## Comparison of the effect of cold atmospheric plasmas on mammalian and bacterial cells

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In the application of non-thermal plasmas to wound treatment both skin and bacterial cells are affected. This study presents a comparison between the behavior of different mammalian and bacterial cell types, on exposure to weak atmospheric plasmas. Three different types of mammalian cells were investigated – SW480, Chinese hamster ovary (CHO) and osteoblast (MG63) cells. The behavior of these cells when exposed to the plasma was compared with the bacterial cells *Staphylococcus aureus*, Coagulase-Negative *Streptococci* and *Pseudomonas aeruginosa*. A further objective is to quantitatively compare how treatment duration and frequency affect the cell response.

The plasma treatments were carried out using the helium plasma jet system shown in Figure 1. This is generated using a dual-pin parallel tungsten electrode source powered using a C2000 Redline generator. The quartz reactor cylinder has a length of 6 cm and nozzle exit area of 2 cm2. In this study the treatment frequency was varied systematically between 150-450 kHz. Due to the differences in the relative size of the mammalian and bacterial cells, the



Figure 1 Helium atmospheric Pressure Plasma Jet system

cell concentrations exposed to the plasma were 2x 10e5/ml in the case of the mammalian cells and 10e9/ml for the bacterial cells. Exposure to the plasma was carried out either directly on polystyrene 6-well plates or by passing the cells through the plasma jet. This was achieved by passing droplets of the suspensions into the plasma using a pneumatic nebulizer. The effect of the plasma treatments was evaluated with respect to cell number, viability, cell cycle, ATP and apoptosis. Lactate dehydrogenase measurements were used to quantify plasma membrane damage of sheared cells.

Passing the cells through the nebulizer system even in the absence of the plasma was found to give rise to a significant level of cell death. This may be due to the harsh mechanical effect of the nebulisation process on the cells. The lethal effect was correlated with the nebulized droplets size (typically in the 3 to 5  $\mu$ m range). The plasma intensity was assessed using emission spectroscopy and the cell treatments were carried out at the frequencies which yielded intensity maxima and minima within the 150-450 kHz range investigated. As expected cell death increased with increased plasma exposure times, with apoptosis is the predominant mechanism in mammalian cells. The level of ATP in CHO cells following plasma treatment was increased after incubation of 24 hours. It is suggested that the increased ATP following treatment might play a major role in energy provision when cellular repair processes are able to operate. The different sensitivity of mammalian and bacterial cells may be related to different cellular and molecular mechanisms in cell types. These findings suggest that such difference in sensitivity could be clinically exploited to improve the use of plasmas in wound healing.

Acknowledgment: SFI grant No. 08/SRC/11411