PRINTING NANOBIOLOGY IN AQUEOUS SYSTEMS

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Water is the major constituent of living organisms and plays a central role in the daily lives of humans. From a chemical point of view, water is associated with unique physical-chemical properties which make it the universal solvent for biological systems [1]; it is therefore important to study the active role of water in the technologies aimed at the realization of functional devices in medical and biotechnological field.
Our studies in the field of printing nanobiology in aqueous solution are proposed to highlight the role of water in the processes of interaction between biomolecules in drug-screening devices fabricated by bioprinting technologies and to emphasize the influence of water evaporation on the diffusion of molecules in droplets of picoliter-scale.
In this regard, we already showed the possibility to use a low-cost and miniaturized drug screening methodology based on direct bio-printing like Dip Pen Lithography (DPL) [2], and non-contact patterning methods, such as Inkjet Printing, to dispense biomolecules under microarrays format without affecting biological activity at solid-liquid interfaces [3] or in picoliter-scale droplets [4].
Accordingly, we carried out studies by the fluorescent probe Alexa 647, dispensed as liquid droplets in picoliter-scale using Inkjet Printing methods, by finely tuning deposition parameters and aqueous ink formulation. To evaluate effects of solvent evaporation, Alexa solutions are spiked with variable quantity of glycerol from 10% to 80 % v/v. We used fluorescence confocal microscopy to quantify fluorescent probe behavior by means of fluctuation techniques that permit mapping concentration and diffusion coefficients of fluorolabeled or fluorescent molecules at nanomolar concentration.
We also showed nanoprinting of DNA oligonucleotides in order to characterize the dynamics of hybridization of DNA sequences in aqueous solution, by printing oligonucleotides with DPL in form of drops of picoliters volumes on a solid substrate.
Having previously shown the possibility to efficiently deposit oligonucleotides onto glass surfaces [2], here we deposited oligonucleotides on nylon substrate for the fabrication of low cost point of care biochips. Accordingly, we optimized oligonucleotides printing on nylon substrate, obtaining efficient deposition at 10 - 1μM concentrations, 70% relative humidity and 30% glycerol v/v. The printed oligonucleotides were then hybridized with a fluorescence-labelled complementary probe to detect and quantify DNA hybridization after DPL deposition. Remarkably, the fine tuning of the printing conditions, such as humidity, tip-surface dwell time, glycerol percentage of molecular ink, are all essential parameters to facilitate the printing process, so limiting the effect of evaporation of the water and obtain regular, circular spots.

References

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