

Study of adherent cell permeabilization in various Plasma Gun plasma delivery protocols

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Plasma discharges have been known for quite a long time as a method to allow for cell permeabilization and likely to involve electric fields, UV photons and charged or excited particles, e.g.[1]. Comparison of different plasma devices, including so called "cold atmospheric pressure plasma" based on dielectric barrier discharge, but also combined role of electrical and chemical factors were more recently investigated with their potential interest as a new method for cell transfection [2]. In a very recent paper [3] not only radical produced by the plasma source but also intense electric field, as high as 150 kV/cm delivered on adherent cells are reported to induce transient increase in intracellular calcium concentrations, thereby playing a key role in cell response to plasma stimulus [3].

In this work, we report on the use of plasma jet, so called Plasma Gun developed at GREMI, as an innovative drug delivery technique for adherent, HeLa and 4T1 cells. The role of different plasma source operation parameters but also plasma delivery protocols has been studied. Propidium iodide and FITC-Dextran were used as permeabilization markers. Cellular uptake was assessed by fluorescence microscopy and flow cytometry. Cell viability and reactive oxygen and nitrogen species produced were measured. Percentage of propidium iodide positive cells was the highest for 1,000, 10,000, and 100,000 pulses corresponding to 100 s-long treatments [4]. We found that cellular uptake was more efficient after a 30-minute incubation time at 37°C. Drug uptake kinetics was also investigated showing that surprisingly best permeabilization levels were achieved when plasma treatment is processed a few minutes before and not simultaneously with the drug injection in the culture medium. Transient permeabilization following plasma treatment was measured for longer delays for drug injection also confirming the non-toxic nature of plasmas when a limited number of pulses are delivered. The specific role of plasma induced transient electric fields is under investigation to try to elucidate not only the plasma mode of action but the ring shape permeabilization patterns, already previously reported by other groups [5], as well.

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