## Inactivation of spores on surfaces using nitrogen cold atmospheric plasma gas

## M.N. Nierop Groot<sup>1</sup>\*, T. Dongmin Kim<sup>1,2</sup>, A. Warda<sup>1,2</sup>, T. Abee<sup>2</sup> and H. Mastwijk<sup>1</sup>

<sup>1</sup>Food and Biobased Research, Wageningen UR, The Netherlands <sup>2</sup>Laboratory of Food Microbiology, Wageningen University, The Netherlands \*masja.nieropgroot@wur.nl

## Abstract

Bacterial spores are resistant to severe conditions and form a challenge to eradicate from food or food packaging material. Currently applied heat and chemical based methods for food packaging have the disadvantage that it not applicable on thermo-sensitive material such as some biodegradable polymers and require a subsequent step to remove residual concentrations of sterilants on packaging material.

Cold atmospheric plasma (CAP) treatment is receiving more attention as potential low temperature, non-chemical based sterilization technology. Cold plasma targets both vegetative cells and spores, but effectiveness depends on plasma parameters such as design of electronic power circuits, carrier gas, but also characteristics of the microbe. Understanding of the antimicrobial mode of action will be a prerequisite for future regulatory status of non-thermal plasma application.

The objective of this study is to determine the efficiency of nitrogen cold atmospheric plasma gas (NCAP) for decontamination of bacterial spores on surfaces. *Bacillus cereus* ATCC14579 spores were used as target to obtain insight in inactivation mechanisms using non-nutrient germination triggers such as high pressure and Ca-DPA and by using a targeted mutants in regulators involved in stress response. In addition, spores of 15 different *B. cereus* strains previously isolated from food and two reference strains (ATCC14579 and ATCC10987) were tested for inactivation by NCAP.

Spores of all 17 tested *B. cereus* strains could be inactivated by NCAP although substantial diversity in sensitivity was observed between the different strains. Notably, these resistance characteristics did not correspond to resistance to heat and/or hydrogen peroxide in line with previous observations (Van Bokhorst-van de Veen et al., 2015). Plasma treated spores were subjected to a subsequent treatment with high pressure at 150 Mpa and 500 MPa to substantiate if only the germination machinery could have been inactivated. High pressure did not trigger germination and outgrowth of NCAP treated spores, in contrast to non NCAP treated spores.

Data presented in this study provide more insight in impact of strain diversity aspects on NCAP effectiveness and provide new leads for its mechanism of inactivation.

## Reference

Bokhorst-van de Veen, H., Xie, H., Esveld E., Abee, T., Mastwijk, H., and M. Nierop Groot (2015). Inactivation of chemical and heat-resistant spores of *Bacillus* and *Geobacillus* by nitrogen cold

atmospheric plasma evokes distinct changes in morphology and integrity of spores. Food Microbiology 55: 26-33.