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<u>Expertise</u>

## Effect on biofilms of chlorine dioxide dosed at 0,2 mg/L by degerming units "Aquacon WH01" and "Aquacon CLO2"

Biofilms in the drinking water-distribution systems could release high concentrations of microorganisms to the streaming water. We tested the effect of chlorine dioxide dosed at 0,2 mg/L by the degerming units "Aquacon WH01" and "Aquacon CLO2" on the biofilms and the levels of the released free floating microorganisms in water. The system used for the tests was the silicone tube model, which has been developed for this type of testing by the Institute of Hygiene and Public Health in Bonn.

The test system consisted of silicone tubes, 4 mm in diameter, through which treated and/or untreated water flowed. The silicone tubes themselves were either fresh ones with no biofilm attached to the inner surface, or older ones which had been plugged in to the water system for several years and which contained well developed biofilms on their inner surface. The presence of microorganisms in the biofilms or of those released from the biofilms to the flowing water was tested for by standard microbiological cultivation methods. The presence and appearance of the biofilms was tested for by means of a scanning electron microscopy.

The final concentration of chlorine dioxide of 0,2 mg/L was that prescribed by the "Drinking Water Regulation". The concentrations of microorganisms in the biofilms were determined in terms of <u>Colony Forming Units</u> (CFU) per unit area (CFU/cm<sup>2</sup>), while the concentrations of the microorganisms

washed out from the biofilms to the flowing water were determined in terms of CFU/cm<sup>2</sup>/sec or CFU/mL. The samples were collected once a week over the period of 70 days.

The washout of bacteria from mature biofilms washed in untreated water displayed the average level of 1450 CFU/mL. After switching the system to the chlorine dioxide-treated water the level of washout dropped to zero.

The presence of chlorine dioxide in the water also displayed the same effect on the levels of microorganisms in the biofilms, albeit over a longer time scale. After 70 days of continuous wash of the biofilms in the treated water the level of microorganisms in the biofilm dropped from the original 10.000.000 CFU/cm<sup>2</sup> to zero. In the tubes with the same biofilm but washed in untreated water the washout remained at 10.000.000 CFU/cm<sup>2</sup> over the whole test period. In the fresh silicone tubes with no biofilm at the beginning of the test period, the effect of washing in with the treated water was a complete suppression of the biofilm formation. When the same fresh silicone tube was washed in untreated water, a biofilm was formed with the level of microorganisms at 388.000 CFU/cm<sup>2</sup>. All these findings were confirmed by the scanning electron microscopy.

Figure 1 shows the time dependence of the microorganism content in biofilms washed in treated water. The analogous dependence for the silicone tubes, which had no biofilms at the beginning of the test period, is shown in Figure 2.



Figure 1. Comparison of the time dependence of the microorganism content in biofilms washed in treated and untreated water.



## Figure 2. Comparison of the time dependence of the microorganism content in silicone tubes washed in treated and untreated water, which had no biofilms at the beginning of the test period.

Supplementary suspension tests with bacterial colonies (*Legionella pneumophila, Pseudomonas aeruginosa, Escherichia coli* and *Enterobacter cloacae*) exposed to 0,2 mg/L of chlorine dioxide revealed that 5 minute contact time brought the level of bacteria under the detection level.

Chlorine dioxide has been known for many years as a proven and very efficient disinfectant. However, till now it was not possible to apply this disinfectant in the fields of drinking water, warm water and Legionella-containing water disinfection because no methods for monitoring concentration of chlorine dioxide (0,05-0,2 mg/L prescribed by the "Drinking Water Regulation") were available.

The levels of chlorine dioxide in water can now be precisely monitored with the device "Aquacon CLO2". The selective measurements are based on a patented "lotronic" method. Tests performed at the Institute of Technical Chemistry at the Hannover University confirmed the suitability of the lotronic method for an automated monitoring of the chlorine dioxide levels in water. In comparison to the common laboratory methods (DIN EN 12671), the lotronic method, when applied for measurements of concentrations relevant in the drinking water treatment (0,1-0,3 mg/L), displays a lower mean relative deviation. The testing of the efficiency of chlorine dioxide against biofilms and bacteria was made possible only after combining two new systems together, the reliable generator of chlorine dioxide, and the efficient monitor of the chlorine dioxide levels which satisfies the prescriptions of the "Drinking Water Regulation".

The performed test show that under the conditions of the performed tests and with the chlorine dioxide concentration level of 0,2 mg/L, the degerming units produced by "lotronic Elektrogerätebau GmbH" and consisting of the chlorine dioxide producing unit Aquacon WH01 and the chlorine dioxide

monitoring unit Aquacon CLO2, are effective in both the inactivation bacteria in biofilms, and the prevention of formation of new biofilms.

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