Atmospheric surface micro-discharge air plasma disinfection against *Clostridium difficile* spores

<u>Tobias G. Klaempfl^{1,3}</u>, Yang-Fang Li¹, Sylvia Koch², Tetsuji Shimizu¹, Julia L. Zimmermann¹, Jürgen Schlegel³, Jürgen Gebel², Gregor E. Morfill¹, Hans-Ulrich Schmidt⁴

¹ Max Planck Institute for Extraterrestrial Physics, Garching, D-85748, Germany
² Association for Applied Hygiene, Bonn, D-53127, Germany
³ Department of Neuropathology, TU München, Munich, D-81675, Germany
⁴ Department of Microbiology, Hospital Munich Schwabing, Munich, D-80804, Germany
⁵ E-mail: klaempfl@mpe.mpg.de

Introduction: According CDC *Clostridium difficile* is a germ that causes diarrhea and other intestinal problems linked to 14,000 U.S. deaths annually. *C. difficile* infections cost at least \$1 billion in extra health care costs at all types of medical facilities including hospitals, nursing homes, and outpatient facilities annually. The transmission of highly-resistant *C. difficile* spores occurs fecal-orally *via* contaminated surfaces in the environment and directly *via* the hands of the personnel. According to the EPA-registered product list, there is no specific disinfectant for the inactivation of *C. difficile* spores [1].

Cold atmospheric plasmas (CAP) are under investigation as promising gas-borne disinfectants or sterilants [2]. Recently, Tseng et al. used a helium radio-frequency cold plasma jet to show the sporicidal effect against *C. difficile* [3]. However, this device cannot be used for surface disinfection of large areas. We are confident that the surface micro-discharge (SMD) plasma technology - a scalable and robust technology - has this capability.

Method: We evaluated the disinfecting effect of SMD air plasma on spores of *C. difficile* NCTC 13366. Spores of *Bacillus subtilis* ATCC 6633 as well as vegetative bacteria of *Enterococcus faecium* ATCC 6057 served as references. Initially 10^6 spores or 10^8 vegetative bacteria were inoculated on dry stainless steel test specimen ($30 \times 6 \text{ mm}^2$) either with or without a small burden of 0.03 % serum albumin and wrapped in Tyvek. These samples were CAP-treated inside a small box (FlatPlaSter2.0) with ~35 mW/cm² plasma power (1 kHz, 10 kV_{pp}, sinusoidal wave form). Furthermore different CAP treatment times and open/closed volume conditions were used. The CAP-treated samples were microbiologically analyzed according to disinfection testing standards.

Results: Survival Curves and decimal reduction values revealed that a rapid surface disinfection takes place for closed volume conditions $(10^3 \text{ spore reduction within few minutes})$. The role of diverse SMD air plasma species, particularly of oxidizing agents like ozone, is discussed in the microbial inactivation. Potentials and limitations of the technology in the field of disinfection are discussed.

Conclusion: Our study clearly showed that SMD air plasma can effectively disinfect dry surfaces contaminated with *C. difficile* spores and therefore, can serve as an alternative disinfection method. Further investigations simulating the clinical practice are necessary.

References

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