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Biofilm thickness and biomass reduction after treatment with DC air corona discharges

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Bacteria on surfaces exist predominantly in a form of biofilm, in which microorganisms are concentrated at an interface and encased in a hydrated matrix of exopolymeric substances (EPS). Bacteria in the biofilm are protected from harsh conditions, and therefore are much more resistant than their planktonic counterparts. In order to avoid using toxic chemicals or high concentrations of antibiotics to achieve the desired decontamination efficiency in biofilms, a search for new alternative methods of decontamination is required; one of these is low-temperature plasma.

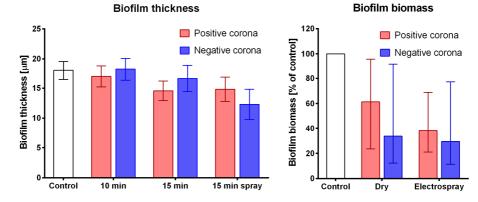


Fig. 1 Left: The biofilm thickness calculated from DAPI fluorescence integrated over the entire biofilm thickness (plotted mean \pm SEM, 8 \leq n \leq 16 in each group). Right: The biofilm biomass calculated as percentage of controls (100%) from crystal violet absorbance after 15 min positive and negative corona treatment with or without water electrospray. (Median with IQR, n = 10 in each group).

DC corona discharges in air with or without water electrospray were used to decontaminate 48-hour biofilm of *Escherichia coli* BW 25113 grown on glass cover slides [1]. Thermostatic cultivation showed up to $5 \log_{10}$ reduction in bacterial concentration within 15 min exposure to the plasma. Water electrospray enhanced the biocidal effect of discharge by $1 \log_{10}$. Confocal laser scanning microscopy of biofilm dyed with DAPI showed thinning of the biofilm (Fig. 1, left) with increasing time of the plasma treatment and adding the water electrospray. The thickness was reduced from 18.1 µm to 12.3 µm after negative corona treatment with water electrospray. The Crystal violet assay for biofilm biomass evaluation showed decrease in the attached biomass after plasma treatment (Fig. 1, right), 61.3% and 32.7% of the biomass remained after exposure to positive and negative corona, respectively. With water electrospray, stronger biomass detachment occurred and only 38.1% and 29.5% of the biomass was attached after positive and negative corona treatment.

Low-temperature plasma has shown the ability to kill biofilms and reduce their thickness.

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[1] Z. Kovalova, et al., *Bioelectrochemistry* (in press 2016).