

Using HY-LiTE for Biocide Efficacy Testing.



The method described can be used to demonstrate the speed and mode of action of different Biocides.

Materials:

HY-LiTE total ATP pens
 HY-LiTE free ATP pens
 Biocides for evaluation
 Disposable sample containers for HY-LiTE testing (e.g. Bijou, 2 ml Microtube or 1.5 ml microcentrifuge tube)
 Disposable plastic pipettes (optional)
 Clean Plastic bottles / jars for incubation of samples with biocide

Equipment:

(optional) Incubator at appropriate temperature e.g. 35°C

Method:

Prepare appropriate stock dilutions of Biocide to be tested, e.g. if a biocide is to be tested at 25, 50 and 100 ppm, prepare stock solutions at 100 x these concentrations.

Collect a sample of White Water to be tested. This should preferably give HY-LiTE readings between 5000 and 50000 RLU.

- Mix well and subdivide into 4 aliquots of 100 ml. Label these as Control, 25, 50 and 100 (or as appropriate)
- To sample Control, add 1% of water (same as used to dilute biocide). Mix well. Draw subsamples and perform test of total and free ATP. Record as Control, Time = 0.
- To each of the other samples add 1 % of appropriate stock solution. Mix well. Start stopwatch. Place at appropriate temperature.
- At appropriate time intervals (e.g. 5, 15 and 30 minutes after Time = 0), mix each of the "test" samples and with a pipette, transfer sub-samples to two separate containers for testing with Total respectively Free ATP pens. Return remaining sample to incubator (if used) and leave until next test time.

Note: Total and Free ATP pens must not be dipped in the same sample container!

Results:

Biocide type:

Temperature:

		Concentration*							
		Control (0ppm)		25 ppm		50 ppm		100 ppm	
		Total (RLU)	Free (RLU)	Total (RLU)	Free (RLU)	Total (RLU)	Free (RLU)	Total (RLU)	Free (RLU)
Contact time (min)*	0								
	0								
	5								
	5								
	15								
	15								
	30								
	30								

* Contact times and Biocide concentrations shown are only examples and may need adjusting.

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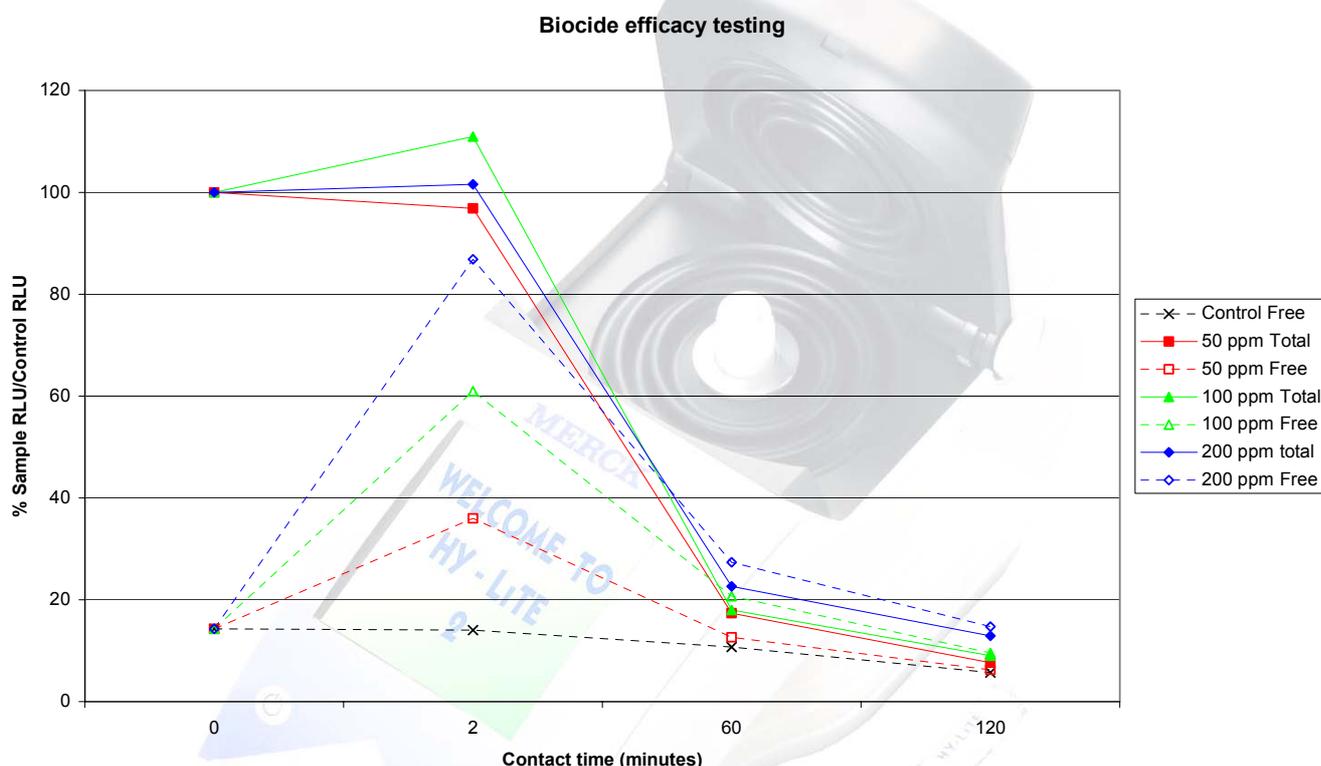


Calculate the RLU for Control (Free) and all other samples as % of Control (Total), and plot this against the contact time (c.f. Fig. 1).

Fig 1:

Example of results of Biocide efficacy test.

The biggest relative change (on short contact time) is seen for 200 ppm Biocide (Free ATP), indicating that this is the most effective dose.



Comments:

It is recommended that all ATP testing be performed in duplicate.

If several concentrations of Biocide are tested, the start of each "test" sample should be staggered by staggering the addition of biocide, to obtain accurate contact. Alternatively, the actual contact times must be recorded.

Concentrations and contact times will vary between different Biocides.

ATP measurements essentially measure bacteriostatic effects, whereas Viable Count determinations measure only bactericidal effects. The results will therefore not necessarily correlate with Viable Counts.

Please note that the effect of biocides on planktonic (free-swimming) micro-organisms are often much higher than on micro-organisms in a Biofilm. If biofilms are present in the system, the concentration of Biocide may therefore need to be adjusted accordingly.