

## Persistence of Legionella antigen after chlorine based biocide treatments

Chlorine based biocides are routinely used in the management of Legionella bacteria within cooling towers, spas, hot-water systems and other high risk equipment for disinfection purposes. We wish to know how long Legionella antigen persists as a detectable residue after a biocide treatment has been applied, to determine when a positive antigen result implies the detection of live bacteria.

Disinfectants are employed normally as a part of the legionella risk assessment on most water systems: chlorine is most commonly applied to cooling towers as sodium hypochlorite solutions which are dosed into a water system either continuously at relatively low concentrations (typically up to 1 ppm free chlorine) with the purpose of preventing colonisation by Legionella and other bacteria, or at larger concentrations (sometimes up to 50 ppm) for short periods of time in response to an observed high bacterial count.

Another example is cold water systems fed from municipal water supplies, which will typically have free chlorine levels below <0.35 ppm. Hot water systems commonly have much lower chlorine levels (as chlorine is driven off by heating). Spas are normally treated with higher levels of chlorine, sometimes up to 5 ppm.

In order to determine the effect that chlorine based biocides have on the antigens detected by the hydrosense® Legionella Field Test™, a set of experiments were carried out with sodium hypochlorite solution as the oxidising agent.

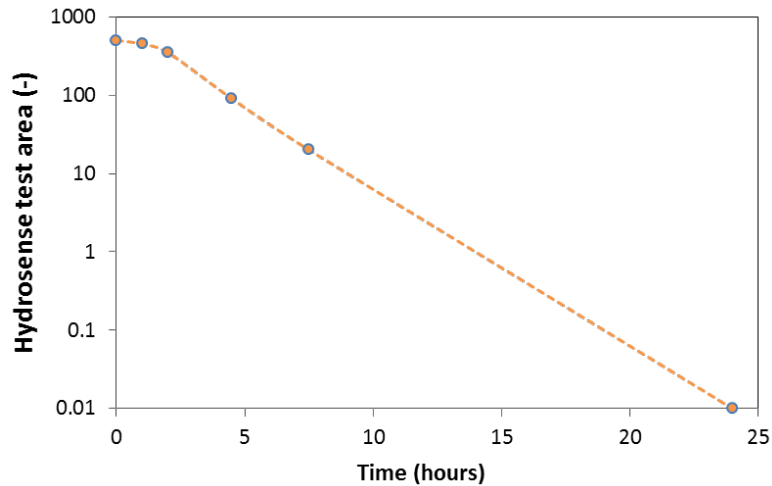
Table 1 shows the results of two levels of chlorine treatment: 0.9 ppm and 4.8 ppm. In both cases it was no longer possible to detect Legionella antigen after 20 minutes.

Time (minutes)	0.9 ppm Cl <sub>2</sub>		4.8 ppm Cl <sub>2</sub>	
	Culture (CFU/mL)	Hydrosense (area)	Culture (CFU/mL)	Hydrosense (area)
0	3000	434	3000	451
5	< 10	289	< 10	16
20	< 10	0	< 10	0
135	< 10	0	< 10	0

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Figure 1 shows the results when a strain of Legionella serogroup 1 (Philadelphia 1 ATCC 33152) with an approximate initial concentration of 3000-3500 CFU/mL is submitted to chlorination by adding sodium hypochloride.



**Figure 1. Reduction in antigen (Hydrosense reader signal) following exposure to 0.3 ppm free chlorine at 5<sup>o</sup> C. Visual limit of detection approximately 15-20 on the y-axis. The value after 24 hours is zero.**

From the graph in figure 1 we can observe that even with a low oxidising treatment of 0.3 ppm, the hydrosense<sup>®</sup> test result continues to fall gradually until no longer detectable by eye after 7 hours, and invisible even to the electronic Reader after 24 hours. Even at lower concentrations, and lower temperatures (which are known to impede the effectiveness of chlorine disinfection) the hydrosense<sup>®</sup> signal has disappeared within 1 day.

Conclusions:

**The hydrosense<sup>®</sup> test is a near real-time information tool on the effectiveness of chlorine based water treatment/disinfection programmes. A negative hydrosense<sup>®</sup> test result indicates the absence of antigen, which means that that the Legionella pneumophila serogroup 1 population is under control.**

**Traditional culture techniques require typically a 1-2 week delay in knowing the effectiveness of a disinfection cycle for Legionella control.**