

IJAE Vol. 115, n. 1/2 (Supplement), 2010

## Effects of cold plasma on pathogenic microorganisms, cells and corneal tissues

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**Background** Low temperature plasma is a partially ionized gas that has long been considered an important tool for material processing and sterilization.

**Aim** The objective of the present study was to evaluate the effects of plasma on different microorganisms, cells and corneal tissues.

**Methods** A prototype of a plasma source has been developed and its effects on different microorganisms and on conjunctival fibroblasts and corneal keratocytes were assessed at different exposure times. Microbial viability was expressed in CFU, the antiviral effect by analyzing the plaque forming units developed after infection of Vero cells and viability of human cells evaluated with MTT test. ROS production was analysed using the 2',7'-dichlorodihydrofluorescein test. The micronucleus test was used to ascertain induction of DNA damage in human cells. Human corneas were also infected *ex-vivo* with bacteria (*P. aeruginosa* or *S. aureus*) or virus (HSV type 1) and treated with plasma. Bacterial CFU or viral plaques were then counted and histology observed. Apoptosis and UV damage of tissues exposed to plasma were also analyzed with the TUNEL test and immunohistochemical analysis, respectively.

**Results** Application of plasma for several minutes caused a significant reduction of microbial viability. Conversely, treatment of human cells did not cause any significant reduction of viability. No effect on HSV vitality was observed. High levels of ROS were found in both microorganisms and human cell cultures. Plasma application to infected corneas caused a significant reduction of microbial viability with no evidence of morphological damage. No induction of thymine dimer formation nor apoptosis was observed in treated tissues. Conversely, a significant increase in the frequency of micronucleated cells was found at all treatment times.

**Conclusion** Cold plasma reduced the viability of microorganisms but not that of human cells. The measurable effects caused by plasma consisted in high ROS production and increases of micronucleated cells. Further study is necessary to investigate the evolution of DNA damage in treated cells.