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Bactericidal activity against *Listeria* spp using Plasma Activated Water

Ana Sainz-García¹, Elisa Sainz-García¹, Félix González-Gallarta², Alpha Verónica Pernía-Espinoza¹, Rodolfo Múgica-Vidal¹, Ignacio Muro-Fraguas¹, Ana González-Marcos¹, Paula Toledano³, Yolanda Sáenz³, Fernando Alba-Elías¹ rod ana paula

¹Department of Mechanical Engineering, University of La Rioja, La Rioja, Spain

²Department of Chemistry, University of La Rioja, La Rioja, Spain

³Center for Biomedical Research of La Rioja (CIBIR), La Rioja, Spain

This work studies the effect of Plasma Activated Water on bacterial inactivation using different hole-setting (number and distribution). A fluid simulation software has been used to know the size, shape and behaviour of bubbles for each hole-configuration.

1. Aim of the work

According to the Food and Agriculture Organization of the United Nations (FAO) the concept “One Health” has been implemented in medical and agri-food fields during the past years. The objective is to balance and optimize the health of ecosystems, animals and humans since they are closely linked [1]. In this regard, there are bacteria such as *Listeria* spp. that could inhabit in all of this field and cause several problems; for instance, food-borne illnesses [2].

This work studies Plasma Activated Water (PAW) generated by an atmospheric pressure cold plasma jet system with dielectric barrier discharge. The PAW used was generated by bubbles method (b-PAW). For generating b-PAW, two gas flows (60-80 slm) and several 3D printed pieces with different hole-configurations (number and distance among holes) were used. *Listeria* spp was chosen as the target bacterium, microbiological experiments were carried out after putting in contact each b-PAW with 10⁸ CFU/mL (30 min, 60 min, 2 h, 4 h, 6 h and 24 h). The general objective was to know which b-PAW reached the higher inactivation against *Listeria* spp. However, that objective was divided into two specific ones. The first one was related to how the number of holes (12 or 48) affects the bactericidal activity of b-PAW and the second one related to which are the effects of the distance among holes in terms of bacterial reduction (symmetrical or random hole-configuration). Chromatographic, UV-vis and colorimetric methods were used for reactive species characterization (NO₂^{*}, NO^{*}, OH^{*}, NO₂⁻, NO₃⁻). Moreover, other parameters such as temperature, oxidation-reduction potential (ORP), electrical conductivity (EC) and pH were also analysed.

Finally, in order to understand inactivation results, bubbles simulations were performed using a fluid-dynamic software (Ansys-Fluent).

2. Results of the work

In terms of bactericidal activity, after 24 h of contact (b-PAW/bacterium) total inactivation was reached regardless the b-PAW studied.

Regarding the first objective, it was suggested higher bacterial reductions when the 3D piece with the maximum number of holes was used. That could be explained due to the fact that the higher the number the holes, the wider the surface interaction air-water when generating b-PAW. It was also showed after fluid dynamic simulation a stronger turbulence with 48 holes compared to 12 holes which is known to increase reactive species diffusion. On the other hand, the research suggested a better bacterial inactivation when using an equal distance among the holes. In this case, simulation illustrated bubble aggrupation after random distances among holes. Besides, since the latest setting reduced the surface interaction air-water the inactivation achieved was lower.

3. Acknowledgments

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