IJAE
Vol. 116, n. 1 (Supplement): 34, 2011
ITALIAN JOURNAL OF ANATOMY AND EMBRYOLOGY

Disinfection of ocular cells and tissues by atmospheric-pressure cold plasma

P. Brun[1], P. Brun[2], M. Vono[1], M. Zuin[3], E. Martines[3], G. Abatangelo[1], W. Giuriati[1], I. Castagliuolo[2], A. Leonardi[4]

[1] Department of Histology, Microbiology and Medical Biotechnology, University of Padua, Italy, Histology Unit
[2] Department of Histology, Microbiology and Medical Biotechnology, University of Padua, Italy, Microbiology Unit
[3] Consorzio RFX, Euratom-ENEA Association, Padua, Italy
[4] Department of Neuroscience, Ophthalmology Unit, University of Padua, Italy

Low temperature plasmas have been proposed in medicine as agents for tissue disinfection and have received increasing attention due to the frequency of bacterial resistance to antibiotics. Our previous studies (1) demonstrated that atmospheric-pressure cold plasma (APCP) generated by a new portable device that ionizes a flow of helium gas inactivated ocular pathogens in vitro.

This study explored whether APCP inactivates ocular pathogens without causing significant tissue damage.

We tested the APCP effects on cultured Pseudomonas aeruginosa, Staphylococcus aureus, A. fumigatus, ocular cells (conjunctival fibroblasts and keratocytes) and ex-vivo cornea. Exposure to APCP for 0.5–5 min significantly reduced microbial viability (colony-forming units) but not human cell viability (MTT assay and Tunel analysis). Since our previous study indicated that exposure to plasma increases intracellular reactive oxygen species (ROS) production, ROS levels in APCP exposed microorganisms and keratocytes were analyzed by 2′,7′-dichlorofluorescein diacetate (HDCF-DA) fluorescence. The potential genotoxic effects of plasma on cells and tissues were evaluated by analyses of thymine dimers (TD), genes and proteins involved in DNA damage and repair (OGG1, GPX, NRF2) at set time intervals. High levels of intracellular reactive oxygen species (ROS) were found in exposed microorganisms and cells. Immunoassay confirmed no induction of thymine dimers in corneal tissues. Conversely, a transient expression of genes and proteins recruited following oxidative stress was determined in ocular cells and corneas by qRT-PCR and Western blotting.

In conclusion, a short application of APCP appears to be an efficient and rapid ocular disinfectant with ROS production likely causing pathogen killing and no substantial effects on ocular cells and tissues. The same APCP treatment to conjuntival fibroblasts and keratocytes caused a time-restricted formation of ROS and a change in some stress-response genes.

Keywords: trattamento al plasma, ingegneria tessuti, infezioni, ROS