Report ME-16-011v2

Effect of the HydroFlux Bracelet on Water Cooling Tower with Reduced Biocide Use

prepared for
HydroFlux Technology, LLC

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Approved for release on 10/13/16 by:

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Introduction

This study aimed to evaluate the effect of the HydroFlux bracelet on microbe growth control and possible reduced use of chemical biocides. A cooling tower at Qualcomm, which had a HydroFlux bracelet installed since February 2016, had its hypochlorite levels reduced starting on August 8th, 2016, leading to a complete shut-off of biocides (hypochlorite and glutaraldehyde) on August 22nd, 2016. The bacteria count and water chemistries were monitored closely from August 1st to September 16th. Water from another cooling tower without the HydroFlux bracelet and with normal levels of hypochlorite and glutaraldehyde, was sampled at the same time points as a control. The results of this study are summarized in this report.

Procedures

These experiments were carried out with the participation of multiple parties: HydroFlux provided the bracelet and commissioned the study, Qualcomm provided the test sites (located in San Diego, California), Fluor collected the water samples for analysis and monitored the cooling tower functions, Trident Technologies controlled the biocide levels and monitored the chemistries of the cooling towers, EnviroMatrix performed the analytical assays and Molecular Express managed the project, evaluated the data and prepared the summary report. Water samples were collected at approximately the same time of the day from the bracelet-installed tower and the control tower in the containers provided by EnviroMatrix, Monday through Friday. The water chemistries (*i.e.,* Aluminium, Calcium, Copper, Iron, Magnesium, Manganese, Silicon, Sodium, Zinc, Chloride, Fluoride, Nitrate, Nitrite, Orthophosphate, Sulfate, Hardness, Specific Conductance, Alkalinity, pH, Total Organic Carbon and Total Suspended Solids) and bacteria count (by Heterotrophic Plate Count) were analyzed on Mondays, Wednesdays and Fridays; while Heterotrophic Plate Count only was performed on Tuesdays and Thursdays. The first water sample was collected on August 1st. The study was designed to reduce the sodium hypochlorite level incrementally from 50% (on August 1st), to 25% (on August 8th), to 0% (on August 22nd) of the normal operation level at the bracelet-installed tower. The actual biocides pump operation records are shown in **Figures 1 and 2**, where it shows that the hypochlorite pump of the bracelet-installed tower only started to reduce on August 8th and was completely terminated by August 22nd; the glutaraldehyde was terminated by August 22nd. The control tower operated without reduction of biocides use during the experiment. The last water sample was collected and analyzed on September 16th, 2016.

Results

The water chemistries and Heterotrophic Plate Count results are summarized in **Tables 1-4.** The bacteria concentrations in the water towers over time is plotted in **Figure 3**. The water chemistries data are plotted as percentage change in **Figures 4-9**. Percentage change is determined by:

$$Percentage\ change = \frac{Value\ on\ sampled\ date}{Initial\ value} \times 100\%$$

The “initial value” is defined as the value on August 1st, except when the initial value is below the reporting limit, then the first data point that is above the reporting limit is used as the initial value. For plotting purposes, data points that are below the detection limits (see “ND” in **Tables 1-4**) are assigned a value of “0%”. Thus, a value of “0%” in **Figures 4-9** indicates that the value is below the reporting limit.

During the course of the experiment, the overall concentration of bacteria as determined by Heterotrophic Plate Count is about an order of magnitude (*i.e.*, 10 times) higher in the control tower than the tower with the HydroFlux bracelet installed (**Figure 3**). Although the bacteria count in the bracelet-installed tower increased on certain sampled days, the bacteria count did not increase exponentially (as expected for normal bacterial growth rates) after the biocide levels were decreased or terminated. The bacteria count in the bracelet-installed tower remained static without the use of biocides.

Most of the trends of the other measured parameters and concentrations are consistent between the control tower and the bracelet-installed tower, except for the concentration of nitrate which increased substantially in the bracelet-installed tower over time.

Conclusions

The goal of this experiment was to investigate the effect of the HydroFlux bracelet on biocide use. Since the bacteria count as measured by Heterotrophic Plate Count did not increase over time with decreased or no biocides, it indicates that the HydroFlux bracelet is as effective in controlling bacterial growth as the chemical biocide system used by Trident

Technologies. However, the Heterotrophic Plate Count method only accounts for live, free-floating, aerobic bacteria in the sampled water that would grow on the non-selective media, and does not account for other microbes, such as fungi, viruses, protozoa and algae.

On September 23rd, one week after the last sample was taken, algae growth was observed in the bracelet-installed tower (with no biocide) and action was taken on September 26th to reintroduce the biocides to this tower (operation record in **Figure 2** shows that hypochlorite was reintroduced on September 27th, and the glutaraldehyde pump remained off on September 27th until at least October 1st). A sample taken on the morning of September 26th yielded bacteria counts of 3,600 and 9,600 CFU/mL for the bracelet-installed tower and the control tower, respectively. This indicates that while the HydroFlux bracelet was effective against bacteria proliferation in this study, it may not control other microbe growth. The algae growth is likely correlated to the increase of nitrate in the bracelet-installed tower over time, since nitrate is one of the essential nutrients for algae. The cause of the increase in nitrate in the bracelet-installed tower is unclear. The nitrate level in the bracelet-installed tower (with normal biocide levels) measured in a previous experiment (February 20th to May 11th, 2016) was between 2.0 to 2.7 mg/L. However, in the study, the nitrate levels increased dramatically from ~1 mg/L to ~3 mg/L in 2 days, and then to ~4.5 mg/L in 3 weeks. A follow-up experiment of installing the bracelet on other cooling towers with reduced biocide levels will support the bacteriostatic properties of the bracelet, and confirm any correlation between the bracelet and the nitrate level. These follow-up experiments may identify the cause of the increase in nitrate as a single event such as contamination, or will indicate if the nitrate level needs to be controlled for the complete microbe control application of the HydroFlux bracelet, or if the selective use of algaecides in combination with the HydroFlux bracelet is necessary. In addition, it is possible that the HydroFlux bracelet may be efficient when used with reduced amounts of biocides, but not the complete elimination of the biocides, and subsequent experiments will aid in determining the optimal level or type of chemical biocides needed.

From this initial ground-breaking study performed on a fully functional cooling tower, it appears that the HydroFlux bracelet exhibited a bacteriostatic activity on the bacteria in the water for 4 weeks without the addition of biocides (except for chlorinated products in the San Diego potable water). The bracelet-installed tower consistently showed less bacteria than the control tower, despite continuous biocides treatment (hypochlorite and glutaraldehyde) of the control tower. The bacterial levels of the bracelet-installed tower were generally within the industry standard of <10,000 CFU/mL. In contrast, the bacterial load in the control tower was consistently above this standard. More impressively, this experiment was conducted in August, which is one of the warmest months in the year and thus, the time of highest potential microbial growth. Given these results, it would be reasonable to continue evaluating the bracelet's antimicrobial effect on additional towers of similar and dissimilar design (*i.e.*, stainless steel or wood) to further understand the utility of the HydroFlux bracelet. Future studies may include the evaluation of specific strains of bacteria that populate the bracelet-installed tower compared to the control tower(s). Given the health related importance of *Legionella pneumophila* in contaminated water cooling towers, an evaluation of the HydroFlux bracelet on a cooling tower known to be contaminated with *L. pneumophila* would be of high value.

Other potential positive effects of the HydroFlux bracelet, such as corrosion and scale control, is beyond the scope of this experiment. In order to limit the number of variables in this experiment, the chemicals used for corrosion inhibition (zinc phosphate) and scale control (sulfuric acid) remained constant for both the bracelet-installed tower and the control tower. A separate, well-controlled experiment is recommended to evaluate the effect of the HydroFlux bracelet on scale control, with the systematic reduced use of sulfuric acid, which may also have an effect on corrosion of the cooling tower. A related experiment, with the reduced use of zinc phosphate, can be followed. These experiments, in combination with the microbial control experiments, will help paint a complete picture of the full benefits of the HydroFlux bracelet.

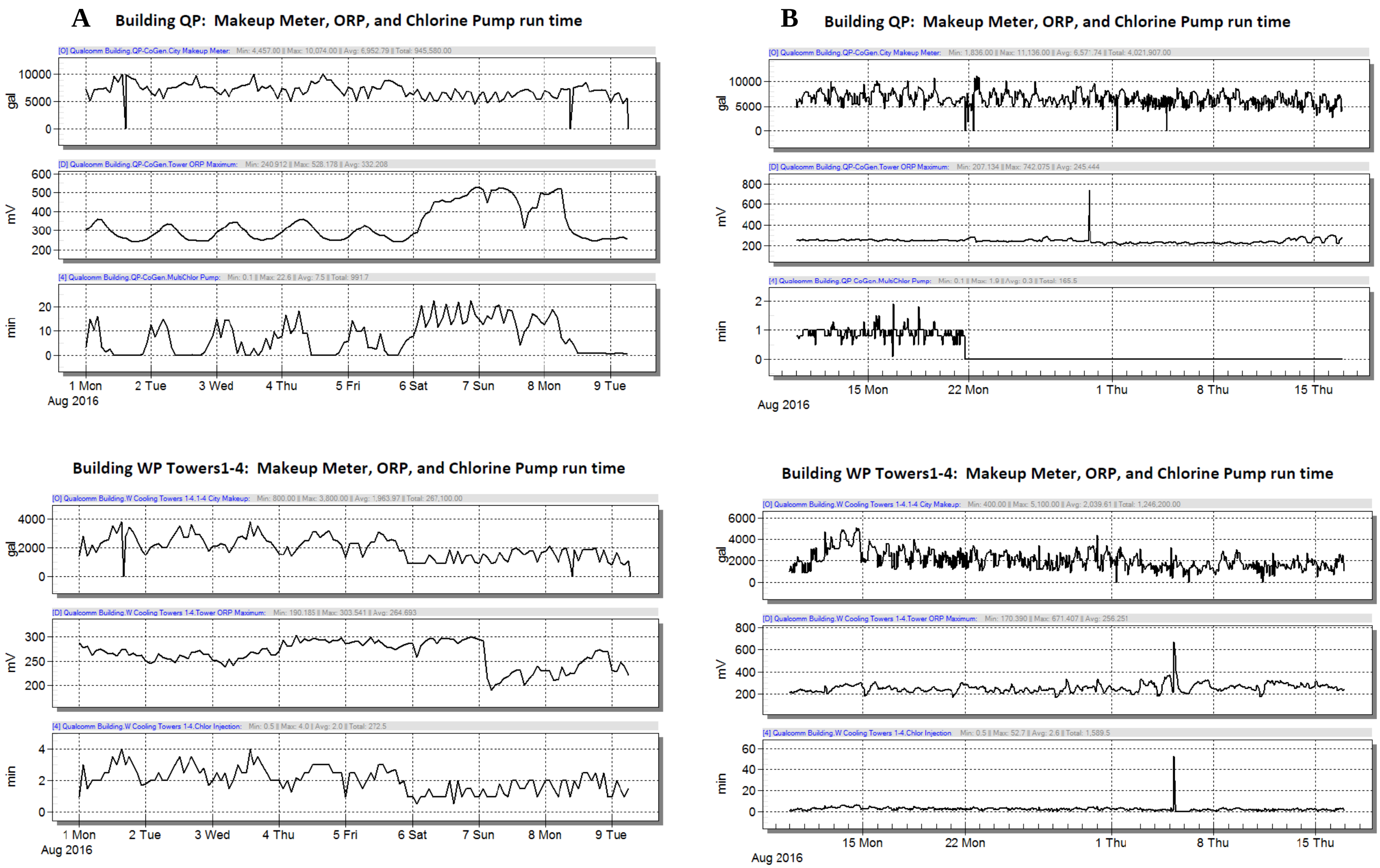

Figure 1. Cooling tower operation record from (**A**) August 1st to August 9th, and (**B**) August 10th to September 16th, 2016. The top three panels are record from the HydroFlux bracelet-installed tower, and the bottom three panels from the control tower. Record provided by Trident Technologies.

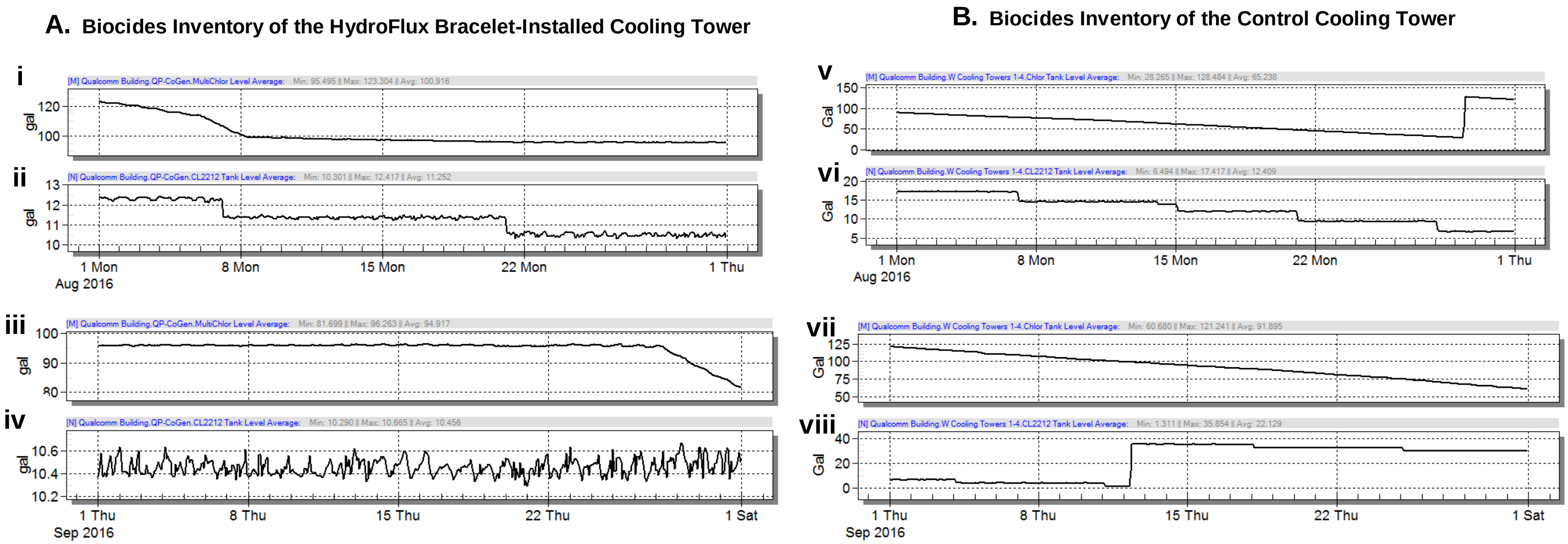

Figure 2. Biocides inventory of (**A)** the HydroFlux bracelet-installed cooling tower and (**B**) the control cooling tower. Panels (**i**), (**ii**), (**v**) and (**vi**) are records from August 1st to September 1st, 2016. Panels (**iii**), (**iv**), (**vii**) and (**viii**) are records from September 1st to October 1st, 2016. Panels (**i**), (**iii**), (**v**) and (**vii**) are records of hypochlorite inventory. Panels (**ii**), (**iv**), (**vi**) and (**viii**) are records of glutaraldehyde inventory.

Table 1. Anion Concentration Analyses Summary. Data of bracelet-installed tower is shown in blue and control tower in red.

Date		Chloride (mg/L)		Fluoride (mg/L)		Nitrate as N (mg/L)		Nitrite as N (mg/L)		Orthophosphate as P (mg/L)		Sulfate as SO_4 (mg/L)	
Reporting limit		0.05		0.10		0.10		0.05		0.25		250	
8/1/2016	M	570	760	3.20	3.17	1.01	1.44	ND	0.06	2.36	3.16	1490	1550
8/3/2016	W	580	570	4.02	3.92	2.99	2.19	ND	0.16	2.28	4.38	1680	1590
8/5/2016	F	550	550	3.36	3.30	3.31	2.61	ND	0.14	2.21	3.76	1580	1550
8/8/2016	M	590	560	3.36	3.26	2.20	1.08	ND	ND	2.04	2.60	1650	1500
8/10/2016	W	590	580	3.50	3.27	3.09	2.11	ND	0.07	2.54	2.88	1770	1520
8/12/2016	F	580	550	3.41	3.15	2.72	1.81	0.06	0.09	4.44	10.7	1870	1690
8/15/2016	M	540	550	3.32	3.16	2.95	1.02	0.07	ND	2.16	3.34	1710	1490
8/17/2016	W	560	550	3.39	3.21	3.12	1.10	0.08	ND	2.18	2.76	1640	1710
8/19/2016	F	550	540	3.33	3.14	4.52	1.67	0.08	0.06	2.34	2.76	1620	1640
8/24/2016	W	570	530	3.4	3.02	4.28	1.48	0.06	0.05	2.03	2.53	1670	1420
8/26/2016	F	580	540	3.46	3.09	2.72	0.75	0.06	0.08	2.20	2.90	1810	1490
8/29/2016	M	610	580	3.63	3.22	3.93	0.76	0.05	0	2.31	2.72	1830	1640
8/31/2016	W	520	580	3.25	3.13	3.44	1.49	0.05	0.05	2.26	2.68	1650	1630
9/2/2016	F	570	590	3.39	3.26	4.00	1.66	ND	0.08	2.40	2.96	1830	1690
9/7/2016	W	570	570	3.32	3.11	4.49	1.29	ND	ND	2.28	2.76	1850	1760
9/9/2016	F	600	590	3.15	3.01	4.41	1.45	ND	ND	2.58	2.72	1810	1710
9/12/2016	M	550	580	3.14	3.05	4.28	1.11	ND	ND	2.42	2.90	1800	1740
9/14/2016	W	560	540	3.54	3.16	3.64	1.05	ND	ND	2.57	2.52	1990	1690
9/16/2016	F	540	570	3.54	3.40	3.88	1.48	ND	ND	2.32	2.74	1990	1810

*ND: Analyte not detected within reporting limit.

Table 2. Metal Concentration Analyses Summary. Data of bracelet-installed tower is shown in blue and control tower in red.

Date		Aluminium (mg/L)		Calcium (mg/L)		Copper (mg/L)		Iron (mg/L)		Magnesium (mg/L)		Manganese (mg/L)		Sodium (mg/L)		Zinc (mg/L)		Silica as SiO_2 (mg/L)	
Reporting limit		1.00		5.00		0.050		0.100		5.00		0.030		5.00		0.500		0.50	
8/1/2016	M	ND	ND	362	341	ND	ND	ND	0.136	138	135	ND	ND	574	565	1.04	1.45	40	38
8/3/2016	W	ND	ND	355	352	ND	ND	ND	ND	137	134	ND	ND	554	550	0.884	1.13	42	38
8/5/2016	F	ND	ND	342	338	ND	ND	ND	ND	124	125	ND	ND	535	531	0.925	1.35	39	33
8/8/2016	M	ND	ND	342	330	ND	ND	ND	0.136	133	129	ND	ND	544	520	0.974	1.40	39	37
8/10/2016	W	ND	ND	380	354	0.051	0.052	0.052	0.11	149	139	ND	ND	577	580	1.14	1.38	37	34
8/12/2016	F	ND	ND	347	355	ND	0.056	0.056	0.205	135	139	ND	ND	552	557	0.898	1.36	38	36
8/15/2016	M	ND	ND	361	380	0.052	ND	ND	ND	141	151	ND	ND	516	562	1.04	1.08	37	35
8/17/2016	W	ND	ND	355	333	ND	ND	ND	ND	134	128	ND	ND	536	523	0.904	1.24	41	38
8/19/2016	F	ND	ND	331	313	ND	ND	ND	ND	126	118	ND	ND	519	506	0.977	1.12	40	38
8/24/2016	W	ND	ND	351	322	ND	ND	ND	0.115	139	125	ND	ND	530	509	0.795	1.03	42	37
8/26/2016	F	ND	ND	393	358	ND	ND	ND	ND	154	140	ND	ND	544	507	1.04	1.18	48	45
8/29/2016	M	ND	ND	366	313	0.051	ND	ND	ND	134	120	ND	ND	530	501	1.22	1.07	45	40
8/31/2016	W	ND	ND	329	335	0.042	0.038	0.038	0.116	127	128	ND	ND	504	558	0.941	1.08	39	38
9/2/2016	F	ND	ND	319	311	ND	ND	ND	0.141	120	119	ND	ND	510	525	0.864	1.09	43	40
9/7/2016	W	ND	ND	328	316	ND	ND	ND	0.134	131	121	ND	ND	540	535	0.939	1.22	44	39
9/9/2016	F	ND	ND	335	325	ND	ND	ND	0.115	130	127	ND	ND	552	535	0.956	1.16	47	45
9/12/2016	M	ND	ND	343	333	ND	ND	ND	0.123	131	128	ND	ND	519	558	0.944	1.19	46	45
9/14/2016	W	ND	ND	362	306	ND	ND	ND	ND	141	121	ND	ND	553	509	1.04	0.968	44	39
9/16/2016	F	ND	ND	336	328	ND	ND	ND	0.129	133	129	ND	ND	506	524	0.966	1.16	43	41

*ND: Analyte not detected within reporting limit.

Table 3. Water Analyses Summary. Data of bracelet-installed tower is shown in blue and control tower in red.

Date		Total Hardness (mg $CaCO_3$/L)		Total Alkalinity (mg $CaCO_3$/L)		Total Suspended Solids (mg/L)		Total Organic Carbon (mg/L)		Specific Conductance (µmhos/cm)		pH	
Reporting limit		100		5		20.0		3.0		1.00		0.10	
8/1/2016	M	1470	1410	81	67	ND	ND	16	18	3910	3950	7.99	7.94
8/3/2016	W	1450	1430	83	60	ND	98	16	16	4250	4130	7.98	7.74
8/5/2016	F	728	1360	81	67	ND	28	15	16	4180	4100	7.94	7.84
8/8/2016	M	1400	1360	98	86	ND	ND	16	19	4190	4080	8.00	8.04
8/10/2016	W	1560	1460	105	78	ND	ND	NA	16	4340	4120	8.14	8.00
8/12/2016	F	1420	1460	91	74	ND	22	22	NA	4310	4100	8.20	8.11
8/15/2016	M	1480	1570	86	112	ND	48	15	26	4130	4090	8.08	7.75
8/17/2016	W	1440	1360	89	91	ND	ND	16	16	4260	4190	8.00	8.05
8/19/2016	F	1350	1270	83	71	ND	ND	29	16	4180	4070	8.06	8.06
8/24/2016	W	1450	1320	83	81	ND	ND	15	15	4150	3830	8.01	8.02
8/26/2016	F	1610	1470	84	72	ND	ND	15	15	4140	3820	8.05	8.04
8/29/2016	M	1460	1270	73	76	ND	ND	16	17	4390	4130	7.94	8.06
8/31/2016	W	1350	1360	74	84	ND	ND	14	16	3780	3890	7.96	8.01
9/2/2016	F	1290	1270	75	84	ND	ND	16	17	4020	4160	7.92	8.06
9/7/2016	W	1360	1290	83	82	ND	ND	15	17	4120	4030	7.94	8.01
9/9/2016	F	1379	1330	78	86	ND	ND	16	17	4380	4310	7.94	8.07
9/12/2016	M	1400	1360	80	80	ND	ND	15	17	4210	4250	7.94	7.99
9/14/2016	W	1480	1260	79	79	ND	ND	15	15	4190	3950	8.00	8.07
9/16/2016	F	1380	1350	75	76	ND	ND	15	15	4000	4050	7.94	8.07

*ND: Analyte not detected within reporting limit; NA: Not analyzed, sample vials were broken during transportation.

Table 4. Heterotrophic Plate Count Summary. Data of bracelet-installed tower is shown in blue and control tower in red.

Date		Heterotrophic Plate Count (CFU/mL)	
8/1/2016	M	>3000	>3000
8/2/2016	T	>3000	>3000
8/3/2016	W	2200	>30000
8/4/2016	Th	16000	29000
8/5/2016	F	2300	18000
8/8/2016	M	3000	>30000
8/9/2016	T	7800	>30000
8/10/2016	W	2600	>30000
8/11/2016	Th	4200	>30000
8/12/2016	F	12000	15000
8/15/2016	M	2800	30000
8/16/2016	T	16000	>30000
8/17/2016	W	2000	15000
8/18/2016	Th	2000	15000
8/19/2016	F	7100	15000
8/23/2016	T	5300	>30000
8/24/2016	W	1400	11000
8/25/2016	Th	2200	13000
8/26/2016	F	1900	17000
8/29/2016	M	2100	26000
8/30/2016	T	2700	26000
8/31/2016	W	1500	16000
9/1/2016	Th	14000	>30000
9/2/2016	F	7400	20000
9/6/2016	T	2900	20000
9/7/2016	W	900	15000
9/8/2016	Th	1500	22000
9/9/2016	F	1600	>30000
9/12/2016	M	1300	4400
9/13/2016	T	12000	30000
9/14/2016	W	12000	4900
9/15/2016	Th	4400	5700
9/16/2016	F	1700	7700

*The highest countable limit is 3,000 CFU/mL from August 1st and 2nd, is 30,000 CFU/mL for August 3rd to September 12th, and is 300,000 CFU/mL thereafter.

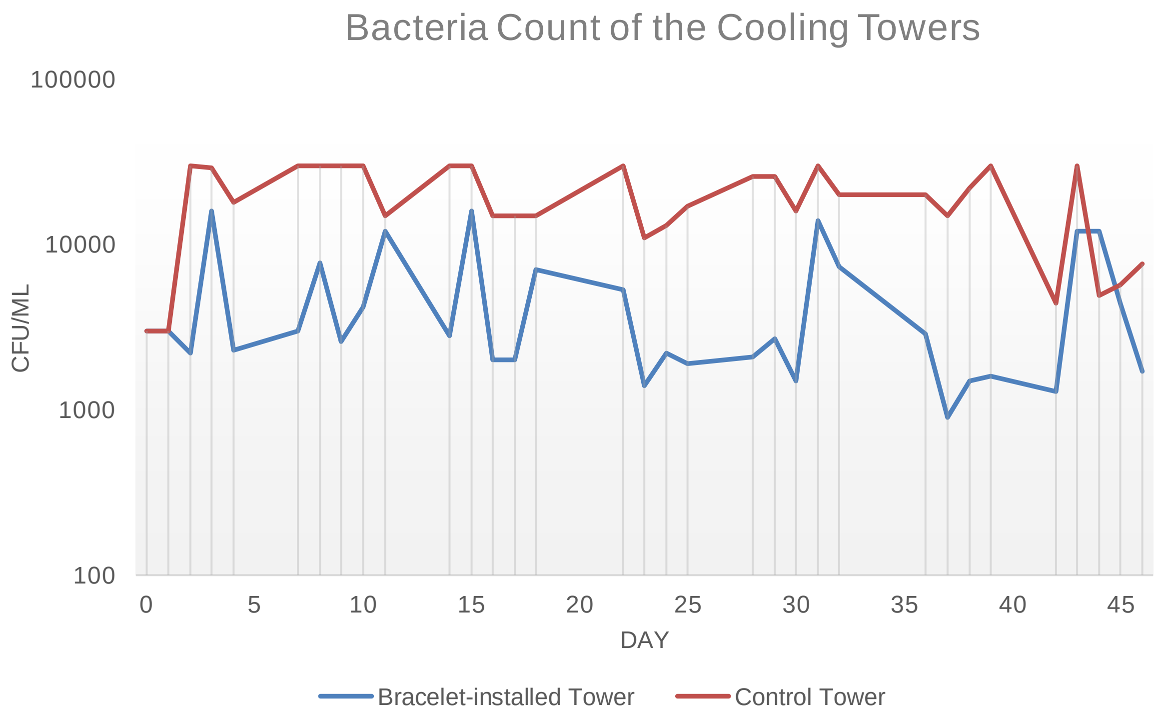

Figure 3. Bacteria count in the Cooling Towers.

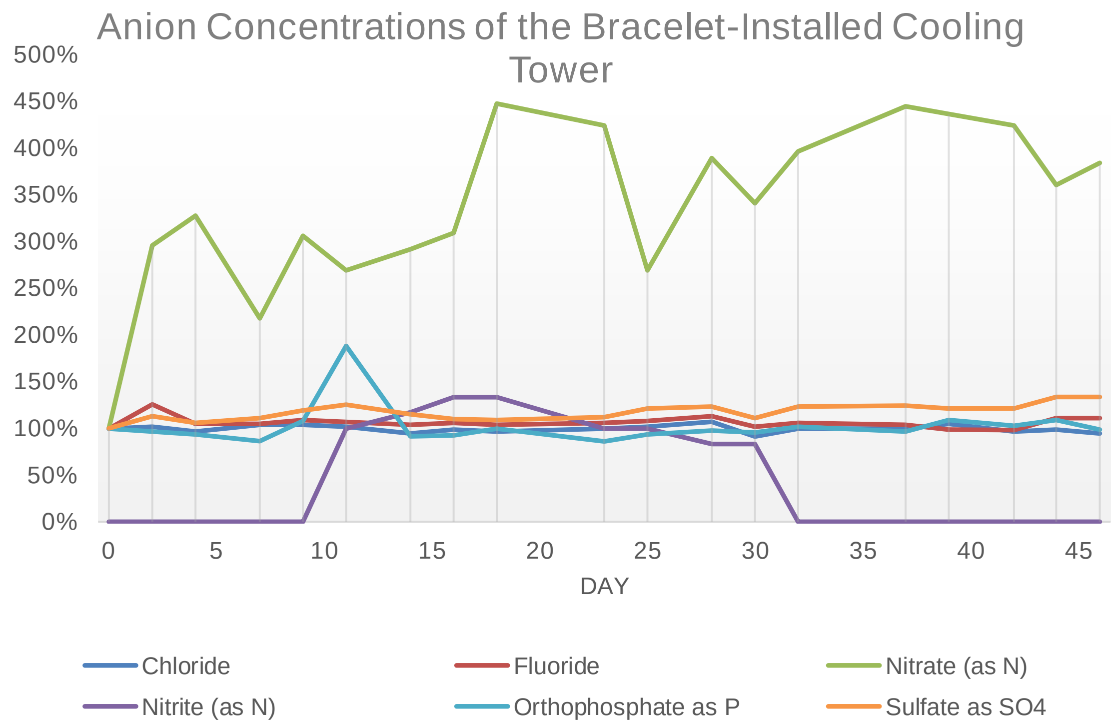

Figure 4. Anion Concentrations of the Bracelet-Installed Cooling Tower. Zero percent represents data that is below the reporting limit.

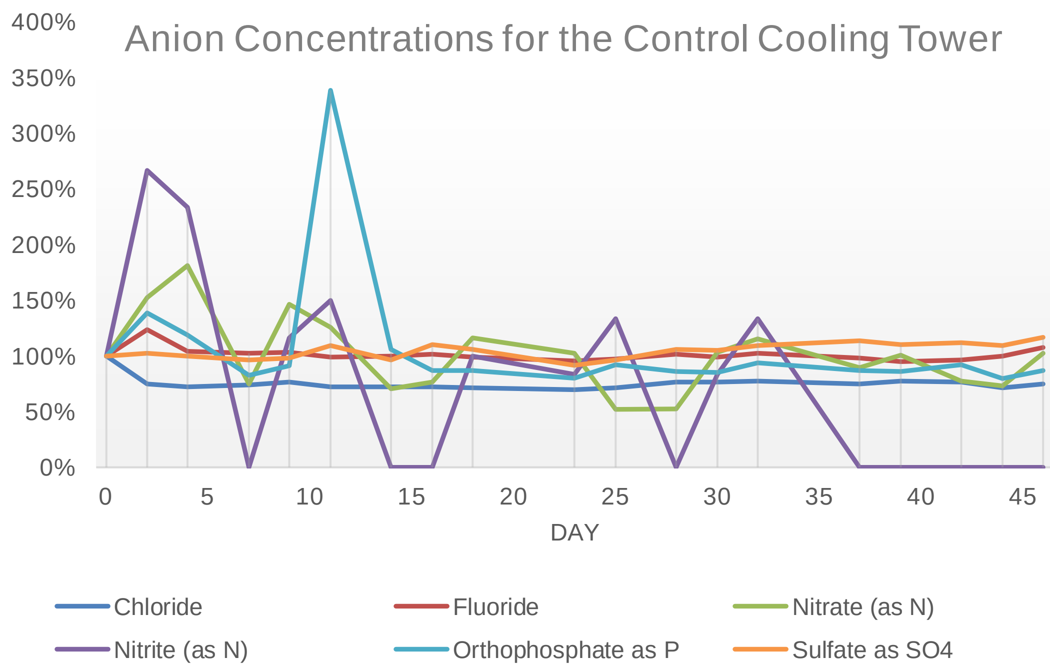

Figure 5. Anion Concentrations of the Control Cooling Tower. Zero percent represents data that is below the reporting limit.

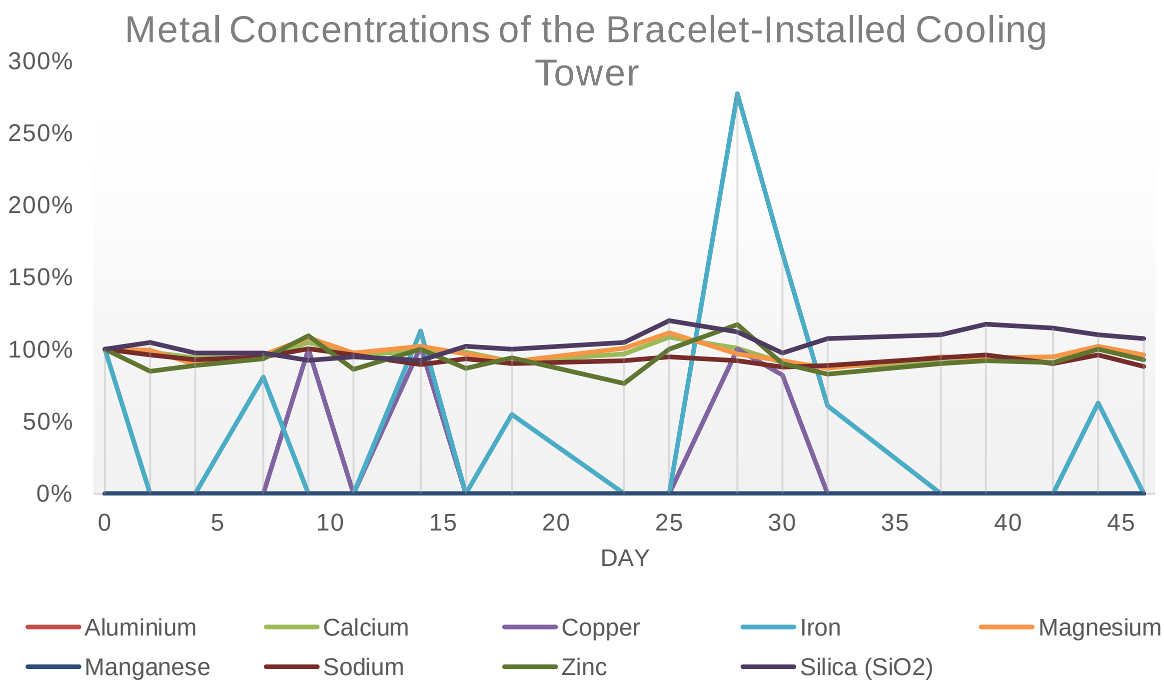

Figure 6. Metal Concentrations of the Bracelet-Installed Cooling Tower. Zero percent represents data that is below the reporting limit.

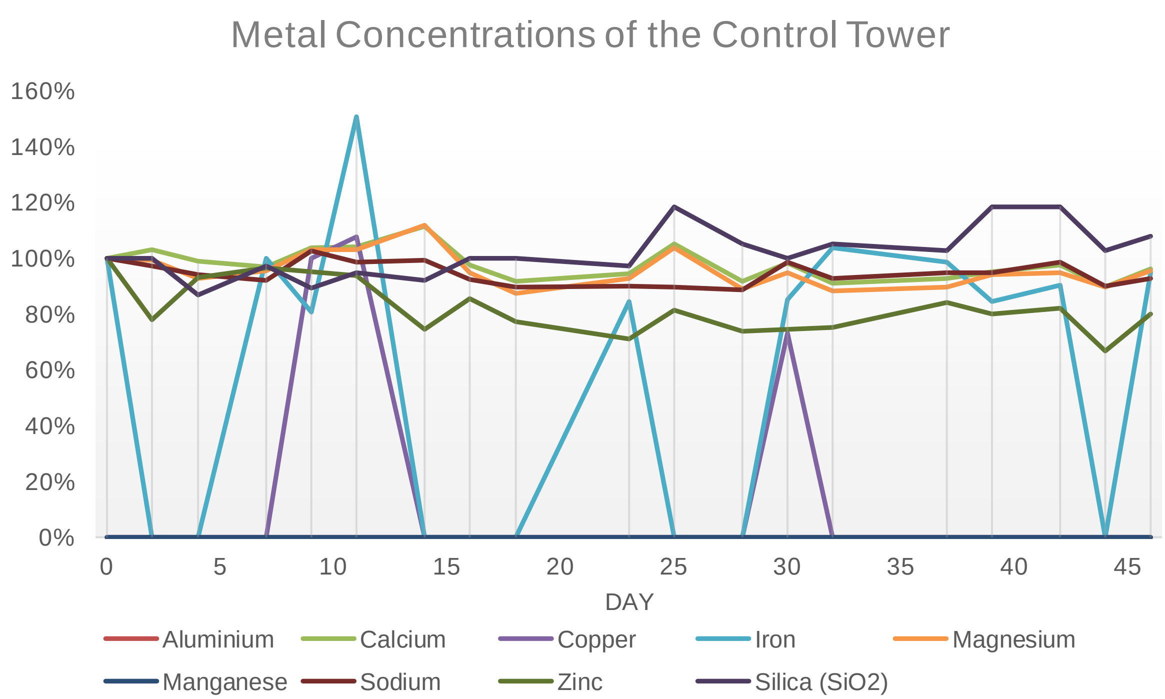

Figure 7. Metal Concentrations of the Control Cooling Tower. Zero percent represents data that is below the reporting limit.

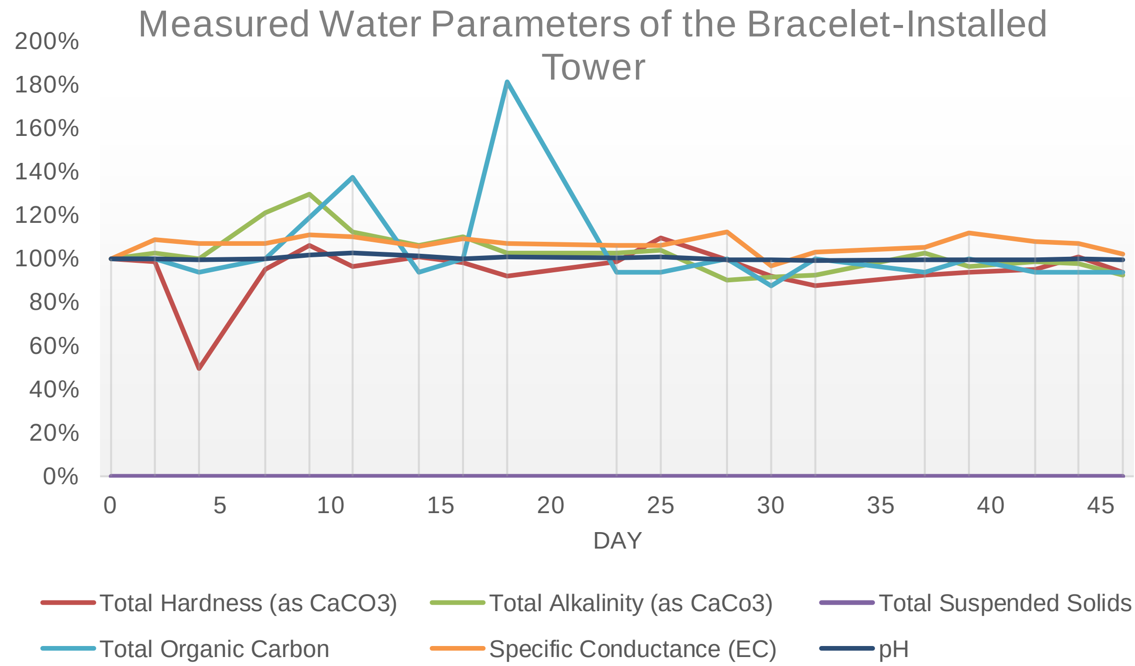

Figure 8. Measured Water Parameters of the Bracelet-Installed Cooling Tower. Zero percent represents data that is below the reporting limit.

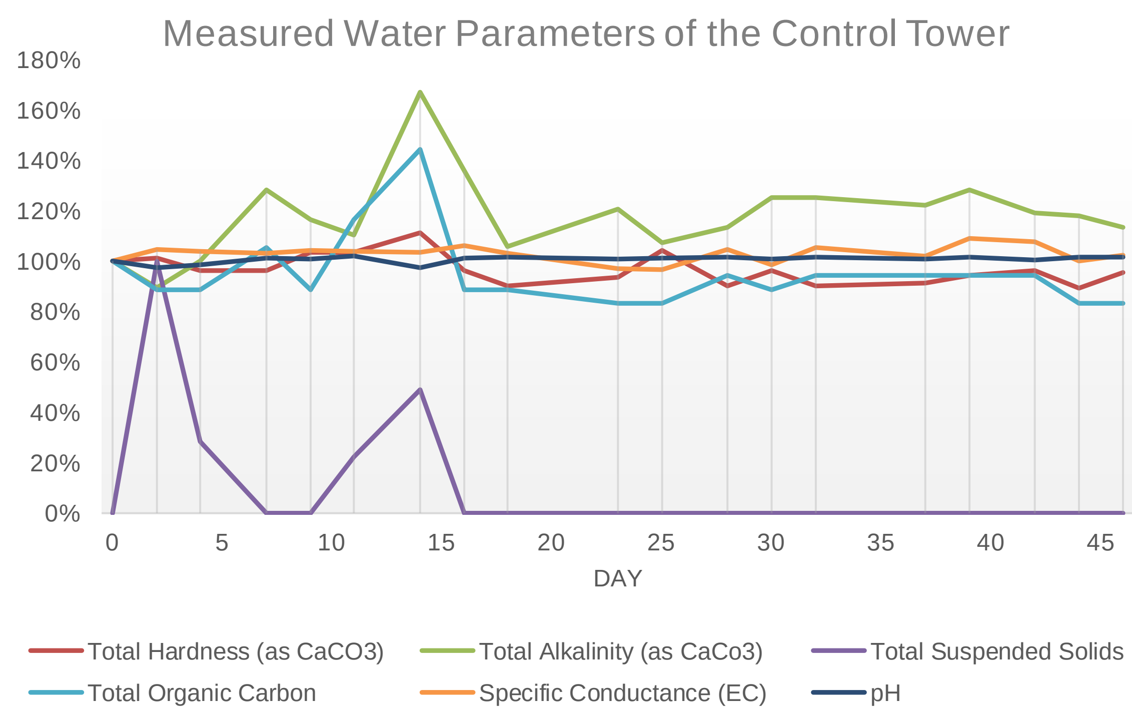

Figure 9. Measured Water Parameters of the Control Cooling Tower. Zero percent represents data that is below the reporting limit.