Free Flow Ion Concentration Polarization Focusing (FF-ICPF)

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Electronic Supplementary Information

1| Effect of high electric power to the stability of the system

A high throughput of the device requires high velocities in the separation direction and hence a high electric field generating high electric power. The high electric power can affect the stability in two ways: (1) in the form of heat and; (2) in the form of changes in pH due to electrochemical reactions.

1.1| Heat analysis

The electric current running through the system results in Ohmic heating. In the typical experiments we performed, the maximal applied voltage is approximately 100 V, resulting in approximately 100 μ A of current (in 0.1xPBS) and a dissipated power in the order of magnitude of 0.1 W. The heat can be dissipated in the system via water cooling due to the flow of the sample through the chip or via heat transfer to the environment. The water cooling power ($P_w[W]$) can be calculated via:

$$P_w = Q_f \rho C_p \Delta T(C.1)$$

where $Q_f[m^3s^{-1}]$ is the volumetric flow of the liquid, $\rho [kg m^{-3}]$ the liquid density, $C_p[JK^{-1}kg^{-1})]$ is the specific heat capacity of the liquid and $\Delta T[K]$ is the temperature difference between inlet and outlet. Using the heat capacity of water (4.19 kJ K⁻¹ kg⁻¹)) and a flow rate of 10 µl min⁻¹, we calculate a water cooling power of approximately 0.7 mW which is almost three orders of magnitude lower than the applied electric heat power. Hence the cooling of the device due to the liquid flow can be neglected.

Heat can also be dissipated from the separation chamber by conduction through the glass at the bottom of the device to the environment. The heat transfer power ($P_T[W]$) can be calculated as:

$$P_T = \kappa_T A \frac{\Delta T}{x} (C.2)$$

where $\kappa_T [W m^{-1} K^{-1}]$ is the thermal conductivity, $A [m^2]$ is the total area of that heat flows through, in our case the size of the separation chamber, x [m] is the heat transfer distance (*i.e.* glass thickness) and $\Delta T [K]$ is the temperature difference between the two sides of the glass. If we equate the Ohmic heating power to the heat transfer power and assume that the temperature of the side of the glass to the environment is constant we calculate a temperature increase in the liquid of 0.95 K compared to the environment (using κ_{T,SiO_2} =1.05 W m⁻¹ K⁻¹), proving that passive cooling of the device is sufficient for typical operation potentials. Nevertheless, higher throughputs in combination with highly conductive samples may require active cooling or the use of materials with higher thermal conductivity compared to glass such as silicon.

1.2| Electrochemical reactions

The electric current applied to the system is transformed to an ionic current via electrochemical reactions on the surface of the electrodes, whereby gaseous products can be formed (O_2 , Cl_2 , H_2). Water electrolysis thereby changes the pH of the solution. The rate of formation of H⁺ and OH⁻ (assuming water electrolysis is the only reaction taking place) ($j \ [mol \ s^{-1}]$) can be simply calculated via Faraday's law:

$$j = \frac{I}{Fz_i}(C.3)$$

where I[A] is the electric current and $F[C mol^{-1}]$ is the Faraday's constant. A current of 100 µA will add approximately 1 nmol s⁻¹ of H⁺ to the anodic reservoir. The smaller the reservoir, the stronger the effect of the added acid on the local pH. In our device we therefore used large reservoirs of 1 ml. The application of 100 µA for 1000 s will cause a pH reduction of 0.81 and 0.19 in 0.1xPBS and 1xPBS, respectively.

2 Original Microscopy images

In the following figures the original fluorescent microscopy images are shown upon which the analysis of the article is based.



Figure ESI1 - Unedited fluorescent microscopy images upon which the analysis for peak mode was based for article, Figure 2.



Figure ESI2 - Unedited fluorescent microscopy images upon which the analysis for plateau mode was based for article, Figure 3.



Figure ESI3 - Unedited fluorescent microscopy images upon which the analysis for the electrophoretic spacers was based for article, Figure 5.

3| Video description

ESI_plateau.avi – Demonstration of plateau mode in FF-ICPF of 1mM BDP(green) 1mM AF647(red) and 1mM CB(blue) in 0.1xPBS. Actuation potential : 200V. Flow rate: 5μ l min⁻¹ (In real time)

https://youtu.be/gKgBbUgRXVQ

ESI_plasma.avi – Demonstration of plateau mode in FF-ICPF of 100µM BDP(green) in undiluted blood plasma. Actuation potential : 120V. Flow rate:5µl min⁻¹ (In real time) https://youtu.be/yUZXsSJzu8Y