquid solution. The resulting co-crystal is irradiated with a laser, which causes the matrix molecules to absorb some of the incident energy. The absorbed energy causes some of the matrix and sample molecules to ionize and desorb into the gas phase. Once the sample molecules have been ionized and desorbed into the gas phase, they are amenable to analysis by mass spectrometry.

[0005] A sample that contains a mixture of chemical compounds is frequently purified prior to analysis by mass spectrometry. One method of purification is by liquid chromatography (LC), in which the mixture of compounds to be separated is dissolved in a liquid phase and the liquid phase is passed over a stationary phase contained in a chromatography column. Compounds that interact more strongly with the stationary phase are retained for a longer period of time on the column, which permits a mixture of compounds to be separated based upon differences in retention times.

[0006] One means of further separating compounds with differing retention times is to fractionate the liquid phase as it exits the chromatography column. The formation of a large number of small amounts of the liquid phase produced by an LC is known as fraction collection. The precision and speed with which fraction collection is performed is critical to achieving high-quality separation and analysis. In addition, after purification by LC, the liquid phase containing the sample molecules can be combined directly with a matrix solution to enable MS analysis. Hereinafter, a liquid refers to any solution containing either sample molecules or matrix molecules, or a mixture of both. A sample refers to a portion of a liquid that has been prepared and provided for analysis, such as by deposition on a sample plate.

[0007] Mass spectrometry analysis is currently being applied to increasingly complicated molecules and mixtures. In addition, advances in medicine, science and technology have created a growing demand for sophisticated analytical tools. This demand, in turn, requires the development of methods for the rapid and efficient preparation, purification and analysis of samples by mass spectrometry. Methods that enable the purification of large numbers of samples by LC and the analysis of large numbers of samples by MS have been developed independently. The demand for a rapid and efficient means to prepare and handle large numbers of LC fractions for analysis by MS, however, remains unmet.

[0008] Typically, systems for automated fraction collection include single-channel devices that use either a touch-down cycle or piezo-electric dispensers for depositing a liquid sample. Touch-down devices use mechanics for physically spotting a liquid droplet onto a fraction collection plate. These devices normally use a narrow capillary connected to

a column that holds the liquid. A tip of the capillary physically contacts the fraction collection plate to deposit a droplet of the liquid.

[0009] Touch-down devices are notoriously prone to misalignment, wear and breakage. Also, preservation of the chromatographic resolution becomes an issue in touch-down collection when small fractions are collected. A significant portion of the liquid deposited in one spot can be carried over to the next spot by sticking to the capillary tip. To remedy carry-over, a make-up flow can be added by teeing-in additional solvent, or by applying a sheath flow, or both. These remedies increase the collected volumes so that a proportionately smaller sample fraction is carried over. However, the larger volumes dilute the sample concentration applied to the fraction collection plate, which, in turn, leads to lower sample densities and lower signal-to-noise in the signal generated by the mass spectrometer.

[0010] A piezo-electric dispenser is a type of non-contact liquid droplet deposition device, in which a piezo-electric element is used to apply a pulse-driven pressure to a dispenser. The pressure forces, or ejects, a droplet of liquid out through a nozzle in the dispenser. Samples deposited using piezo-electric devices typically exhibit reduced chromatographic resolution because a larger dead volume is required to eject the droplet, and the concentration of sample molecules to matrix molecules is correspondingly reduced. Further, problems exist with keeping the dispenser nozzle clean if matrix solvent contains larger amounts of non-volatile material. Application of a sheath flow is not possible in this case.

[0011] Other methods of micro deposition of LC samples include electrospray and streaking. Electrospray is a non-contact sample deposition process, which is sensitive to solvent composition and flow rate and which spreads the sample out over larger areas. Streaking is a contact form of deposition, which requires flash evaporation or freezing to preserve chromatographic resolution. Both electrospray and streaking processes are sensitive to solvent composition.

[0012] Accordingly, a need exists for improved systems for depositing liquid droplets on a plate for subsequent analysis by mass spectroscopy. The present invention fulfills this and other needs.

SUMMARY OF THE INVENTION

[0013] This invention provides a novel non-contact liquid droplet deposition system and method. Advantageously, the invention supports the preparation and handling of a larger number of sample targets, resulting in higher throughput, while providing flexibility by allowing greater control of liquid flow rates, sampling speed, and solvent composition. Additionally, a system and method in accordance with the invention provides for enhanced chromatographic resolution of samples produced using liquid chromatography without sacrificing throughput or flexibility.

[0014] The system and method according to embodiments of the invention provide a rapid, accurate, and efficient interface for depositing the output of one or more liquid chromatography columns onto sample plates for analysis in a mass spectrometer. The system is flexible, permitting the deposition of samples at nearly any spacing, drop size, and solvent composition. Also, the samples can be deposited on a variety of sample plate materials, including both conducting and insulating materials. Non-contact deposition is achieved by a novel method of droplet desorption from fine capillary tips onto a sample plate through the application of a high voltage pulse to the sample plate, which generates an electric field between the sample plate and the capillary tip.

[0015] A method that embodies this invention includes positioning a sample plate below a liquid droplet, and applying an electric field between the liquid droplet and the sample plate. The electric field polarizes the droplet. The polarized droplet experiences a force along the applied electric field and is pulled toward the sample plate.

[0016] Another method embodying this invention includes providing an array of liquid droplets at a distance above a positionable sample plate, and applying an electric field between the droplets and the sample plate. Under the influence of the electric field, the droplets move to target locations on the sample plate.

[0017] In another embodiment of the invention, a liquid droplet deposition system includes a holding mechanism, and an array of capillaries, held and positioned by the holding mechanism, wherein each capillary contains at least a portion of the liquid, and the array of capillaries provides one or more droplets of the liquid simultaneously. The system further includes a sample plate being positionable beneath the plurality of capillaries, and a

power supply. The power supply connected to the capillaries and/or the sample plate for applying a voltage difference between the liquid and the sample plate.

[0018] In still yet another embodiment, a sample is formed of a number of successive droplet depositions, in which each deposition includes positioning a sample plate and a droplet-forming capillary such that the droplet-forming capillary is above a target location on the sample plate. A successive application of a plurality of voltage pulses pulls individual droplets to the same location on the sample plate.

BRIEF DESCRIPTION OF THE DRAWINGS

[0019] FIG. 1 is a simplified diagram of a liquid chromatography sample deposition system according to the invention.

[0020] FIG. 2 illustrates an alternative embodiment of a sample deposition in accordance with the invention.

[0021] FIGS. 3A-3C are simplified diagrams of a deposition system to illustrate a liquid droplet deposition method according to the invention.

[0022] FIGS. 4A-4F show various methods of applying an electric field pulse in accordance with methods of the invention.

[0023] FIG. 5 shows an example of a graphical user interface that can be used to control the sample deposition apparatus of the invention.

[0024] FIG. 6 shows an example of a graphical user interface for use in preparing a calibration plate.

DETAILED DESCRIPTION

[0025] The invention provides an apparatus and methods for depositing liquid samples on a sample plate. The apparatus is useful to, for example, prepare sample plates for subsequent analysis by MALDI-mass spectrometry. Often, the samples are obtained from a liquid chromatography column which is used to fractionate a sample. The sample deposition system is used to collect fractions in the form of discrete droplets that are then subjected to analysis.

[0026] Figure 1 is a simplified diagram illustrating an LC MALDI sample deposition system 100 according to an embodiment of the present invention. The system 100 can include a motion table 105 and a controller. The motion table 105 is movable at least in

a longitudinal direction shown, under direction of the controller. The motion table 105 may also move laterally, vertically, or rotationally.

[0027] A sample plate holder 110 rides on the motion table 105, preferably fixed to the motion table 105. In one embodiment, the sample plate holder 110 is a subassembly of the motion table 105. Other configurations are possible; for example the sample plate holder 110 and the motion table 105 may be provided as a single unit. The sample plate holder 110 provides a base to which a sample plate 115 can be clamped. One or more sample plate holders can be situated on the motion table 105, thus providing an accurate mechanism for accurately and reliably positioning one or more sample plates on the system 100. For example, a motion table can have sample plate holders for two, three, four, five, or more plates. The motion table, which can consist of, for example, a conveyer-type system, moves the sample plate holders, sequentially or non-sequentially, to the loading position under the dispenser. In some embodiments, the sample plate holder 110 uses a built-in spring to hold the sample plate, and one or more pins to accurately align the sample plate.

[0028] The sample plate 115 can be formed of a material that maintains its shape for a sufficient length of time to deposit and analyze the samples. For example, the sample plate can be a rigid or semi-rigid material and preferably has a planar upper surface 117. The upper surface 117 may include an array of wells or small divots in the upper surface 117, each for providing an anchor for a deposited portion of a sample. For example, each well can be an independently addressable target location embedded in the upper surface 117, suitable for micro-arraying applications. Such wells are divots are not required, however. The sample plate 115 can be formed of glass (e.g., a glass slide), metal (e.g., stainless steel, aluminum, titanium, or other metal), a polymer, a ceramic, a porous or nonporous membrane, gel, frozen liquid, and the like. The sample plate can itself be an electrical conductor, in which case one can apply an electrical charge or ground directly to the sample plate. Alternatively, the electrical charge or ground can be applied to an electrode plate which underlies the sample plate, as described below.

[0029] The system 100 typically further includes a holding mechanism 118 adapted to hold one or more very small diameter columns 125, or capillaries. In one embodiment of the invention, the capillary 125 is connected to a high pressure liquid chromatography (HPLC) source that provides the HPLC liquid. The capillary 125

terminates at a capillary tip 127. The capillary tip 127 is connected to the capillary 125 at one end. The second end of the capillary tip 127 terminates to an open tip. The capillary tip 127 can be formed of a rigid or semi-rigid material including, but not limited to, metal, or silica glass. The material used for the capillary tip 127 may also depend on desired electrical characteristics of the material for transferring electric energy to the liquid provided therein, as explained in further detail below.

[0030] The liquid can include sample molecules, matrix molecules, or a mixture of sample molecules and matrix molecules. In one embodiment of the invention, the liquid is a sample solution containing sample molecules, and the matrix molecules are independently provided to the sample plate 115. Methods of applying matrix molecules to the sample plate 115 are known, including, but not limited to, depositing a matrix solution into individual wells formed in the upper surface 117, coating the entire upper surface 117 with a matrix composition, etc. The sample solution is mixed with the matrix solution or composition upon deposition to the sample plate 115. In another embodiment, the matrix solution is mixed with the sample solution in the capillary 125. For example, the matrix can be mixed with the liquid as the liquid exits the chromatography source through an appropriate junction fitting.

[0031] The capillary 125 and capillary tip 127 are adapted to receive a flowing portion 130 of the liquid. Preferably, the portion 130 of the liquid is a droplet. The droplet may have a controlled, adjustable size or volume, depending upon flow-rate through the capillary 125 and a desired size and density of the sample. The formation of the droplet is typically driven by the chromatography pump, rather than by the electric field. Surface tension of the droplet suspends the droplet at the capillary tip 127 until the droplet is pulled away by the influence of an applied electric field. The portion 130 may be a collection of droplets, such as a spray, or even a continual stream of the liquid.

[0032] In accordance with the present invention, the system 100 can include a power supply 135. In some embodiments, the power supply 135 allows for adjustment of the output voltage. The power supply 135 may also include electrodes that are connected to ground, or zero potential. The power supply 135 is configured to energize either the sample plate or the liquid, to create a potential difference between the liquid and the sample plate 117, either of which may also be pre-charged to a particular polarity. For example, in some

embodiments the liquid in the capillary and/or in the droplet at the end of the capillary tip is grounded, while the sample plate is charged. Alternatively, the liquid is charged and the sample plate is grounded.

[0033] In a preferred embodiment, a voltage pulse is provided to a liquid droplet 130, via the liquid, and to the sample plate 115. The application of this voltage pulse therefore creates a potential difference between the sample droplet 130 and the sample plate 115. The voltage pulse can be provided with any combination of electrical connections to the sample plate and the liquid, as will be illustrated below. The voltage level of this pulse can be set either manually or automatically, and the timing and duration of the voltage pulse can be controlled either manually or by software that runs the deposition system 100. Further, the power supply 135 is illustrated in Figure 1 as having a connection to the sample plate 115 and the second, open end of the capillary tip 127. However, the actual physical connection from the power supply 135 may be made in any location that energizes either the sample plate or the liquid. Therefore, the present invention is not to be limited to the specific embodiment shown in Figure 1.

[0034] The system 100 may additionally include a waste and wash plate, which are not shown in Figure 1 for ease of explanation. The waste and wash plate allows cleaning of the capillary tips, from which samples are deposited, and provides a location for dropping extra sample before and/or after the sample plates are spotted with samples.

[0035] Figure 2 illustrates a liquid droplet deposition system 200 according to another exemplary embodiment of the invention. The system 200 includes a holding mechanism 118 that is configured to hold an array of capillaries 125. Each capillary connects to a capillary tip 127, which bears a droplet 130 of the liquid. The sample plate 115 can be positioned to any desired location below the array of capillaries 125. The sample plate 115 may optionally include an array of target locations 116, such as a well or an etched outline of a location, for example. The array of target locations 116 is illustrated as having only a single row, however those skilled in the art would recognize that multiple rows and columns of target locations 116 are possible, based in part on the desired spot size of the liquid droplet.

[0036] In the embodiment shown, eight capillaries 125 are held in position by the holding mechanism 118, however other numbers of capillaries may be provided by the

system 200 of the present invention. Therefore, the number of capillaries 125 in the array of capillaries is not limited to any specific number. The number of capillaries may be limited by a number of target locations 116 of the sample plate 115 used in the system 200.

[0037] A power supply 135 is shown in the embodiment as connected between the holding mechanism 118 and the sample plate 115. As discussed, the connection may be made anywhere between the droplet and the sample plate. In an embodiment, a separate power supply, or a separate electrical connection from a power supply, may be provided for each capillary 125 and tip 127 for greater flexibility. In such an embodiment, the size and diameter of each liquid droplet generated by a particular capillary could be adjusted independently, and each droplet could be deposited on the sample plate independently of deposition from other capillaries. In another embodiment, the sample plate 115, or a substrate connected with the sample plate 115, is configured with an array of electrically addressable deposition sites or independent counter electrodes. The sites or electrodes could correspond to the target locations 116, for application of a differential voltage to the sample plate 115. For example, the sample plate or an electrode plate that underlies the sample plate can include circuitry which connects each sample location to a separate connector, which connector is plugged into a corresponding connector on the sample deposition system. Each particular target location is thus independently addressable.

[0038] Figures 3A-3C are simplified diagrams of a system to illustrate a liquid droplet deposition method according to the invention. In Figure 3A, at least a portion of a liquid is provided to a capillary 125. A holding mechanism 118 may be adjusted to position the capillary and a capillary tip 127 to a predetermined position. A sample plate 115 is moved into a position below the capillary tip 127.

[0039] Referring now to Figure 3B, the flow of liquid through the capillary 125 forms a droplet 130 at an open end of the capillary tip 127. The size and contents of the droplet are controlled for a specific desired sample spot size and/or sample density. In some embodiments, each droplet has a volume of less than 10 microliters. Preferably, the volume of each droplet ranges between 100 and 200 nanoliters. As shown in Figure 3C, a voltage differential 135 is applied between the droplet 130 and the sample plate 115, to attract the droplet 130 away from the capillary tip 127 to a target location 116 on the sample plate 115, and to form a sample spot 132.

[0040] A method according to the invention uses a high voltage pulse to create an electric field between the liquid sample and the sample plate. In one embodiment, a charge is applied to liquid droplets formed at the end of the capillary tips. In another embodiment, a charge is applied to the sample plate. The duration of the voltage pulses, and the interval between each pulse, are adjustable and controlled for desired throughput of sample deposition, and based in part on the size of the desired spot size.

[0041] Figures 4A – 4E illustrate various methods of creating an electric field between a portion of the liquid and the sample plate. Turning first to Figure 4A, there is illustrated a method of depositing a liquid droplet, whereby the sample plate 110 is connected to a positive or negative high voltage power supply. The voltage is applied in one or more pulses, which correspond to a time period in which deposition is to be made.

[0042] A liquid droplet, formed at the end of a capillary tip 127, can be neutrally charged, which, during application of a positive or negative voltage pulse to the sample plate, creates a potential difference between the liquid droplet and the sample plate. The potential difference between the liquid droplet and the sample plate generates an electric field along which the liquid droplet travels. In Figure 4A, the liquid is grounded through a metal tee 140 that is a part of the holding mechanism 118, and the capillary tip 127 is formed of a nonconductive material such as fused silica glass. In Figure 4B, the capillary tip 127 is made of a conductive material, such as metal, and is connected directly to ground.

[0043] Figure 4C illustrates an alternative method for creating an electric field for depositing a sample. In this case, the capillary tip 127 is disposed in a sheath 128. The sheath 128 surrounds the capillary tip 127 along its length. A sheath liquid 129 is fed into the sheath 128. The sheath liquid 129 is polarized to a polarity that is opposite the polarity of a voltage pulse to be applied to the sample plate 110. Alternatively, the sheath liquid 129 may have a neutral polarity or a ground potential. The sheath liquid contacts a liquid droplet at the end of the capillary tip 127.

[0044] An electrode may also be used to apply a charge to either the liquid or the sample plate, as shown in Figures 4D and 4E. In Figure 4D, the sample plate 110 is grounded, and an electrode is placed in contact with the liquid droplet 130 for a short duration. The electrode may be persistently connected to the capillary tip 127, or physically movable to touch the liquid directly if the capillary tip is formed of nonconductive material.

Further, the electrode may be switchably coupled to the liquid or droplet according to a desired interval. In Figure 4E, the liquid is grounded according to any method, including those methods mentioned above. An electrode is then coupled to the sample plate 110 to apply a high voltage pulse. As discussed, the electrode can include a switch for physical connection to the sample plate when a voltage pulse is needed.

[0045] Figure 4F illustrates another embodiment of a system and method according to the invention. The sample plate 110 is mounted over an electrode plate 150, which is connected to a voltage source. Preferably, the voltage source is configured to apply a voltage pulse to the electrode plate 150 at predetermined intervals, and for a predetermined adjustable duration. In this embodiment and the other aforementioned embodiments, the duration of the voltage pulses ranges from 100-300 milliseconds, and is preferably about 200 milliseconds. However, other pulse durations may be used without departing from the scope of the invention. For example, application of a pulse of duration longer than 300 milliseconds can achieve an electrospray of sample to the sample plate, if desired. In this embodiment and the other aforementioned embodiments, the distance between the sample droplet and the sample plate ranges from one to ten millimeters, and is preferably about five millimeters. However, other distances may be used without departing from the scope of the present invention.

[0046] The voltage difference generally ranges from 500 to 3000 volts. However, other voltage differences may be used without departing from the scope of the invention. The charged electrode plate 150 creates an electric field, shown generally in the simplified diagram of Figure 4F. The droplet 130 formed at the end of the capillary tip 127 is polarized according to the electric field 155, and is then attracted toward the electrode plate 150 to the sample plate 110, along the electric field 155 path.

[0047] In another exemplary embodiment, a liquid portion is deposited to a sample plate as a succession of droplets pulled to a single location on the sample plate. According to this embodiment, a target location on the sample plate is positioned with respect to a liquid to be deposited. A succession of electric fields are generated, preferably by a number of voltage pulses, which attract a like number of liquid droplets to the target location. By increasing the number of pulses without moving the sample plate relative to the capillary, the volume of liquid at the target location is increased. This embodiment improves

resolution of the deposited sample solution, and reduces back-mixing of molecules in the droplets to the liquid.

[0048] The use of multiple pulses at each target location is particularly useful for collecting fractions from a chromatograph. As an illustrative example, to collect a particular fraction volume one might provide a pulse of 200 milliseconds once per second for ten seconds. The number of pulses at each target location, and the frequency and duration of the pulses, can be adjusted to result in a desired volume of sample at each location. The number of pulses at each location can vary as needed for a particular application. Typically, where two or more pulses are used at each sample location, 100 or fewer pulses are used, in some embodiments the number of pulses is between 50 or fewer, and can be 20 or fewer. The frequency of pulses can also vary greatly, depending upon the particular application. For example, a pulse cycle can consist of an equal amount of time for voltage-on voltage-off, or the voltage-on time can be longer or shorter than the voltage-off time in each cycle. In some embodiments, the charge is provided as alternating current, thus resulting in pulses of current in opposite directions separated by a period in which the current flow is zero. For purposes of the invention, the directionality of the current is not significant to the transfer of liquid.

[0049] The systems of the present invention typically include at least one computer (or other information appliance) operably connected to or included within various system components. The computer typically includes system software that directs the sample plate transport and the pulsing systems to, e.g., move a plate into position under the capillaries, move a plate so that a desired target location is positioned under a corresponding capillary, control the duration, frequency, and intensity of pulses, control the rate at which liquid is pumped through the chromatography system, and the like. Additionally, the sample deposition system is optionally coupled to an appropriately programmed processor or computer which functions to instruct the operation of these instruments in accordance with preprogrammed or user input instructions, receive data and information from these instruments, and interpret, manipulate and report this information to the user. As such, the computer is typically appropriately coupled to the instrument (e.g., including an analog to digital or digital to analog converter as needed).

[0050] The computer typically includes appropriate software for receiving user instructions, either in the form of user input into a set of parameter fields, e.g., in a GUI, or

in the form of preprogrammed instructions, e.g., preprogrammed for a variety of different specific operations. The software then converts these instructions to appropriate language for instructing the operation of one or more the handling system, the fluid direction system, or the like to carry out the desired operation, e.g., varying or selecting the rate or mode of movement of various system components, or the like. The computer then receives the data from the one or more sensors/detectors included within the system, and interprets the data, either provides it in a user understood format, or uses that data to initiate further controller instructions, in accordance with the programming, e.g., such as in fluid flow rates, fluid volumes, pulse duration and frequency, or the like.

[0051] In certain embodiments, Microsoft WINDOWS™ software written using instrument control language (ICL) scripts is adapted for use in the sample deposition systems of the invention. Optionally, standard desktop applications such as word processing software (e.g., Microsoft Word™ or Corel WordPerfect™) and database software (e.g., spreadsheet software such as Microsoft Excel™, Corel Quattro Pro™, or database programs such as Microsoft Access™ or Paradox™) can be adapted to the present invention by inputting character strings corresponding to reagents or masses thereof. For example, the systems optionally include the foregoing software having the appropriate information, e.g., used in conjunction with a user interface (e.g., a GUI in a standard operating system such as a Windows, Macintosh or LINUX system) to manipulate relevant information. One example of a suitable graphical user interface is shown in Fig. 5. This GUI provides for operator input of various parameters including, for example, the number of sample target locations per sample plate, the duration of each pulse, the particular calibration plate to be used, and other parameters as shown. Fig. 6 shows an example of a GUI for use in calibrating the instrument.

[0052] The computer can be, e.g., a PC (Intel x86 or Pentium chip-compatible DOS™, OS2™, WINDOWS™, WINDOWS NT™, WINDOWS95™, WINDOWS98™, WINDOWS 2000™, WINDOWS XP™, LINUX-based machine, a MACINTOSH™, Power PC, or a UNIX-based (e.g., SUN™ work station) machine) or other common commercially available computer which is known to one of skill. Software for performing the sample deposition methods is optionally easily constructed by one of skill using a standard programming language such as Visual basic, Fortran, Basic, Java, or the like. Any controller

or computer optionally includes a monitor which is often a cathode ray tube ("CRT") display, a flat panel display (e.g., active matrix liquid crystal display, liquid crystal display), or others. Computer circuitry is often placed in a box (e.g., adjacent to the sample deposition system of the invention), which includes numerous integrated circuit chips, such as a microprocessor, memory, interface circuits, and others. The box also optionally includes a hard disk drive, a floppy disk drive, a high capacity removable drive such as a writeable CD-ROM, and other common peripheral elements. Inputting devices such as a keyboard (e.g., a touch screen, etc.) or mouse optionally provide for input from a user.

[0053] It is understood that the examples and embodiments described herein are for illustrative purposes only and that various modifications or changes in light thereof will be suggested to persons skilled in the art and are to be included within the spirit and purview of this application and scope of the appended claims. All publications, patents, and patent applications cited herein are hereby incorporated by reference for all purposes.

WE CLAIM:

1. A method of depositing a liquid droplet, the method comprising:

providing a sample plate at a distance from a liquid droplet; and

generating an electric field between the liquid droplet and the sample plate to polarize the liquid droplet, wherein the liquid droplet is either charged or grounded;

wherein the liquid droplet is pulled to the sample plate along the electric field.

2. The method of claim 1, wherein providing a sample plate comprises moving the sample plate to a target position.

3. The method of claim 1, wherein the liquid droplet is pulled to a target location on the sample plate.

4. The method of claim 1, wherein the liquid droplet is provided at a tip of a column.

5. The method of claim 4, wherein the column comprises a capillary.

6. The method of claim 1, wherein generating an electric field includes applying a voltage to the liquid droplet.

7. The method of claim 6, wherein generating an electric field comprises connecting the sample plate to ground.

8. The method of claim 7, wherein the sample plate is connected to ground indirectly via an electrode plate that underlies the sample plate.

9. The method of claim 1, wherein generating an electric field comprises applying a voltage to the sample plate.

10. The method of claim 9, wherein the voltage is applied to the sample plate indirectly via an electrode plate that underlies the sample plate.

11. The method of claim 9, wherein generating an electric field comprises connecting the liquid droplet to ground.

12. The method of claim 1, wherein generating an electric field comprises applying a voltage to more auxiliary electrodes.

13. The method of claim 1, wherein the distance between the liquid droplet and the sample plate is less than ten millimeters.

14. The method of claim 13, wherein the distance between the liquid droplet and the sample plate is approximately five millimeters.

15. The method of claim 1, wherein the electric field has a duration of 100-300 milliseconds.

16. The method of claim 15, wherein the electric field has a duration of approximately 200 milliseconds.

17. The method of claim 1, wherein the voltage has a range of 500-3000 volts.

18. The method of claim 17, wherein the voltage is approximately 1000 volts.

19. The method of claim 1, wherein two or more drops are deposited at each target location on the sample plate.

20. A liquid droplet deposition system, comprising:

a holding mechanism;

a plurality of columns, held by the holding mechanism;

a sample plate positionable with respect to the plurality of columns; and

a power supply configured to generate an electric field between each column and the sample plate along which a droplet of liquid which can form at an end of the column is pulled to the sample plate.

21. The liquid droplet deposition system of claim 20, wherein the each column comprises a capillary.

22. The liquid droplet deposition system of claim 21, wherein each column comprises:

a column that contains a liquid from which the liquid droplet is formed; and

a capillary tip, connected to the column at a first end, and including an open tip at a second end for providing the droplets.

23. The liquid droplet deposition system of claim 20, wherein the column comprises a liquid chromatography column.

24. The liquid droplet deposition system of claim 20, wherein the sample plate is coupled to a movable sample plate holder.

25. The liquid droplet deposition system of claim 20, wherein the sample plate is positioned below the array of capillaries.

26. The liquid droplet deposition system of claim 20, further comprising means for moving the sample plate to a target position.

27. The liquid droplet deposition system of claim 20, wherein the power supply includes a voltage source for applying a voltage to the sample plate.

28. The liquid droplet deposition system of claim 27, wherein system comprises an electrode plate through which the voltage is applied indirectly to the sample plate indirectly.

29. The liquid droplet deposition system of claim 27, wherein the system comprises an electrical connection which grounds a liquid droplet at an end of the column.

30. The liquid droplet deposition system of claim 20, wherein the power supply permits the independent application of a voltage to each of a plurality of liquid droplets.

31. The liquid droplet deposition system of claim 20, wherein the power supply permits the independent application of a voltage to different parts of the sample plate.

32. The liquid droplet deposition system of claim 31, wherein the power supply further includes a ground connection for grounding the liquid droplet.

33. The liquid droplet deposition system of claim 20, wherein the power supply includes a voltage source for applying a voltage to the liquid droplet.

34. The liquid droplet deposition system of claim 20, further comprising a controller which controls one or more parameters selected from the group consisting of: movement of the sample plate, intensity of the electric field, duration of the electric field, and frequency of application of the electric field.

35. The liquid droplet deposition system of claim 34, wherein the controller comprises a computer which is programmed with software which provides a user interface which allows an operator to set parameters for the sample deposition.

36. A liquid droplet deposition method, comprising:

providing a plurality of liquid droplets above a positionable sample plate; and

attracting each droplet to a target location on the sample plate with an electric field formed between the plurality of droplets and the sample plate.

37. The method of claim 36, wherein the plurality of liquid droplets includes eight or more droplets.

38. The method of claim 36, wherein attracting each droplet to a target location is performed in a succession to a single target location.

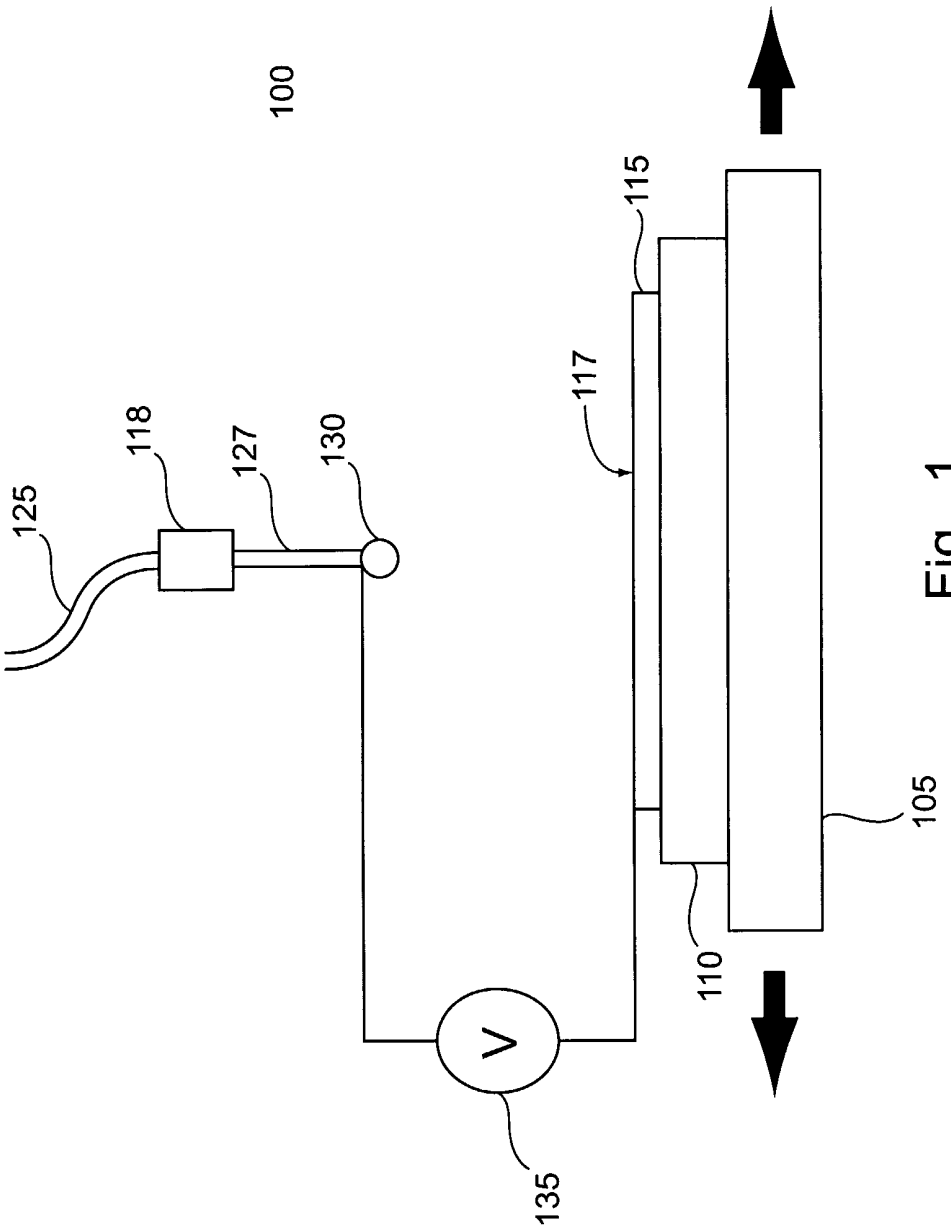

Fig. 1

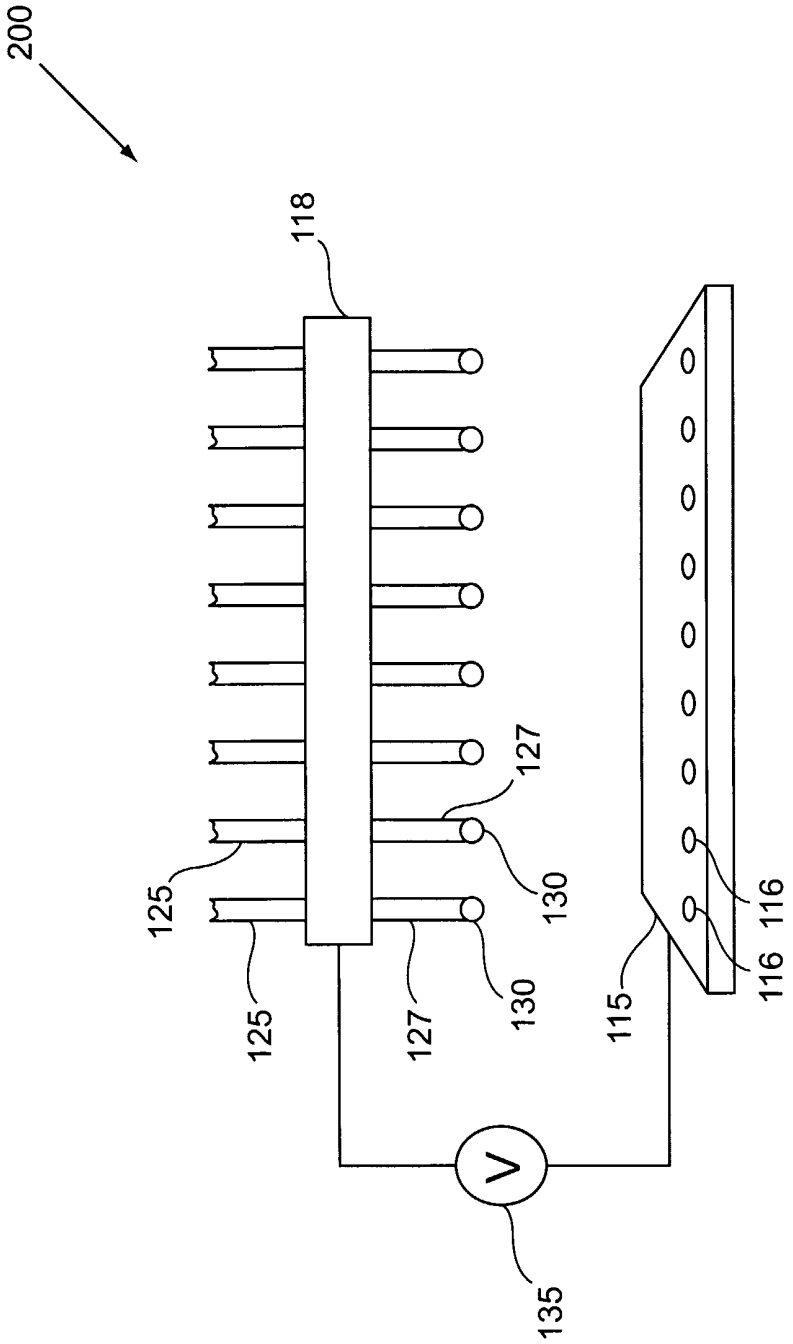

Fig. 2

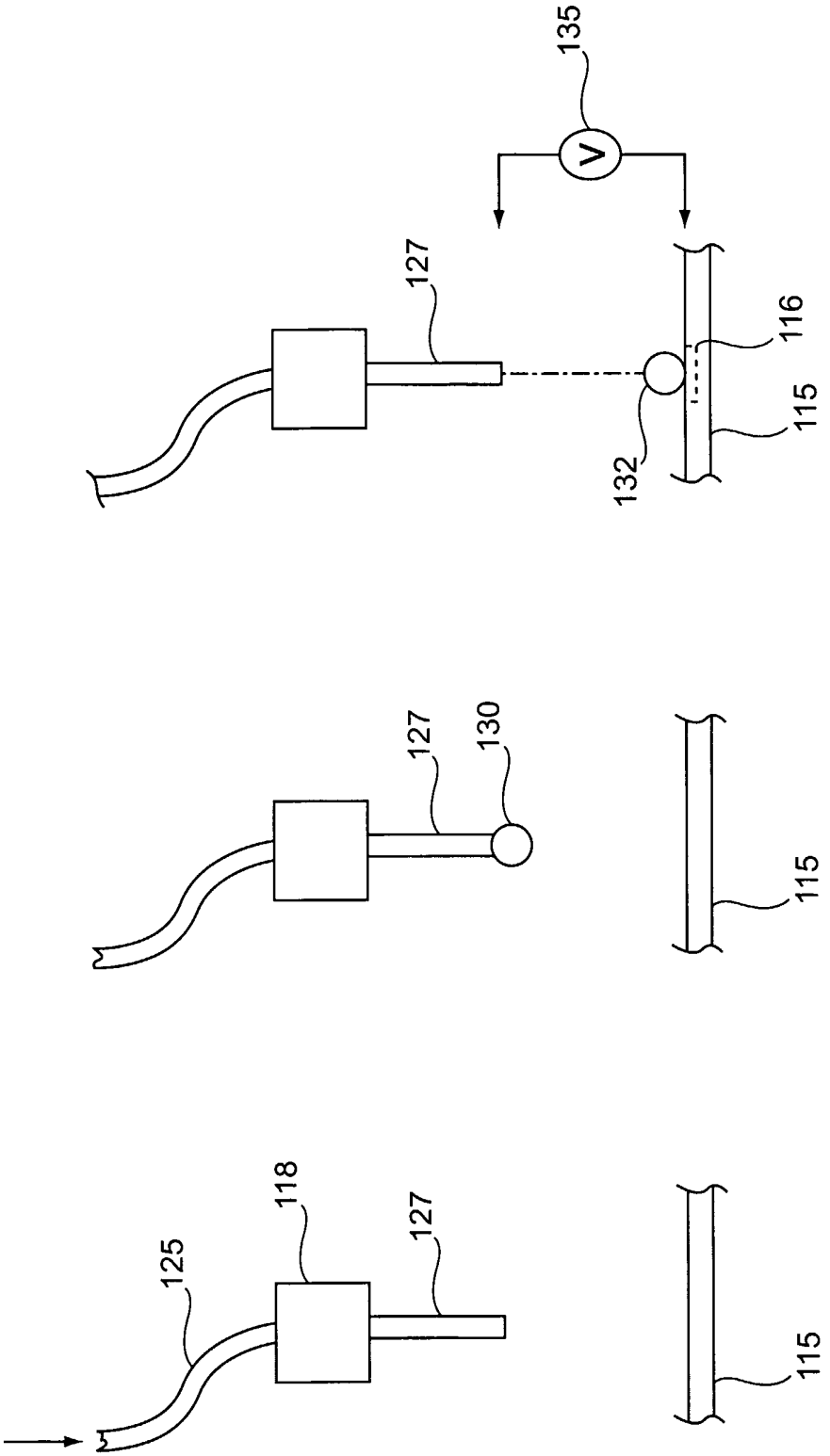

Fig. 3C

Fig. 3B

Fig. 3A

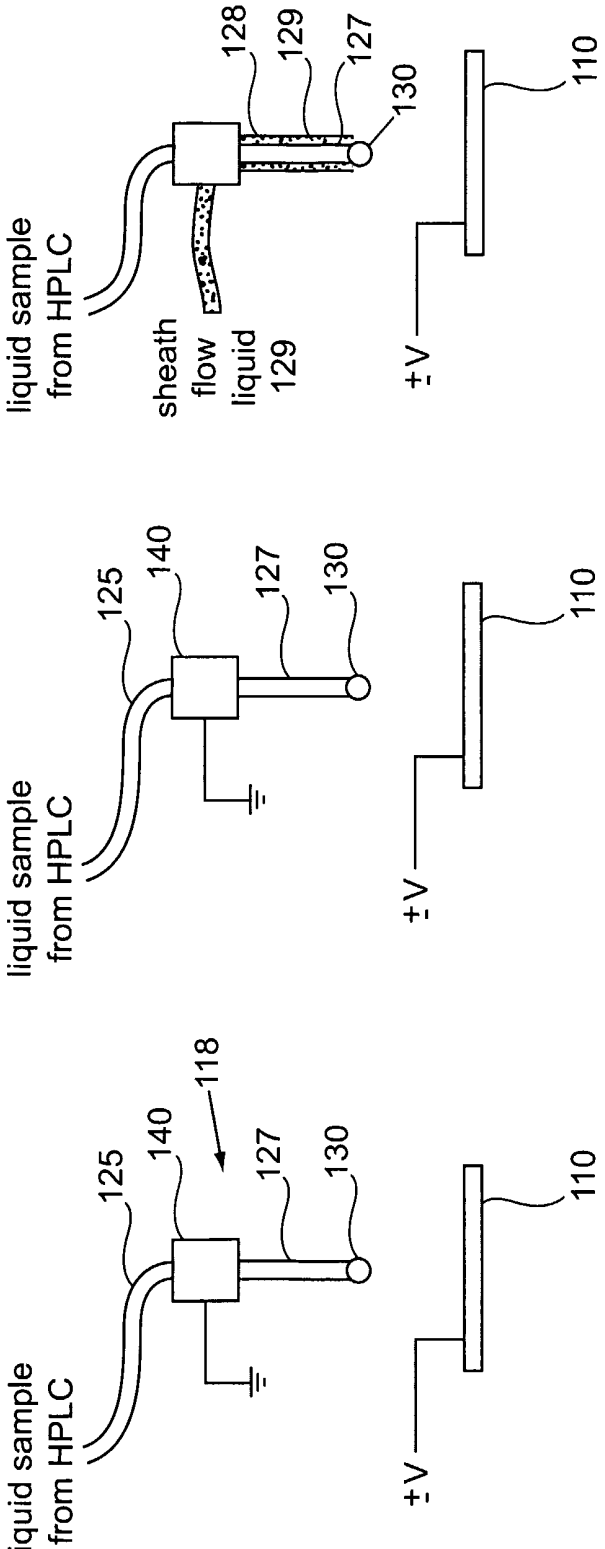

Fig. 4C

Fig. 4B

Fig. 4A

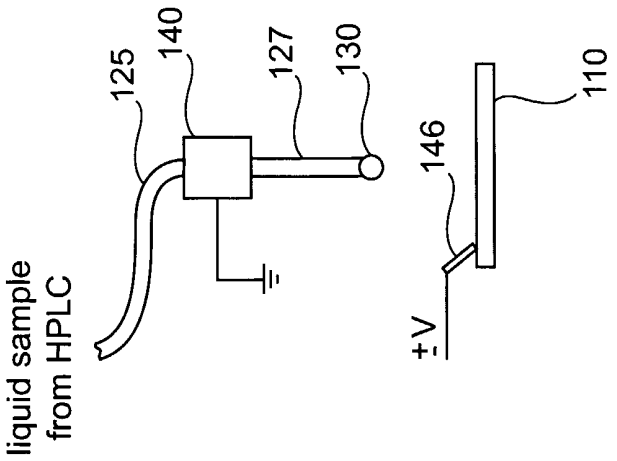
liquid sample
from HPLC
125
140
127
130
146
110
±V

Fig. 4E

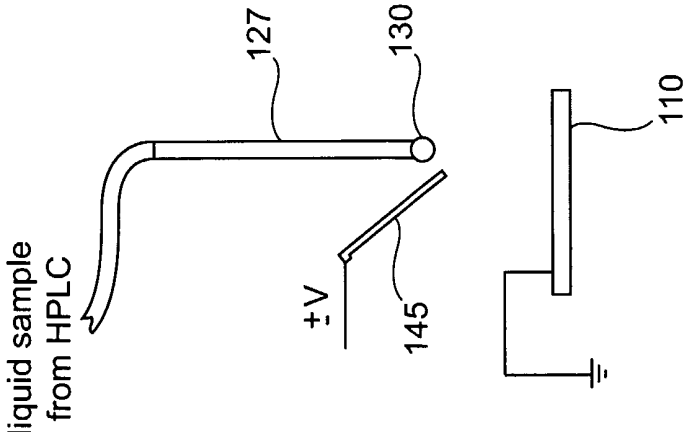
liquid sample
from HPLC
127
130
±V
145
110

Fig. 4D

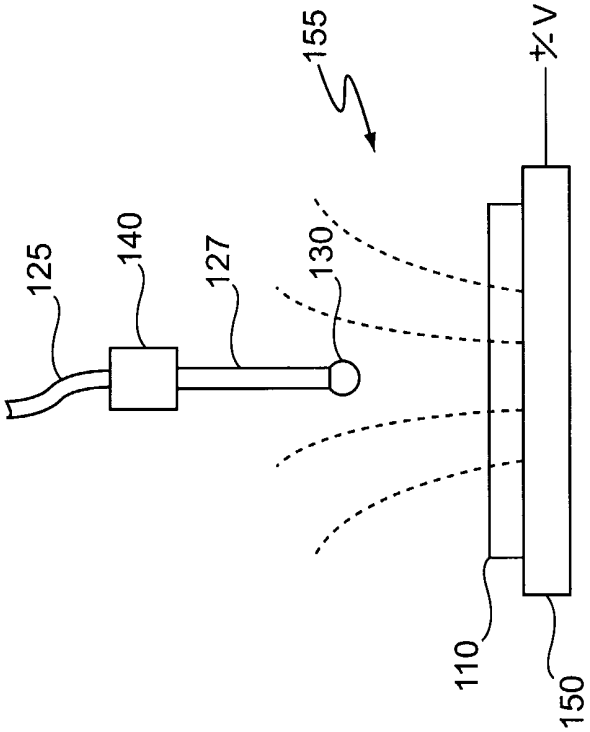

Fig. 4F

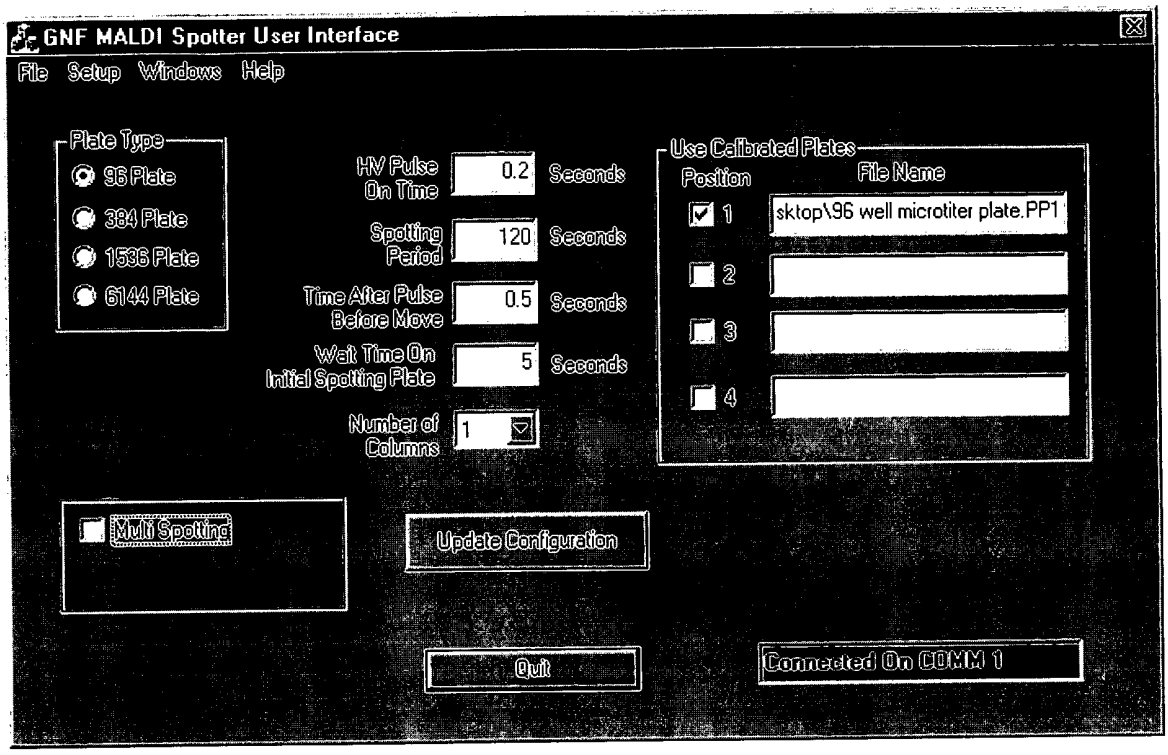
GNF MALDI Spotter User Interface
File Setup Windows Help
Plate Type
96 Plate
384 Plate
1536 Plate
6144 Plate
HV Pulse On Time
0.2
Seconds
Spotting Period
120
Seconds
Time After Pulse Before Move
0.5
Seconds
Wait Time On Initial Spotting Plate
5
Seconds
Number of Columns
1
Use Calibrated Plates
Position
File Name
1
sktop\96 well microtiter plate.PP1
2
3
4
Multi Spotting
Update Configuration
Quit
Connected On COMM 1

Fig. 5

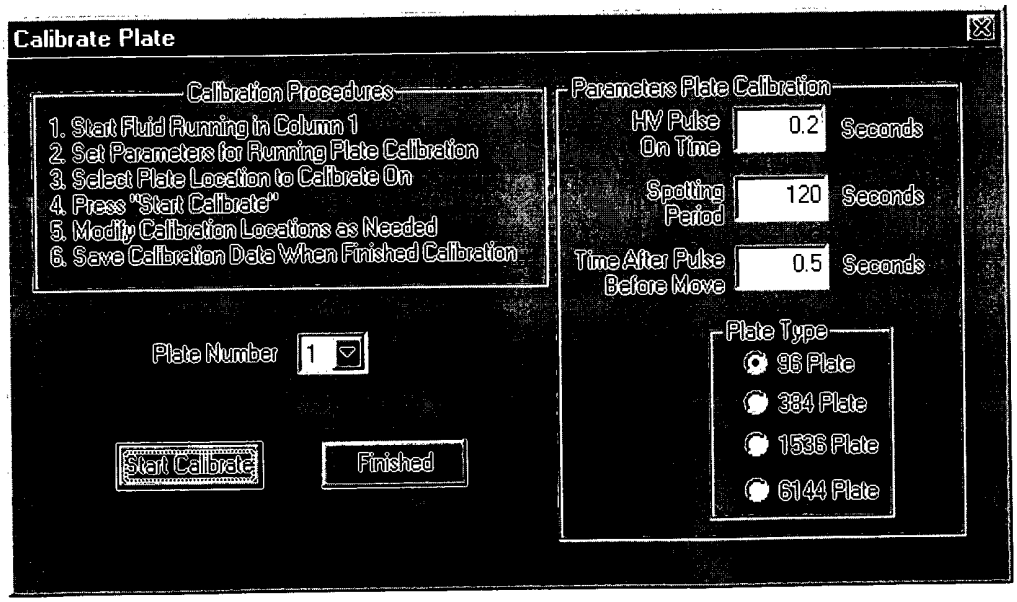
Calibrate Plate
Calibration Procedures
1. Start Fluid Running in Column 1
2. Set Parameters for Running Plate Calibration
3. Select Plate Location to Calibrate On
4. Press "Start Calibrate"
5. Modify Calibration Locations as Needed
6. Save Calibration Data When Finished Calibration
Plate Number
1
Start Calibrate
Finished
Parameters Plate Calibration
HV Pulse On Time
0.2
Seconds
Spotting Period
120
Seconds
Time After Pulse Before Move
0.5
Seconds
Plate Type
96 Plate
384 Plate
1536 Plate
6144 Plate

Fig. 6

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US02/01536

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) :B01L 3/00; B01L 3/02; G01N 1/14
US CL :222/420; 73/863.32, 864.01, 864.25; 422/100; 436/180; 137/825

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : Please See Extra Sheet.

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

Please See Extra Sheet.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- Y	US 6,149,815 A (SAUTER) 21 November 2000 (21.11.2000), abstract, Figs. 1-2, col. 7, line 24- col. 9, line 61 and col. 14, lines 28-50	1-14, 17-25, 27-34, 36-38 --- 15-16, 26, 35
Y	US 3,476,291 A (GLASER) 04 November 1969 (04.11.1969), abstract, Figs. 1-4, col. 3, lines 42-46 and col. 5, lines 43-52	15-16
Y	US 5,916,524 A (TISONE) 29 June 1999 (29.06.1999), abstract and col. 11, lines 32-46	15-16
Y	US 6,132,582 A (KING et al) 17 October 2000 (17.10.2000), abstract, Fig. 1 and col. 13, lines 35-55	26, 35

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

*	Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"A"	document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"E"	earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family
"O"	document referring to an oral disclosure, use, exhibition or other means		
"P"	document published prior to the international filing date but later than the priority date claimed		

Date of the actual completion of the international search 12 JUNE 2002	Date of mailing of the international search report 10 JUL 2002
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer THOMAS P. NOLAND Telephone No. (703) 305-4765

Deborah Perry-Leeper VEGA
Paralegal Specialist
Technology Center 2800

Form PCT/ISA/210 (second sheet) (July 1998)★

B. FIELDS SEARCHED
Minimum documentation searched
Classification System: U.S.

222/420; 73/23.37, 23.41, 61.55, 863-863.03, 863.31, 863.32, 864.01, 864.25; 422/100, 930; 436/180; 137/2, 803, 825, 829; 141/31; 347/55; 101/489, Dig. 37; 250/288

B. FIELDS SEARCHED
Electronic data bases consulted (Name of data base and where practicable terms used):

IBM_TDB, US-PGPUB, EPO, JPO, DERWENT, USOCR, USPAT
search terms: droplet, liquid, drop, liquid-drop, liquid-droplet, sample, sampler, sampling, specimen, plate, sample-plate, sampler-plate, sampling-plate, specimen-plate, electric, electrical, field, electric-field, electrical-field