

transported along these lines. By arranging two electrode pads of opposite polarity separated by a suitable distance, the curve of electric field lines will reach the substrate surface thus transporting target molecules not only horizontally but also vertically towards the probe on the substrate surface. Additional pads can be positioned between the two opposing electrodes. Switching sequences can be employed to ensure that the target molecules pass every probe on the substrate.

[0188] It is also possible to mix liquid crystals (LC) into the sample solution. Because LC are highly polar and highly elliptically shaped particles, they can easily be manipulated by external electric fields to move in desired directions along the field lines. As the LC are moved, the LC create a flow in the surrounding liquid, thereby moving the liquid more readily to bring target molecules in contact with probe molecules.

[0189] FIG. 33 illustrates an electric field gradient which can be used to drive negatively charged molecules in a liquid sample. The liquid sample can be, for example, an aqueous solution that is polar. When a negatively charged molecule, such as DNA or RNA, is subjected to an electrical field  $E$ , a dipole moment,  $P$ , is induced. By applying an inhomogeneous electric field to the dipole, the dipole will be forced toward the lower energy density region. Therefore, by applying an electrical field to the hybridization chamber such that the lower energy density region is along the surface of the substrate, the negatively charged molecules are forced towards the surface of the substrate. The hybridization process can be accelerated due to the higher possibility of collision between the target DNA/RNA molecules and the probes on the substrate.

[0190] FIG. 34 illustrates an embodiment in which Lorentz forces are applied to move charged molecules in the liquid sample. The spacers along the sides of the hybridization chamber can be formed to conduct electricity. This can be accomplished, for example, by forming metal coated areas on the cover, the substrate, or a middle layer at each side of the hybridization chamber to serve as spacers as well as electrodes. A voltage applied across the two electrodes drives charged target molecules in the hybridization liquid to move in parallel with the substrate surface. A pair of magnets establishes a magnetic field across the hybridization chamber in perpendicular to the motion of the charged molecules. The magnetic vector is oriented so that the Lorentz force will push the target molecules to migrate towards the probes on the substrate surface.

[0191] In one embodiment, the voltage can be held constant while the orientation of the magnetic field vector is periodically reversed. The Lorentz force reverses directions periodically causing the charged molecules to follow a zig-zag route between the cover slip and the surface of the microarray from one electrode to the other. The polarity of the voltage can also be switched to change the direction of the molecules movement. This can create improved contact between the target molecule and the probes on the substrate.

[0192] It is possible to split the two electrodes on each side or add two additional electrodes on the other two sides of the hybridization chamber, as shown in FIG. 35a and b, which show a top view of two embodiments of the invention. In FIG. 35a, the charged target molecules can move in lateral or diagonal directions towards the opposite ends of the

substrate. In FIG. 35b, the charged molecules can now move in two perpendicular directions in the microarray substrate surface. By switching the four electrodes on and off in a designed sequence, the target molecules can be driven to contact all probes on the substrate.

[0193] FIG. 36 illustrates an embodiment for generating movement of target molecules by localized heating and/or cooling. An increase of temperature in a localized position in the hybridization liquid can cause the liquid at and near this location to expand and rise. In a cooled environment, the liquid then cools, contracts and descends. A convection driven circulation can be established by utilizing this heating/cooling fluid dynamic. A hybridization apparatus can be fabricated based on this principle. As illustrated in FIG. 36a, a Peltier heat pump is provided on the cover slip. The heat pump heats one position of the liquid while simultaneously cooling another position to establish a convective circulation between the two positions. The temperature change caused by such heating and cooling may be kept small so that the temperature remains within the range at which hybridization or associations of target and probe occurs. In other embodiments, the temperature differential need not be provided by a Peltier heat pump, and can be provided with any heating element and cooling element.

[0194] The establishment of temperature differential caused by the heat pump utilizes gravity to cause convective circulation. When the liquid layer between the cover slip and the substrate is very thin, vertical flow between cover slip and substrate may be difficult to establish. A way to establish more efficient convection is to stand the cover slip and substrate assembly on its edge during hybridization, as shown in FIG. 36b. Furthermore, a number of such heating-cooling pairs can be arranged across the microarray to establish multiple circulations. The heating-cooling poles can be reversed. In addition, phases of the heat-cool cycles among different pairs can be programmed to establish a "global" liquid circulation throughout the entire hybridization chamber.

[0195] For example, if the positions or temperatures of all heating-cooling pairs remain stable, a particular target molecule will be trapped in a local circulation around a particular pair. However, if, in the middle of a circulation, a temperature or position change is introduced to the pairs, a new circulation pattern will be established. By controlling the changing temperature or position, this method can be used to transport the molecule into a different circulation around a different pair. By alternating circulation patterns in a programmed fashion, any target molecule in the fluid can be transported anywhere on the substrate.

[0196] An effective interaction between the sample solution and the probes on the substrate can also be achieved by pneumatically driving the sample solution in and out of the hybridization chamber through microfluidic channels. In accordance with embodiments of this method, microfluidic channels are fabricated on a cover slip, which is placed on top of the microarray for hybridization with the microchannels facing the array.

[0197] In one embodiment as illustrated in FIG. 37, probes on the microarray substrate may be arranged in such a way that there is extra space between columns or rows for wall portions found in the cover slide to contact the substrate to form channels without contacting probes. Open-top