Biological Control in Cooling Water Systems Using Non-Chemical Treatment Devices

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Executive Summary

Microbial growth in cooling water systems causes corrosion, decreases energy efficiency, and has the potential to cause human infection. Control of microbial growth in these systems is typically achieved with the use of chemical biocides. Recently, non-chemical water treatment methods have seen increased use as an alternative. However, few objective studies with an untreated system as a reference are available to verify the efficacy of these devices to control microbial growth in cooling towers. Therefore, the specific objective of this investigation was to provide a controlled, independent, and scientific evaluation of several classes of non-chemical treatment devices (NCDs) for controlling biological activity in a model cooling tower system.

Five NCDs were evaluated for efficacy in reducing planktonic (bulk water) and sessile (biofilm) microbial populations within a pilot-scale cooling system. The devices included magnetic, pulsed electric field, electrostatic, ultrasonic, and hydrodynamic cavitation. Two model towers were designed and operated to simulate field conditions (e.g., heat load, residence time, liquid loading rate, evaporative cooling, blowdown and make-up system). One tower served as the untreated control (T1) while the NCD was installed on the second tower (T2). Each device trial was conducted over a 4-week period. Heterotrophic plate counts (HPC) were used to monitor biological growth in both planktonic and attached phase. Physicochemical monitoring included temperature, conductivity, pH, alkalinity, hardness, total dissolved solids (TDS), ORP, and chloride. Make-up water for each system was dechlorinated city tap water. According to information documented in the literature, the makeup water chemistry used in this study is representative of that found in many building cooling tower systems.

Under the experimental conditions used in this study, no statistically significant difference (p values in a t-test above 0.05) in planktonic and sessile microbial concentrations

(HPC) was observed between the control tower and a tower treated by any of the five NCDs evaluated in this study (i.e., biological and chemical parameters were comparable in T1 and T2 for all device trials).

Standard chemical treatment of pilot-scale cooling towers by the addition of free chlorine (positive control) was able to achieve appreciable reduction in both planktonic (2-3 orders of magnitude) and sessile microbial growth (3-4 orders of magnitude) in these towers. These positive controls were repeated three times throughout the study and the results clearly showed that free chlorine was able to control biological growth in every instance, even after heavy microbial colonization of model cooling towers.

The results of this study conducted under well-controlled conditions show that NCDs did not control biological growth under the conditions of the testing in the pilot scale cooling tower systems. As with any research project, the conclusions that can be drawn to full-scale applications are limited by the extent to which the variables possible in full-scale are understood and appropriately modeled against controls. The study can only properly conclude that the devices did not successfully control biological growth under the conditions tested. As with any protocol for biological control, it is prudent for building owners and engineers to consider taking water sample tests for all HVAC systems that require biological control. If the testing shows an issue, appropriate adjustment of technology or protocols followed by additional testing is important to prevent potential health or operating issues.

TABLE OF CONTENTS

1.0 INTRODUCTION 1

1.1 COOLING SYSTEM WATER TREATMENT 1

1.2 NON-CHEMICAL WATER TREATMENT 3

1.2.1 Magnetic Treatment 3

1.2.2 Pulsed-power and Electrostatic Treatment 5

1.2.3 Ultrasonic Cavitation Treatment 9

1.2.4 Hydrodynamic Cavitation Treatment 12

2.0 EXPERIMENTAL PROTOCOL 16

2.1 SPECIFIC OBJECTIVES 16

2.2 MATERIALS AND METHODS 17

2.2.1 Cooling Tower System Description 17

2.2.2 Device Trial Protocol 21

2.2.3 Chemical Disinfection Test Protocols 28

2.2.3.1 Pre-Device Trial Chlorination Test Protocol 29

2.2.3.2 ED/UD Chlorination Test 30

2.2.3.3 UD/HCD Chlorination Test 31

2.3 NON-CHEMICAL DEVICES 33

2.3.1 Magnetic Device (MD) 33
2.3.2 Pulsed Electric Field Device (PEFD) 34
2.3.3 Electrostatic Device (ED) 38
2.3.4 Ultrasonic Device (UD) 39
2.3.5 Hydrodynamic Cavitation Device (HCD) 40
3.0 EXPERIMENTAL REPRODUCIBILITY 43
3.1 MAKE-UP WATER QUALITY 44
3.1.1 Biological Parameters 45
3.1.2 Summary 46
3.2 T1 (CONTROL) CONDITIONS 48
3.2.1 T1 (Control) System Operation 48
3.2.2 Biological Parameters 50
3.2.3 Summary 53
4.0 DEVICE TRIAL RESULTS AND DISCUSSION 55
4.1 MAGNETIC DEVICE (MD) TRIAL 55
4.1.1 Tower System Operation 56
4.1.2 Biological Parameters 58
4.1.3 Summary 64
4.2 PULSED ELECTRIC FIELD DEVICE (PEFD) TRIAL 1/2 66
4.2.1 Tower System Operation 66
4.2.2 Biological Parameters 68
4.2.3 Summary 72
4.3 PULSED ELECTRIC FIELD DEVICE (PEFD) TRIAL 2/2 73

4.3.1 Tower System Operation 74
4.3.2 Biological Parameters 76
4.3.3 Summary 81
4.4 ELECTROSTATIC DEVICE (ED) TRIAL 83
4.4.1 Tower System Operation 83
4.4.2 Biological Parameters 85
4.4.3 Summary 91
4.5 ULTRASONIC DEVICE (UD) TRIAL 92
4.5.1 Tower System Operation 92
4.5.2 Biological Parameters 95
4.5.3 Summary 99
4.6 HYDRODYNAMIC CAVITATION DEVICE (HCD) TRIAL 100
4.6.1 Tower System Operation 100
4.6.2 Biological Parameters 102
4.6.3 Summary 106
4.7 CHLORINATION TEST RESULTS 107
4.7.1 Pre-Device Trial Chlorination Test 107
4.7.2 ED/UD Chlorination Test 110
4.7.3 UD/HCD Chlorination Test 111
4.8 HPC ANALYSES USING ATP MICROBIAL EQUIVALENTS AND POUR PLATE METHOD 115
5.0 SUMMARY AND CONCLUSIONS 117
6.0 REFERENCES 121

APPENDIX A: SCALING INDICES CALCULATION & INTERPRETATION 128
APPENDIX B : EXPERIMENTAL REPRODUCIBILITY 130
APPENDIX B.1 – MAKE-UP WATER QUALITY 130
APPENDIX B.2 – T1 (CONTROL) WATER QUALITY 134
APPENDIX C: MD TRIAL 141
APPENDIX C.1 – EXPERIMENTAL RESULTS 141
APPENDIX C.2 – STATISTICAL ANALYSES 152
APPENDIX C.3 - PHOTOGRAPHS OF TOWER SYSTEMS 157
APPENDIX D : PEFD TRIAL 1/2 161
APPENDIX D.1 – EXPERIMENTAL RESULTS 161
APPENDIX D.2 – STATISTICAL ANALYSES OF CHEMICAL PARAMETERS 172
APPENDIX D.3 – PHOTOGRAPHS OF TOWER SYSTEMS 177
APPENDIX E : PEFD TRIAL 2/2 184
APPENDIX E.1 – EXPERIMENTAL RESULTS 184
APPENDIX E.2 – STATISTICAL ANALYSES 195
APPENDIX E.3 – PHOTOGRAPHS OF TOWER SYSTEMS 200
APPENDIX E.4 – BIOFILM SAMPLING COUPON COMPARISON 206
APPENDIX F : ED TRIAL 207
APPENDIX F.1 – EXPERIMENTAL RESULTS 207
APPENDIX F.2 – STATISTICAL ANALYSES 218
APPENDIX F.3 – PHOTOGRAPHS OF TOWER SYSTEMS 223
APPENDIX F.4 – BIOFILM SAMPLING COUPON COMPARISON 230
APPENDIX G : UD TRIAL 231

APPENDIX G.1 – EXPERIMENTAL RESULTS 231
APPENDIX G.2 – STATISTICAL ANALYSES 242
APPENDIX G.3 – PHOTOGRAPHS OF TOWER SYSTEMS 247
APPENDIX G.4 – BIOFILM SAMPLING COUPON COMPARISON 255
APPENDIX H : HCD TRIAL 256
APPENDIX H.1 – EXPERIMENTAL RESULTS 256
APPENDIX H.2 – STATISTICAL ANALYSES 267
APPENDIX H.3 – PHOTOGRAPHS OF TOWER SYSTEMS 272
APPENDIX H.4 – BIOFILM SAMPLING COUPON COMPARISON 280

1.0 INTRODUCTION

This section will provide a brief background for the investigation discussed in this report. First, the basic principles of cooling system water treatment will be discussed. Next, an introduction to several non-chemical treatment technologies will be provided. Each of the technologies discussed in this section was tested during this investigation. These included magnetic, pulsed-power, electrostatic, ultrasonic, and hydrodynamic cavitation treatment mechanisms.

1.1 COOLING SYSTEM WATER TREATMENT

Disinfection is essential for the maintenance of an efficient cooling water system. Pathogens must be removed from industrial process waters to prevent spreading of pathogenic organisms and associated health problems. In addition, the presence of microorganisms in cooling water systems can lead to bacterially-induced corrosion of the infrastructure and inefficient heat transfer due to coating of surfaces with heavy microbial growth (biofilm). It is currently a standard practice to use chemical biocides as means of limiting microbial growth. The most popular of these chemical agents is free chlorine, which is utilized as a primary disinfectant in the United States as well as throughout the world.

The exclusive use of chemical disinfectants for water treatment has fallen out of favor in recent years for several reasons. Chemical disinfectants must be replenished regularly. Many biocidal agents are highly toxic and may cause serious health problems to those handling them.

As a result, safety training must be provided for workers who are responsible for the application of treatment chemicals. Extended use of chemical disinfectants can lead to the establishment of microbial resistance, which forces industries to regularly alternate the primary biocide [Gaines *et al.,* 2007]. Chemical disinfectants may also combine with other compounds present in treated or receiving waters to form disinfection byproducts (DBPs), which may have adverse ecological effects. In particular, the formation of toxic trihalomethanes in waters containing chlorine residual has been well-documented [Lee *et al.,* 2007].

Chemical water treatment has been the industry standard for control of biological growth. However, due to the limitations described above there has been significant interest in the use of non-chemical (physical) water treatment technologies. Physical non-chemical water treatment devices have been marketed since the late 19th century [Faunce & Cabell, 1890], but only recently have they seen widespread application. Magnetic treatment exhibited potential applicability for corrosion and scale control [Baker and Judd, 1996]. However, it has not been shown to effectively control microbial growth. Other physical water treatment technologies, including pulsed-power systems, electrostatic systems, ultrasonic systems, and hydrodynamic cavitation systems, have been developed and marketed by a variety of manufacturers over the past several decades. While the manufacturers of some devices claim that their products are capable of controlling scaling, corrosion, and microbial growth, only a limited amount of independently performed research has substantiated those claims [Baker & Judd, 1996; Kitzman *et al*., 2003; Phull *et al*., 1997; Vega-Mercado *et al*., 1997].

The validity of claims regarding device efficiency must be evaluated using objective criteria. Studies that report anecdotal uncontrolled observations, experiments performed under laboratory conditions that do not simulate field conditions, or studies conducted or supported by

the device manufacturer have less scientific merit than controlled prospective studies conducted under conditions that do simulate typical cooling tower operation.

The investigation described in this report has been funded by the American Society of Heating, Refrigeration and Air-Conditioning Engineers (ASHRAE) with the intention of providing an independent and unbiased study to determine if these non-chemical treatment devices can control biological growth in cooling tower systems.

1.2 NON-CHEMICAL WATER TREATMENT

1.2.1 Magnetic Treatment

Magnetic water conditioners have been applied to reduce scaling and corrosion in industrial systems for several decades. There are no claims, however, that magnetic devices control microbial growth in cooling tower systems. These devices function by allowing the water to pass through a fixed magnetic field. This field is purported to alter the water chemistry to prevent the formation of "hard" scales on cooling surfaces. These "hard" scales effect heat transfer and are difficult to remove. Factors which affect the ability of a magnetic water conditioner to prevent scale formation include "chemical properties of the water, strength and configuration of the magnetic field, thermodynamic properties of the water and fluid flow characteristics" [Quinn *et al.,* 1997].

All molecules can be classified as either polar or non-polar. "A non-polar molecule is one in which the center of gravity of the positive nuclei and the electrons coincide, while a polar

molecule is one in which they do not" [Quinn *et al.,* 1997]. As a result, molecules which maintain symmetry, such as the diatomic gases, are non-polar, while molecules which do not, such as water, are polar. When non-polar molecules are exposed to a magnetic field, displacement of molecular charges leads to the formation of an induced dipole. This induced dipole allows non-polar molecules to align themselves in the direction of the applied magnetic field. In addition, polar molecules also align themselves according to the magnetic field direction. According to Quinn et al. (1997), the capacity of a magnetic conditioner can be determined by the gauss strength, flux density, surface area of the exposure , the number of fields and the distance between alternating poles. In order for a fixed magnetic field to be effectively used for water conditioning, certain conditions must be met. These conditions are as follows [Quinn *et al.,* 1997]:

- The water path must be perpendicular to the magnetic lines of force.
- Water should first cut the south magnetic lines and then proceed to break wider and more dense alternating reversing polarity lines, until exiting the magnetic chamber through the single north pole flux path.
- Water must be under pressure and moving with the least amount of turbulence possible, just before entering and during its travel through the magnetic fields.

Under these conditions, several studies have indicated that magnetic conditioners are able to prevent the formation of scaling compounds. Calcium carbonate that is exposed to a magnetic field may be converted from the calcite form, which is responsible for scale formation, into the aragonite form, which exists as a "soft" precipitate that may easily be removed from cooling

surfaces. [Quinn *et al.,* 1997]. A study conducted by the National Aeronautics and Space Administration (NASA) in 1975 reported negligible corrosion rates for magnetically conditioned water, while chemically treated water was reported to have a corrosion rate between 1-50 mils/yr [Kuivinen, 1975]. Researchers have repeatedly verified that magnetic fields increase the potential of coagulation in waters containing at least moderate levels of hardness [Bogatin, 1999]. Later studies revealed that the efficiency of a magnetic treatment system was independent of the hardness of the water being treated [Gabrielli *et al.*, 2001].

1.2.2 Pulsed-power and Electrostatic Treatment

Pulsed-power treatment, also referred to as pulsed electric field (PEF) treatment [Rieder *et al.,* 2008] or electropulse treatment [Danilenko *et al.,* 2005], involves the bombardment of the substance to be treated with pulses of electromagnetic energy. These pulses may inactivate microorganisms, including pathogens, present in the treated liquid. However, the optimal mechanism by which this process occurs has not been definitively established. The biocidal application of pulsed-power technology has been evaluated for use in the food processing industry [Feng *et al.,* 2004], as well as for cooling tower process water treatment [Opheim, 2001; Kitzman *et al.,* 2003].

Laboratory studies have demonstrated the efficacy of pulsed-power disinfection technology, particularly in food and beverage production. The work of Feng *et al*. (2004) demonstrated a high reduction in aerobic plate counts prepared from germinated brown rice in circulating water treated with pulsed-power. The reactions which produced this antimicrobial

effect, however, were found to be electrochemical, rather than physical. Additionally, this effect was observed in a closed system, as opposed to a cooling water system which allows for the intrusion of airborne bacteria and bacteria from incoming make-up water. Oil field reinjection water treatment using pulsed-power technology has also been investigated in laboratory settings, resulting in several log reduction of saprophytic, sulfate-reducing, and iron bacteria [Xin *et al.*, 2008]. However, these experiments were performed under batch conditions, as opposed to a field setting. Treatment times required for effective microbial reductions were long, with significant effects not appearing until nearly 15 minutes of continuous treatment.

Evidence of pulsed-power's ability to inactivate pathogens has also been claimed in several field studies. Application of pulsed-power systems on ice skating rink cooling towers in Connecticut demonstrated several log reduction of microorganisms (quantified using heterotrophic plate counts) both in bulk water and in biofilm coupon samples prepared from glass slides [Opheim, 2001]. These antimicrobial effects were observed after two months of treatment, and heterotrophic plate counts during the first two months of operation following installation ranged from 10^3 to 10^6 CFU/mL.

A similar investigation involved the application of a pulsed-power treatment system to evaporative coolers at Alcoa's Mt. Holly Works in Goose Creek, SC [Kitzman, 2003]. This study compared the efficacy of pulsed-power treatment at varying cooling tower cycles of concentration to chemical and hydrodynamic cavitation treatment. Pulsed-power treatment demonstrated average plate counts of aerobic and anaerobic planktonic bacteria of 65,000 CFU/mL and 85 CFU/mL, respectively and it appears that microbial control using this treatment system was more consistent than that observed using chemical treatment. It is important to note that the installation of the pulsed-power and hydrodynamic cavitation systems at Mt. Holly

Works included a cyclonic separator for solids removal. During the investigation, each of the three cooling towers received large quantities of airborne carbon dust from a nearby carbon silo. While the pulsed-power and hydrodynamic cavitation experimental towers were able to remove this additional carbon via sidestream filtration, the chemically treated tower had no means of filtration, and as a result turbidity levels in chemically treated tower (>120 NTU) were significantly higher than in the towers receiving physical water treatment (10-15 NTU). It is uncertain whether the observed antimicrobial effects of the physical water treatment systems were the result of the physical treatment mechanisms or the removal of large amounts of carbon via sidestream filtration. Additionally, although plate counts were lower in the tower with the pulsed-power device than in the chemically treated tower, none of the treatment methods produced plate counts below the industry standard of 10^4 CFU/mL.

Multiple mechanisms have been proposed for the inactivation of microorganisms by pulses of electromagnetic energy. It has been suggested that pulsed-power treatment can disrupt protective cellular structures in a mechanism known as electroporation. Laboratory studies documented that pulsed-power systems are capable of generating a transmembrane potential that is sufficient to cause electroporation [Zimmermann, 1986; Tsong, 1991]. The electromagnetic pulses lead to "[t]he bi-electrical breakthrough of the phosphorus lipid double layer in biological membranes," rupturing the cell and causing death [Rieder *et al.,* 2008]. This mechanism is also supported by research which demonstrated the relative resistance of Gram-negative bacteria to pulsed-power treatment in comparison to Gram-positive bacteria [Min *et al.,* 2007]. This study attributed the resistance of Gram-negative bacteria to the presence of cell walls, and it also documented pulsed-power treatment resistance in bacterial spores and mold ascospores.

Laboratory investigations have verified that exposure to electromagnetic energy disrupts and weakens cellular membranes. The electromagnetic fields in these studies [Zimmermann, 1986; Tsong, 1991] were very strong (volts to kilovolts), and they were applied over very short distances and for relatively long exposure times (milliseconds – microseconds) in comparison to the conditions generated by commercially available pulsed-power treatment systems. The strengths of the applied fields in these studies were far higher than those produced by commercial treatment systems, and the distance of exposure was comparatively short (cm). While these investigations have demonstrated that the induction of electroporation is possible through the application of pulses of electromagnetic energy, it has not been demonstrated that the electromagnetic exposures generated by commercially available treatment systems are powerful enough to produce these effects.

Another hypothesis regarding the mechanism for inactivation of microorganisms by pulsed-power treatment proposes that free radicals (OH^-, ClO^-) are formed in water treated with electromagnetic bombardment [Vega-Mercado *et al.*, 1997; Oshima *et al.*, 1997]. Further investigation of this theory confirmed the formation of hydroxyl radicals upon exposure to electromagnetic pulses [Feng *et al.*, 2000]. As a result, hybridization of pulsed-power systems with low-dose chemical disinfectant feeds have been proposed, as the electromagnetic pulses have demonstrated the ability to increase free chlorine concentrations [Abderahmane *et al.*, 2008]. This indicates that the antimicrobial effects of pulsed-power treatment systems may be the result of electrochemical reactions, and that their mechanism of operation may not be strictly physical. While both the electroporation mechanism and the free radical formation mechanism have been substantiated with supporting laboratory research, it has not yet been determined which may be effective for microbial inactivation in field conditions.

The mechanisms of operation for electrostatic treatment systems are essentially identical to that involved in the operation of pulsed-power treatment systems. The primary difference is that electrostatic systems apply a static electric field, rather than pulses of energy. The claims of the manufacturers of these devices also include scaling, corrosion, and microbial control [Huchler, 2002]. However, there is no published literature indicating that the application of relatively weak static electric fields for a very short exposure time over a relatively large distance is capable of producing any antimicrobial effects.

1.2.3 Ultrasonic Cavitation Treatment

The use of ultrasonic energy to inactivate microorganisms has been under investigation for several years. The first studies, attempting to utilize ultrasound as means of microbial inactivation, date back to 1929, when the technology was originally considered to be too energy intensive for commercial applications [Harvey & Loomis, 1929]. However, significant advances have been made in the field of ultrasonic technology over the past several decades, and it is now being re-evaluated as a potential disinfection agent. The use of ultrasound alone for disinfection remains energy-intensive, especially for high volume applications, and a number of studies have been conducted to evaluate the disinfection capacity of ultrasound in combination with other disinfection technologies [Joyce, 2003].

The interaction of ultrasonic energy with water results in cavitation process through a process known as sonication. Several processes resulting from the collapse of these cavitation bubbles are responsible for bacterial inactivation [Mason & Joyce *et al.*, 2003]:

- Forces due to surface resonance of the bacterial cell are induced by cavitation. Pressures and pressure gradients resulting from the collapse of gas bubbles which enter the bacterial solution on or near the bacterial cell wall can cause cellular damage. Bacterial cell damage results from mechanical fatigue, over a period of time, which depends on frequency.
- Shear forces induced by microstreaming occur within bacterial cells.
- Chemical attack due to the formation of radicals ($H^{\bullet}$ and $OH^{\bullet}$) during cavitation in the aqueous medium can also cause cellular damage. These radicals attack the chemical structure of the bacterial cell wall and weaken the cell wall to the point of disintegration.
- Among the final products of the sonochemical degradation of water is hydrogen peroxide (H_2O_2), which is a strong bactericide.

Bench-scale investigations of the application of both low- (20-40 kHz) and high- (0.5-0.85 MHz) frequency ultrasonic energy to bacterial mediums have indicated that ultrasound may be responsible for both the death of bacteria and the disruption of cellular agglomerations [Mason & Joyce *et al.*, 2003]. These two processes can also counteract, resulting in no net biocidal effect. However, tests involving the use of an ultrasonic treatment unit to decontaminate 20 L of bacterial suspension contained within a flow loop have demonstrated a kill efficiency of near 85% [Mason & Duckhouse *et al.*, 2003].

Several studies have investigated the application of ultrasound for the disinfection of both drinking water and wastewater. Some of these investigations have evaluated ultrasound as a stand-alone treatment process [Furuta *et al.*, 2004; Hua & Thompson, 2000; Scherba *et al.*, 1991]

or in combination with other treatment technologies. Hybrid disinfection systems combining ultraviolet treatment with ultrasonic pretreatment generated coliform reductions of 3.3 to 3.7 log units, whereas ultraviolet treatment alone generated reductions of 2.5 log units [Blume & Neis, 2004]. Treatment with an ultrasonic horn combined with 5 mg/L H_2O_2 dosing was able to generate a 15-minute disinfection percentage of 90% for fecal coliform removal, while 5 mg/L H_2O_2 dosing alone was only able to produce a disinfection percentage of 9% and treatment with the ultrasonic horn alone was only able to produce a disinfection percentage of 47% [Jyoti & Pandit, 2003]. A system combining an ultrasonic horn with 0.5 mg/L ozone dosing was able to produce a 99.9% disinfection efficiency for heterotrophic plate count bacteria after 15 minutes of treatment, while 15 minute stand-alone treatment regimens of 0.5 mg/L ozone dosing and ultrasonic horn treatment produced heterotrophic plate count bacteria removal percentages of 46% and 50%, respectively [Jyoti & Pandit, 2004].

While research has demonstrated that microbial inactivation is possible through the application of ultrasonic energy, it has not successfully demonstrated that this technology may be applied to cooling system process water. The treatment times utilized in the studies performed by Jyoti & Pandit were long (15 minutes) compared to the very short treatment times required for cooling system process water. Since ultrasonic bombardment produces no residual, it may not be effective in controlling the microbial population of a cooling water system which receives large quantities of airborne material at various points throughout the system during operation. Some form of residual treatment is likely necessary in order to prevent microbial fouling from occurring, and as a result ultrasonic treatment systems alone may not be capable of significant microbial control in cooling water systems.

1.2.4 Hydrodynamic Cavitation Treatment

When fluids are subjected to sudden high pressure changes, very small vapor bubbles may form within the fluid in a process known as cavitation. These bubbles quickly collapse, leading to extremely high local temperatures, pressures, and fluid velocities. The implosion of these small bubbles of fluid vapor within a liquid may lead to inactivation of surrounding organisms. This process of fluid vapor bubble formation as a result of "fluctuations in fluid pressure" is known as hydrodynamic cavitation [Gaines, 2007], and it has been proposed as a possible method of disinfection for potable water, wastewater, and industrial process water.

The efficacy of hydrodynamic cavitation disinfection has been demonstrated by a number of laboratory researchers. A group of investigators from India recently demonstrated the biocidal effect of hydrodynamic cavitation on zooplankton, achieving disinfection efficiency of approximately 80% [Sawant, 2008]. Additionally, elimination of *Legionella pneumophila* bacteria using a laboratory-scale hydrodynamic cavitation treatment system has been observed [Stout, 2002]. Hydrodynamic cavitation systems were able to generate a fecal coliform disinfection efficiency of 89% after 60 minutes of treatment [Jyoti & Pandit, 2003]. This treatment time is far longer than may be utilized by a cooling water treatment system, however.

Field studies involving the application of hydrodynamic cavitation treatment systems for cooling tower make up water have also demonstrated positive results with regards to the control of microbial populations. A group of researchers funded by the American Society of Heating, Refrigerating, and Air-Conditioning Engineers (ASHRAE) investigated the biocidal capabilities of a hydrodynamic cavitation system installed on a cooling tower located in an automotive test lab [Gaines, 2007]. This system was capable of sustaining heterotrophic plate count values

below approximately 10^4 CFU/mL for over 2 months. However, several HPCs in excess of 10^4 CFU/mL were observed during the investigation. Field application of a hydrodynamic cavitation treatment system was also performed alongside pulsed-power treatment testing at Mt. Holly Works in Goose Creek, SC [Kitzman, 2003]. In this study, the hydrodynamic cavitation device was able to successfully maintain an average aerobic plate count of 95,500 CFU/mL and an average anaerobic plate count of 87 CFU/mL in its cooling water system. However, the installation of this system was accompanied by the installation of cyclonic separator for solids removal. Since the systems during this investigation received large amounts of airborne carbon fines from a nearby carbon silo, and the chemically-treated tower in this comparative study had no form of filtration, a conclusion may not be made whether or not the observed reduction in microbial activity in the system treated by hydrodynamic cavitation was the result of the physical treatment process or the removal of high levels of excess carbon through filtration.

Several mechanisms for the inactivation of microorganisms via hydrodynamic cavitation have been proposed. Studies have demonstrated that cavitation may result in the formation of free radicals when applied to aqueous solutions [Kalumuck *et al.*, 2003]. These hydrogen, hydroxyl, and hydrogen peroxide radicals have the potential to eliminate pathogens, but only in the area directly surrounding the bubble which collapses [Gaines, 2007]. The collapse of these small fluid vapor bubbles also releases high pressure pulses (up to 1450 psia), shear forces, and shock waves which are capable of causing nearby cells to rupture [Brennen, 1995]. Brennen's work has demonstrated that extremely high temperatures (as high as 8540 °F) occur for a few microseconds at the interface between the liquid and the vapor bubble during collapse. These temperatures may also lead to the death of nearby microorganisms [Gaines, 2007]. Additionally, when vapor bubble collapse occurs asymmetrically (i.e. near a surface), jets of fluid with

extremely high velocities are formed, as well as eddies with large amounts of energy [Naude & Elllis, 1961; Benjamin & Ellis, 1966]. These high fluid velocities may also be responsible for cellular inactivation.

The development of hydrodynamic cavitation disinfection systems, much like that of pulsed-power systems, must direct future efforts towards determining which inactivation mechanisms to exploit. While microorganism inactivation has been documented via several pathways, researchers must determine which pathway allows for the most efficient disinfection. Several hydrodynamic cavitation systems are commercially available for use as a primary form of disinfection, relying on asymmetric bubble collapse, pressure shock waves, and extreme local temperatures for microbial inactivation. However, systems which employ the use of hydrodynamic cavitation in combination with a low dose of chemical disinfectant and ultrasonic cavitation technologies are also under development.

Researchers from the University of Mumbai in India have investigated a number of hybrid hydrodynamic cavitation disinfection systems, coupling this treatment technology with ultrasonic cavitation, ozone dosing, and hydrogen peroxide dosing [Jyoti & Pandit, 2003; Jyoti & Pandit, 2004]. The combination of hydrodynamic cavitation with these disinfection technologies demonstrated a synergistic effect. Hydrodynamic cavitation coupled with 5 mg/L H_2O_2 dosing generated 90% removal of fecal coliforms after 60 minutes, compared to just 21% removal produced after 60 minutes of 5 mg/L H_2O_2 dosing alone [Jyoti & Pandit, 2003]. When hydrodynamic cavitation treatment alone was employed during this study, a fecal coliform disinfection efficiency of 89% was observed after 60 minutes, indicating that the addition of hyrodgen peroxide did little to increase the disinfection capacity of the cavitation device. Hydrodynamic cavitation coupled with 0.5 mg/L dosing of ozone generated a fecal coliform

disinfection efficiency of 80% after 15 minutes of treatment, compared to disinfection efficiencies of 68% and 57% observed after 15 minutes of 0.5 mg/L ozone dosing and hydrodynamic cavitation treatment, respectively [Jyoti & Pandit, 2004]. However, the treatment times utilized during these studies are longer than is practical for cooling water treatment.

The application of hydrodynamic cavitation disinfection systems requires continued research for optimization of efficiency. The use of hybrid systems which couple hydrodynamic cavitation with other processes (both chemical and physical) should be further investigated, as the results of some preliminary hybrid process studies [Jyoti & Pandit, 2003; Jyoti & Pandit, 2004] have demonstrated potentially high disinfection efficiencies. Additionally, different hydrodynamic cavitation system designs must be compared to determine which means of inducing cavitation produces the strongest biocidal effect. The ability of hydrodynamic cavitation systems to eliminate *Legionella* has been demonstrated in the laboratory, but further field research is necessary to verify these findings. Additionally, hydrodynamic cavitation systems may affect the heat transfer efficiency of the water being treated, a subject which warrants further investigation in order to be substantiated [Gaines, 2007]. Like ultrasonic cavitation treatment, hydrodynamic cavitation does not produce any form of residual treatment.

2.0 EXPERIMENTAL PROTOCOL

The following section will provide a detailed outline of the project performance. The objectives of the investigation will be described, and the materials and methods used for the completion of the study will be presented. This section will include a description of all parameters monitored during each experiment, frequency and method of measurement, and specific instrumentation used for sample analysis. In addition, a description of each non-chemical treatment device evaluated during this investigation will be provided.

2.1 SPECIFIC OBJECTIVES

The primary objective of this investigation was to evaluate the ability of five (5) non-chemical water treatment devices to significantly reduce the microbial population within a pilot-scale cooling water system. This objective was accomplished in a well-controlled study in which two identical cooling water systems were operated simultaneously. One tower received treatment from the non-chemical device being tested, while the other tower received no treatment for the duration of the experimental trial. Each device was activated at the beginning of the trial and allowed to operate for the duration of the experiment (4 weeks). In addition, the performance of non-chemical devices for the control of biological growth in pilot-scale cooling

towers was compared to standard chemical treatment protocol (i.e., chlorination) that was performed several times during this study as positive control (Section 4.7.) for experimental validation.

2.2 MATERIALS AND METHODS

2.2.1 Cooling Tower System Description

Two pilot-scale model cooling tower systems were used to evaluate the performance of each device. The two model cooling towers used in this study were designed to be identical. A schematic outlining the cooling system setup for each tower is shown in **Figure 1**.

In each pilot-scale system, water is stored in a 60 gal. holding tank prior to being pumped at a rate of 7 gpm by a 2 hp centrifugal pump into a stainless steel heating bath. The system flow rate is controlled by the use of a side stream placed immediately after the pump discharge. This sidestream returns a portion of the flow back to the 60 gal. holding tank. The rate of return flow is controlled by a needle valve, allowing the tower operator to manually adjust the system flow rate to approximately 7 gpm.

Immediately prior to entering the heating bath, the flow of water is split into two paths, and each flow path continues into a coil of 1/2" OD copper tubing. The two coils (approximately 105 ft. and 44 ft.) wrap around a 15 kW immersion heater, and the entire heating apparatus is surrounded by a stainless steel box containing dechlorinated water. The box is sealed by a lid made of ½ in. thick plexiglass in order to minimize evaporative losses. The immersion heater is

controlled by a thermostat, which was adjusted throughout the experimental trials to maintain a water bath temperature of approximately 120°F. This heating bath temperature provides enough thermal energy to elevate the temperature of the system water to 95-100°F.

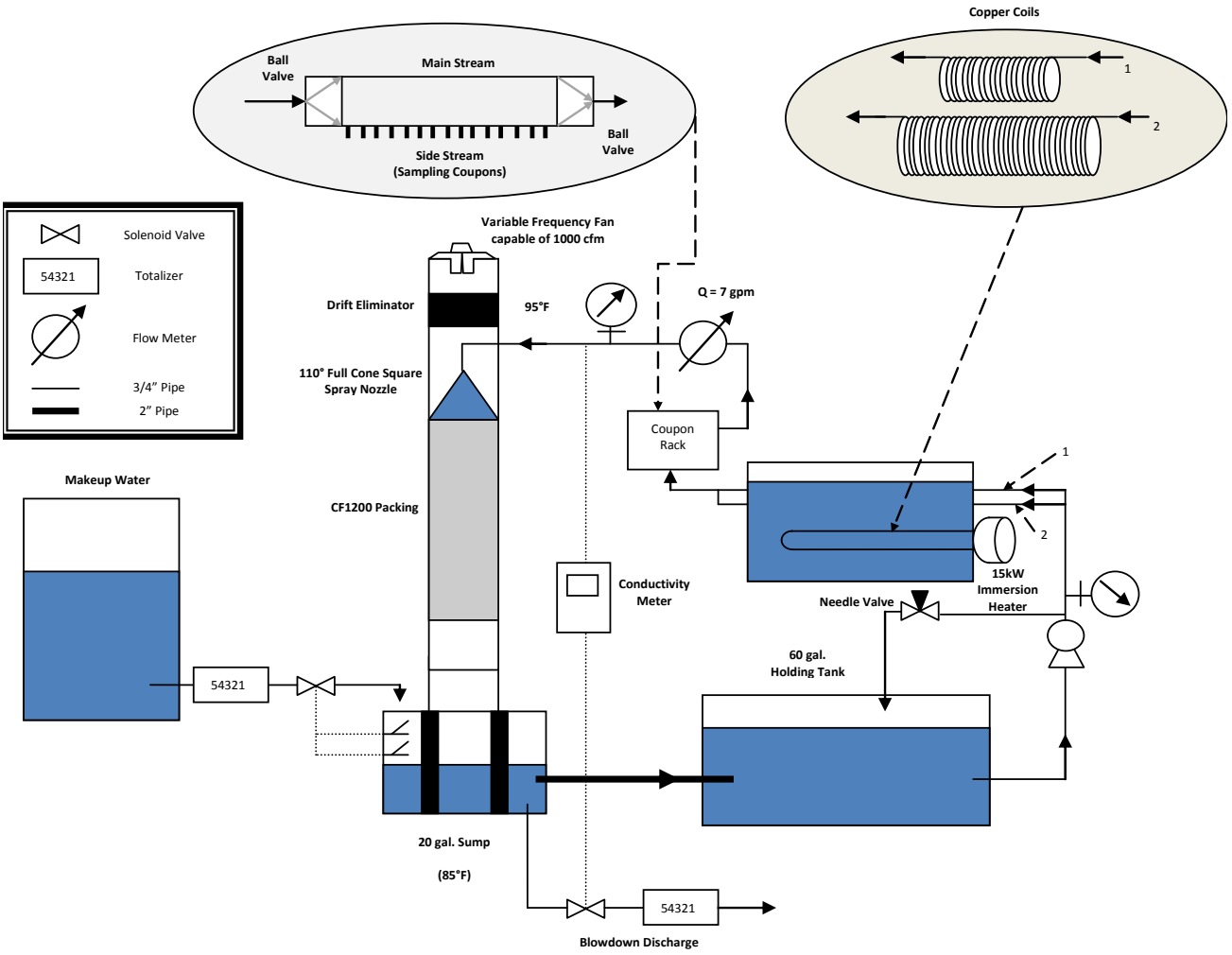


Figure 1 - Pilot-Scale Cooling System Schematic

Once the system water passes through the two copper coils, the flow paths are combined. The flow is then diverted through a sampling rack containing a series of sampling coupons. The

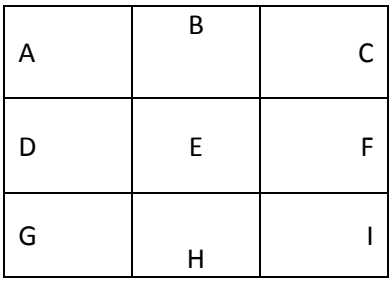
sampling coupons were 5.61 cm^2 stainless steel washers which were scrubbed and autoclaved at 121°F prior to installation in the experimental towers. These coupons were installed at the beginning of each device trial, and they were used to quantify biofilm growth within each of the cooling tower systems. Coupons were installed parallel to the direction of flow.

Upon exiting the sampling rack, the system flow passes through a number of sensors for data collection. Flow passes past a pH probe, an ORP probe, a conductivity probe, and a thermometer designed to record the water temperature prior to tower entrance. Each of these probes is connected to an AquaTrac Multiflex data collection system, which records data values at 1-hour intervals. The flow then passes through a flow meter to ensure that system flow rate of 7 gpm is maintained. Immediately prior to tower entrance, the flow passes over an additional conductivity meter. This conductivity meter is connected to a blowdown control system which uses conductivity readings to control when the tower goes through blowdown based on a user-defined blowdown conductivity setpoint. The setpoint is chosen based on the make-up water conductivity, and it was selected to produce 4-5 cycles of concentration in the cooling tower system.

Flow enters each of the cooling towers by way of a 110° full cone square spray nozzle. This allows the flow to be distributed evenly over the surface of the CF1200 packing (Brentwood Industries) which is installed in each tower. The height of the packing in each tower is adjusted so that the spray from the nozzle contacts the packing at its uppermost edge, diverting flow through the interior of the packing rather than down the side wall of the tower. A total of three units of packing (1 ft^3 each) were installed vertically in each tower system, for a total packing height of 3 ft.

Once the water has travelled through the packing, it is deposited into a 20 gal. sump. To ensure an even flow distribution across the packing, water flow was measured at several points across the packing's cross sectional area (**Table 1**). In order to minimize water losses from splashing, screening was placed around the perimeter of each towers' support legs.

Table 1 - Water flow (gpm) distribution across packing



Packing Measurement Locations (Cross-Sectional View)

	A	B	c	D	E	F	G	H	I
T1	0.465	0.343	0.375	0.655	0.396	0.417	0.517	0.480	0.306
T2	0.422	0.322	0.338	0.449	0.475	0.549	0.581	0.607	0.602

Upon entering the sump, the water temperature decreased to 85-90°F, thereby maintaining a temperature differential across the packing of approximately 10°F. This cooling is accomplished by a variable frequency axial fan placed at the top of the tower, above the water entrance. The rate of airflow generated by this fan is controlled by a potentiometer to produce the desired 10°F temperature differential. A distribution of the airflow across the width of the tower is shown in **Table 2**. The 20 gal. sump is connected to the 60 gal. holding tank via a 2 in. diameter PVC pipe, and as water travels through the system it is pulled from the 20 gal. sump back into the 60 gal. holding tank, completing the cooling water cycle.

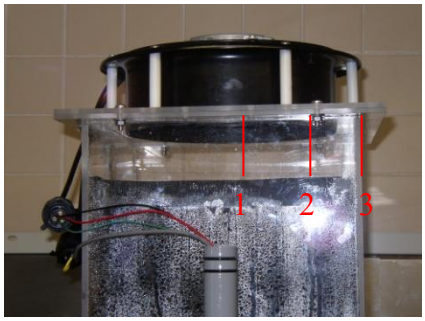
Make-up water used for all experiments in this study was dechlorinated City of Pittsburgh tap water. Dechlorination was accomplished by passing the water through a fixed-bed activated carbon adsorber [Loret *et al.,* 2005]. The cylindrical activated carbon adsorber had a height of 6 ft. and a diameter of 12 in. The column contained 33 L of activated carbon (TIGG 5DC 0830, coconut shell based, 8 x 30 mesh size, activity = 1000; manufactured by TIGG Corporation), and the flow rate through the column during make-up water generation was maintained at or below 3 gpm in order to ensure a minimum contact time of 3 min. Make-up water for each cooling tower was stored in four 125-gal polyethylene tanks to provide enough water for two days of tower operation (approximate tank residence time = 48 hrs). In between device trials, the carbon column was flushed by running water through it at twice the flow rate necessary for chlorine removal (> 6 gpm) for a minimum of 1 hr.

2.2.2 Device Trial Protocol

For each device trial, a control tower and a test tower were utilized. The control tower (T1) received no treatment during the testing process, while the device tower (T2) received treatment from the device being evaluated. The device was activated at the beginning of the study, and it was not turned off until the investigation had been completed. For the remainder of this report, the control tower in each device trial will be referred to as T1 (Control), and the device tower will be referred to as T2 (Device). Lights in both the shower room containing the two test towers and the locker room containing the make-up water storage tanks were kept on

throughout the duration of each device trial. No algal growth was observed in either of the towers or the make-up water storage tanks during any of the device trials or chlorination tests.

Table 2 - Airflow distribution across tower. Highlighted value indicates operating airflow rates



Fan Potentiometer Setting	Airflow Velocity (ft/s)		
	1	2	3
10	900	1020	810
9	678	920	720
8	691	800	503
7	660	760	330
6	545	680	215

A total of five (5) non-chemical water treatment devices were tested over the course of this investigation. A device testing schedule is shown in Table 3. Before the beginning of each device trial, several measures were taken to ensure consistent starting conditions. Each tower received 4 gal. of dilute acetic acid and 250 mL of 5.25% sodium hypochlorite solution, and the towers were allowed to operate for several hours in order to eliminate any residual microorganisms present in the system and to remove scale formed during the previous trial. Both towers and their corresponding sumps and holding tanks were scrubbed with 5% acetic acid to remove as much scale as possible. Each system was drained completely using a shop vacuum,

and refilled with clean make-up water. The draining and refilling process was repeated a minimum of 2 times for each tower prior to the beginning of a new device trial. Make-up water storage tanks were also drained and refilled prior to the beginning of a new device trial. Device trials began less than 24 hours after the final refilling of both the tower systems and the make-up water storage tanks. Additionally, the plastic packing in each of the towers was replaced prior to the initialization of a new test. The new packing was installed after the tower had been drained and rinsed to reduce the amount of residual solid material which it collected.

Table 3 – Non-chemical device testing schedule

Device Name Abbreviation	Treatment Technology	Test Date Range(s)	Total Days of Testing	Phase I Completed	Phase II Completed
MD	Magnetic	2/11/09 - 3/2/09, 3/13/09 - 4/20/09	56	YES	NO
PEFD	Pulsed Electric Field	5/2/09 - 5/30/09, 6/12/09 - 7/10/09	58	YES	NO
ED	Static Electric Field	7/18/09 - 8/15/09	29	YES	NO
UD	Ultrasound	9/2/09 - 9/30/09	29	YES	NO
HCD	Hydrodynamic Cavitation	10/27/09 - 11/24/09	29	YES	NO

In order to maximize the cooling potential of the packing, each tower underwent a "seasoning" process prior to trial onset. This process was in accordance with the packing manufacturer's specifications. To season the packing, each tower was allowed to operate with a heat load for approximately 1 hour. Following this period of operation, each tower system was shut off, allowing heated water deposited on the packing surface to evaporate, leaving a thin layer of deposited minerals on the packing surface. This process was repeated a minimum of two times prior to the beginning of each device trial, and the entire process occurred over

approximately 3 days. Each tower system was drained and replenished following the final seasoning of the packing.

Throughout the course of the study, water temperature measurements were taken at regular intervals to determine both the temperature of the water entering the tower system and the water in the sump. Measurements of the water temperature entering each of the tower systems were taken every hour using an AquaTrack MultiFlex data logging device. A manual reading of this temperature for each tower system was recorded on a daily basis. Sump temperature measurements were taken every 15 minutes using a portable thermologger (Omega Scientific). Additionally, manual sump temperature measurements were taken daily using a handheld thermometer.

A number of physical, chemical, and biological parameters were monitored during the performance of each device trial. The parameters measured, as well as their corresponding frequencies of measurement, are shown in Table 4. Immediately following this table is a description of the protocol used for biofilm sampling. A picture of the biofilm sampling coupon setup is shown in **Figure 2**.

Table 4 – Physical, chemical, and biological parameters analyzed during investigation

Parameter	Source	Frequency of Measurement	Standard Method	Device
Temperature Entering Tower	Tower	Continuous	-	AquaTrac Multiflex
Sump Temperature	Tower	Continuous	-	Digital Thermometer
pH	Make-up and Tower	Daily	-	Fisher Accumet pH meter Model 25
ORP	Tower	Daily	-	Fisher Accumet pH meter Model 25 with ORP selective electrode
Conductivity	Make-up and Tower	Continuous	Method 2510	Sybron-Barnstead Conductivity Bridge (Model PM-70CB)
Alkalinity	Make-up and Tower	Daily	Method 2320 B	-
TDS	Tower	Daily	Method 2540	-
TDS	Make-up	Monthly	Method 2540	-
Chloride	Make-up and Tower	Weekly	Ion Chromatography	Dionex 4500 Ion Chromatograph with conductivity detector
Chlorine	Make-up	Weekly	DPD Method	Hach DR 2010 Spectrophotometer
Calcium Hardness	Tower	Daily	Method 3111 B, Method 2340 B	Perkin Elmer 1100B atomic absorption spectrophotometer
Magnesium Hardness	Tower	Daily	*	*
Total Hardness	Tower	Daily	*	*
Calcium Hardness	Make-up	Monthly	*	*
Magnesium Hardness	Make-up	Monthly	*	*
Total Hardness	Make-up	Monthly	*	*
Planktonic Heterotrophic Plate Count	Make-up and Tower	Bi-weekly	Method 9215	-
Sessile Heterotrophic Plate Count	Tower	Weekly	Method 9215 (See below for procedure)	-
ATP	Make-up and Tower	Bi-weekly	[QGA Quick-Reference Guide, 2007]	LuminUltra Technologies Ltd. Quench-Gone Aqueous (QGA) ATP test kit

Biofilm Sampling Protocol

Close valves to bypass coupon rack

1. Put on gloves
2. Unscrew coupon holder from rack and remove
3. Swab bottom of coupon before unscrewing
4. Hold coupon over sterile conical tube and unscrew nut
5. Release coupon into 5 mL conical tube – bottom side down
6. Swab top of coupon and add 10 mL of sterile water to tube
7. Place swab into tube and agitate vigorously to remove attached material from swab. Cut or break swab and leave in the tube
8. Transport sample to lab as soon as possible
9. Vortex for 30 sec before testing the sample liquid. Process for HPC using appropriate dilution scheme
10. Total CFU recovered is calculated by multiplying CFU x 10 mL sample volume
11. Divide total CFU by surface area of the coupon (5.61 cm^2). Report coupon results as CFU/cm^2
12. Save coupon and sterilize for reuse

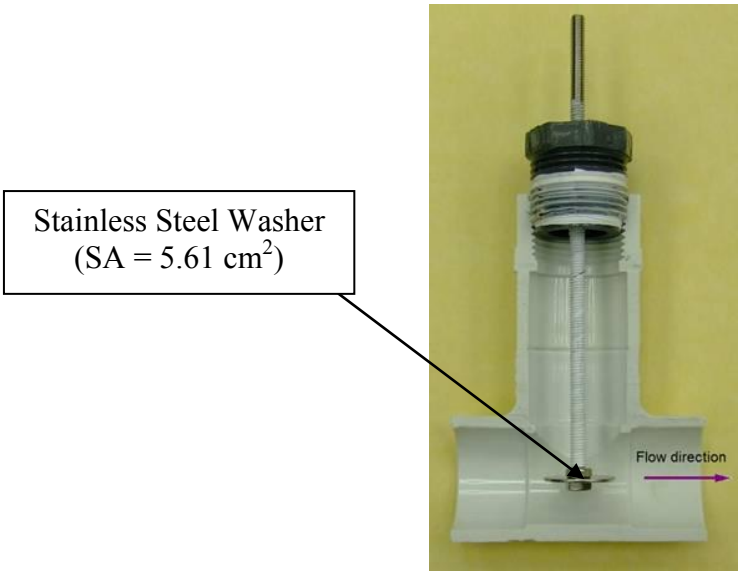


Figure 2 – Diagram of biofilm sampling coupon setup

Bulk water samples were collected using sterilized sampling bottles. Biofilm samples were collected using the protocol described below. All biological samples were kept chilled during transport to the laboratory. Upon arrival, samples were shaken thoroughly and subject to a series of dilutions. The dilutions used were determined using pre-device trial testing, which indicated the levels of microbial growth to be expected in each of the towers, as well as in the make-up water.

A series of three dilutions was plated for each bulk water and biofilm sample. Ten-fold dilutions were accomplished by transferring 1.0 mL of sample water to a test tube containing 9.0 mL of sterilized deionized water. Hundred-fold dilutions were accomplished by transferring 0.1 mL of sample water into a test tube containing 9.9 mL of sterilized deionized water. The range of dilutions used for make-up water analysis was 10^{-2} – 10^{-4} for this investigation, while the bulk water tower dilution range was 10^{-3} – 10^{-5} and the biofilm sample dilution range was 10^{-4} – 10^{-6}. Fresh sterilized pipette tips were used for each volumetric transfer during the dilution process. Dilutions were plated according to Standard Method 9215 pour plate protocol.

The measurement of cellular ATP was performed using a test kit manufactured by LuminUltra™ Technologies Ltd. This test kit was used to measure relative light units (RLUs) passing through a sample. By comparing the measured number of RLUs to a standard of a known concentration of ATP, it was possible to determine the concentration of ATP present in each tower system biological sample. The following equation was used to calculate cellular ATP (cATP) [QGA Quick-Reference Guide, 2007]:

$$cATP\ \left(\frac{pg}{mL}\right) = \frac{RLU_{Measured}}{RLU_{UltraCheck}} \times \frac{10000}{Sample\ Volume\ (mL)}$$

In this equation, $RLU_{UltraCheck}$ refers to the measured RLUs passing through a standard of known ATP concentration. For all measurements, a sample volume of 50 mL was used. Cellular ATP was used to determine the number of microbial equivalents present in each sample for comparison with heterotrophic plate counts. This was done using the following equation [QGA Quick-Reference Guide, 2007]:

$$Microbial\ Equivalents\ \left(\frac{\#}{mL}\right) = \ cATP\ \left(\frac{pg}{mL}\right) \times 1000$$

2.2.3 Chemical Disinfection Test Protocols

During the course of investigating the effectiveness of NCDs for the control of biological growth in cooling towers, three chlorination tests were performed. These tests, which involved the addition of chlorine to the device tower (T2), are essential to provide scientifically defensible evidence that industry-tested disinfection methods are capable of controlling microbial growth in the experimental system operated in this study. Demonstrating the effectiveness of these disinfection tests indicates that the comparison between accepted and experimental treatment mechanisms is valid.

The selection of free chlorine as a positive control was based on common practice in cooling water treatment and a previous study where several chemical biocides, including free chlorine, were evaluated in model cooling towers that simulated real-world cooling tower operational conditions [Thomas *et al*., 1999]. The Thomas *et al*. investigation was performed

using a series of cross-flow cooling tower cells. Prior to the performance of disinfection trials, the researchers demonstrated that the tower cells were capable of generating and maintaining a heterotrophic bacterial population $>10^6$ CFU/mL after 48 hrs of operation. In the first phase of the Thomas et. al. study, the chlorine treatment protocol (0.5-1.5 ppm as free residual oxidant) reduced planktonic heterotrophic bacteria by at least 3 orders of magnitude (99.9%) and reduced heterotrophic bacteria in biofilms by 3-4 orders of magnitude (99.9+%) compared to controls. This study demonstrated that chlorination may be used as an effective means of biological control when applied to a model cooling tower system, and its application to a pilot-scale system can produce results which directly reflect those observed in full-scale cooling systems.

2.2.3.1 Pre-Device Trial Chlorination Test Protocol

The first chlorination test was performed prior to the beginning of the device trials. During this test, both T1 (Control) and T2 (Device) operated untreated from 1/15/09 – 1/22/09. After samples were taken on 1/22, a spike dose of chlorine (80 mL of 5.25% sodium hypochlorite supplied by Fisher Scientific) was added to each of the towers, resulting in an initial chlorine dose of approximately 14 mg/L. Following this spike dose, chlorine stock solution was pumped into each tower system to maintain a chlorine concentration of approximately 1 mg/L for 3 days. For this chlorination test, the stock solution was prepared by adding 60 mL of 5.25% sodium hypochlorite solution per gallon of dechlorinated water, resulting in a free chlorine concentration of 832 mg/L.

For the first day of chlorination, chlorine stock solution was pumped into each tower at a rate of 5 mL/min. Flow rates for the 2nd and 3rd days of chlorination were approximately 3 mL/min and 2 mL/min, respectively, as a result of decreased chlorine demand due to microbial inactivation. Overall, the total volume of chlorine stock solution added to each tower during this 3-day test was 14.4 L, corresponding to a total chlorine mass added to each tower of approximately 12 g. The average makeup water consumption rate for each tower was approximately 130 gal/day and the average initial chlorine feed for each tower can be estimated at approximately 8 mg/L. Considering that the chlorine residual in each tower was maintained at approximately 1 mg/L, the disinfectant demand for this experimental system heavily colonized with microbial growth can be estimated at approximately 7 mg/L.

2.2.3.2 ED/UD Chlorination Test

The second chlorination test was performed immediately after the ED trial and immediately before the towers were prepared for the UD trial. During this chlorination test, T2 (Device) received chemical treatment, while T1 (Control) remained untreated. Chlorination began on 8/21 after biological samples were taken, and it was performed for three days. The sodium hypochlorite purchased from Fischer Scientific was consumed during the cleaning processes following the MD and PEFD device trials, and new sodium hypochlorite solution had to be purchased. The sodium hypochlorite solution used for this chlorination test was 6.0% household bleach.

The chlorination process began with a spike dose of chlorine (80 mL of 6.0% sodium hypochlorite), resulting in an initial chlorine dose of approximately 16 mg/L. This shock dose was followed by a steady flow of chlorine stock solution in order to maintain a free chlorine concentration of approximately 1 mg/L. The stock solution was prepared by adding 52 mL of 6.0% sodium hypochlorite per gallon of dechlorinated water for an approximate concentrion of 832 mg/L. The flow rate of chlorine stock solution on days 1, 2, and 3 of chlorination were 5 mL/min, 3 mL/min, and 2 mL/min, respectively. Overall, the total volume of chlorine stock solution added to each tower during this 3-day test was 14.4 L, corresponding to a total chlorine mass added to each tower of approximately 12 g. The average makeup water consumption rate for each tower was approximately 130 gal/day and the average initial chlorine feed for each tower can be estimated at approximately 8 mg/L. Considering that the chlorine residual in each tower was maintained at approximately 1 mg/L, the disinfectant demand for this experimental system heavily colonized with microbial growth can be estimated at approximately 7 mg/L.

2.2.3.3 UD/HCD Chlorination Test

The third chlorination test was performed immediately following the UD trial and immediately before the towers were prepared for the HCD trial. Chlorination of T2 (Device) began on 10/1 following biological sampling, and it was performed using a steady dose of chlorine stock solution. The sodium hypochlorite solution used for this chlorination test was 6.0% household bleach manufactured by Target Corporation. The stock solution was prepared by adding 52 mL

of 6.0% sodium hypochlorite per gallon of dechlorinated water for an approximate concentrion of 832 mg/L.

No shock dose of chlorine was used for this test, but instead a gradual increase in chlorine concentration was performed over a period of 5 days until the residual concentration reached 1 mg/L. The flow rate of chlorine stock solution on day 1 was approximately 8 mL/min. The flow rate was decreased to 6 mL/min on day 2, 5 mL/min on day 3, 3 mL/min on day 4, and 2 mL/min on day 5. Overall, the total volume of chlorine stock solution added to each tower during this 5-day test was 34.6 L, corresponding to a total chlorine mass added to each tower of approximately 29 g (in the absence of an initial shock dose of chlorine). The average makeup water for each tower was approximately 130 gal/day and the average initial chlorine feed for each tower can be estimated at approximately 11.4 mg/L. Considering that the chlorine residual in each tower was maintained at approximately 1 mg/L, the disinfectant demand for this experimental system heavily colonized with microbial growth can be estimated at approximately 10.4 mg/L. While this demand was higher than the previous 2 chlorination tests, this may be attributed to the fact that no initial shock dose of chlorine was applied during this test.

2.3 NON-CHEMICAL DEVICES

2.3.1 Magnetic Device (MD)

The magnetic device evaluated in this investigation consists of a 13" flow-through cylinder which exposes water to 4 alternating magnetic poles. A diagram of the device is shown in **Figure 3**. Since the device utilizes fixed magnetic fields to condition the water, it requires protection from electromagnetic fields generated by high-voltage electronic devices. In order to prevent any interference with the MD by the 3-phase centrifugal pump used to pump water through the cooling system, a lead shield was installed around the device during the testing period.

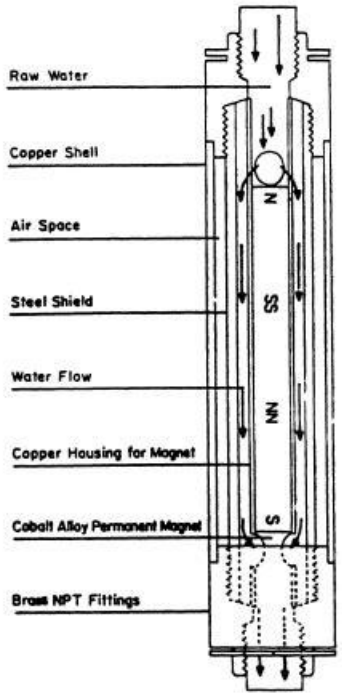


Figure 3 – Magnetic device (MD) diagram [Lin & Nadiv, 1988]

The MD is marketed as a scale-inhibiting water conditioner. The manufacturer does not claim that the device is capable of microbiological control. According to the manufacturer, the device operates by keeping mineral ions, such as calcium and magnesium in suspension, thereby preventing them from forming scale on cooling surfaces [Cho, 2002]. Instead, mineral compounds form in suspension, leading to the formation of a precipitate, which is easily removed from cooling system surfaces. This reduction in scale formation leads to increased system operational efficiency, while at the same time reducing water and energy consumption.

2.3.2 Pulsed Electric Field Device (PEFD)

The pulsed-power (electrostatic) non-chemical treatment device evaluated in this investigation is composed of two primary components: the signal generator and the treatment module. The signal generator is housed in a stainless steel box, and it contains all of the system's replaceable parts. LEDs installed in the front of the box are used to ensure proper operation, while ventilation slits on both sides of the unit allow for fan-powered cooling of the interior electronics. A power connection is also included in the signal generator, and it operates at a voltage of 115V / 60Hz. The treatment module, which consists of a 1" diameter PVC cylindrical flow-through reactor, is connected to the signal generator via an umbilical cable. A diagram of the treatment module is shown in **Figure 4**.

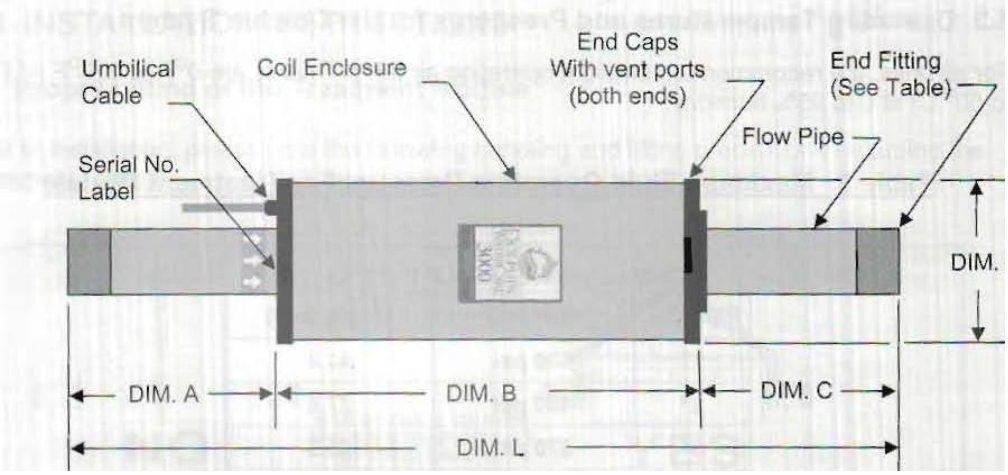


Figure 4 – Pulsed electric field device (PEFD) treatment module diagram [PEFD Technical Manual, 2008]

Operation of the pulsed electric field device involves the application of "pulsed, high-frequency electric fields into flowing water" [PEFD Technical Manual, 2008]. The characteristic waveform generated by the system's signal generator is shown in **Figure 5**. According to the manufacturer, the device is capable of controlling scale formation, equipment corrosion, microbial populations, and algal growth in a cooling water system.

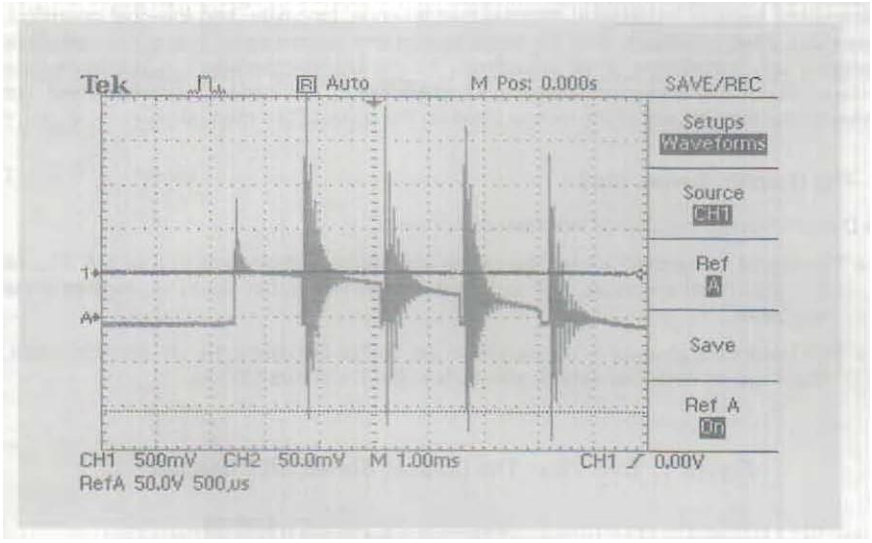


Figure 5 – PEFD characteristic waveform [PEFD Technical Manual, 2008]

The PEFD manufacturer claims that scaling is controlled via the interaction between the pulsating electromagnetic field produced by the signal generator and suspended particles in the water. According to the literature provided by the manufacturer, the PEFD "activates the suspended particles by removing the static electric charge on their surface" [PEFD Technical Manual, 2008]. By removing this electric charge, interactions between molecules found in water lead to the formation of a powdery calcium carbonate precipitate, as opposed to the formation of a thick calcium carbonate scale on system surfaces. The precipitated calcium carbonate may then be removed either through filtration or normal tower blowdown.

Biological control by PEFD is proposed to occur by two separate pathways. The first is encapsulation, where the precipitate formed as a result of the interaction between suspended particles clusters around microorganisms which are present in the cooling water system. Bacteria normally would repel these particles due to their static charge. However, by removing this static charge the PEFD allows for these suspended particles to collect around microbes, encasing them and effectively inactivating them by preventing any further microbial reproduction from occurring. The second pathway by which microbial control is proposed to occur is electroporation, which involves a physical rupturing of the membrane of planktonic bacteria. The "high frequency, pulsing action of the PEFD electric fields" leads to the formation of holes within the outer membrane of microorganisms which pass through the treatment module [PEFD Technical Manual, 2008]. As a result of this membrane damage, microorganisms spend the remainder of their 1-2 day lifespan repairing external cellular damage, eliminating their reproductive capacity and effectively rendering them inactive. The combined effect of encapsulation and electroporation is suggested to lead to a reduction in total bacterial counts in systems which utilize the PEFD.

In addition to the control of planktonic microbial population, it is proposed that the PEFD is also capable of biofilm formation prevention. This control may result from the interaction between planktonic and sessile microorganisms in cooling water systems. Planktonic bacteria generate specific nutrients which are then absorbed and utilized by sessile bacteria. The elimination of planktonic bacteria via encapsulation and electroporation greatly reduces the concentration of these nutrients present in the system's water supply. As a result, the sessile microbial population present in the system is claimed to be greatly reduced.

Corrosion control by the PEFD is a result of the prevention of scale formation and the reduction of the sessile and planktonic microbial populations within the system water. The formation of calcium carbonate precipitates is thought to act as a preventive measure against corrosion, acting as a "cathodic corrosion inhibitor," which "greatly slows the corrosion process by blocking the reception of electrons that are thrown off by the corrosion process" [PEFD Technical Manual, 2008]. Several microorganisms which commonly populate cooling water systems generate corrosive byproducts, such as hydrogen sulfide. By controlling the reproduction of these organisms, the PEFD may be able to reduce the concentrations of these corrosive byproducts, reducing the overall corrosion rate of the system water. As a result, the device manufacturer claims that both localized and uniform corrosion rates are significantly decreased by the application of their device.

The PEFD was installed in the cooling tower system used in this study according to the manufacturer's specifications. The treatment module was placed directly after the centrifugal pump and immediately before the heat bath. According to the manufacturer, the treatment module may also be placed directly after the heat exchanger but before the entrance of water into the cooling tower.

2.3.3 Electrostatic Device (ED)

The ED is an electrostatic treatment device designed to "control scaling, inhibit corrosion, [and] minimize biological fouling without chemical additives" [Environmentally Sustainable Cooling Tower Treatment, 2008]. The device was composed of a 1" flow-through reactor vessel as shown in **Figure 6.**

Figure 6 – Electrostatic device (ED) photograph

The technology by which the ED operates is similar in principle to that employed by the PEFD. While the PEFD bombards the water with pulses of electromagnetic energy, the ED exposes the water in the reactor chamber to a steady electrostatic field. Mineral ions are kept in suspension through the application of this field. This field increases molecular collisions between suspended particles, causing them to form precipitates which may easily be removed from cooling systems rather than hard scale on system surfaces. This process removes ions from

solution, which allows for the dissolution of scale which has already formed [Environmentally Sustainable Cooling Tower Treatment, 2008]. Possible mechanisms for the control of biological growth in the system would be very similar to those described for the PEFD.

2.3.4 Ultrasonic Device (UD)

The UD operates by diverting water from the cooling system sump or holding tank through a venturi and into an ultrasonic treatment cell. Once the flow velocity has been increased by passing through the venturi, air is introduced into the water stream. According to the manufacturer, the vacuum pressure generated by the venturi during normal operation should be between 0.4 and 0.75 bar below atmospheric pressure [UD Operation Manual, 2008]. The water/air mixture then enters an ultrasonic treatment chamber containing 6 ceramic transducers. Upon exiting the treatment cell, the water passes through a basket filter prior to discharge back into the cooling system sump.

The ultrasonic device was installed according to the manufacturer's specifications, and a representative from the manufacturer approved the final installation. A sidestream was constructed for the application of this device, with the sidestream intake positioned near the outlet end of the 60 gallon storage tank and the outflow positioned near the storage tank's inlet. A diagram of the ultrasonic device is shown in **Figure 7**.

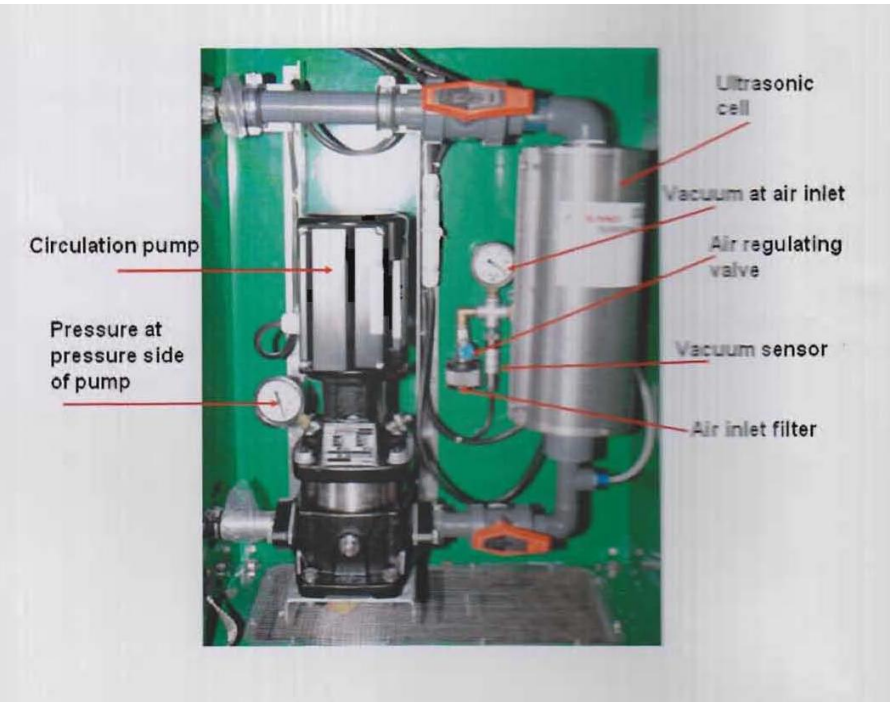


Figure 7 – Ultrasonic device (UD) diagram [UD Operation Manual, 2008]

2.3.5 Hydrodynamic Cavitation Device (HCD)

Operation of the HCD involves the diversion of water from the cooling system sump or holding tank into the device, where treatment is administered and the water is returned to the sump from which it was initially withdrawn. Water drawn from the system sump enters a pressure-equalization chamber. The flow of water is then split into two separate streams and each of these streams enters a vortex nozzle. This nozzle "…impart[s] a specific rotation and velocity to the water streams" [HCD Technology: A Primer, 2008] to create a conical flow path for each of the streams, and these streams are forced to collide in a low-pressure stabilizing chamber.

The collision of these two conical streams creates a vacuum region which results in the formation of cavitation bubbles. In turn, the collapse of these bubbles generates local regions of high shearing forces, temperatures, and pressures, leading to microbial inactivation. Additionally, "…the hydrogen-bonding molecular arrays of water are broken down, thereby allowing entrapped gasses, such as CO_2, to be released and off-gassed to [the] atmosphere." [HCD Technology: A Primer, 2008].

The manufacturer of the HCD claims that the device is capable of controlling biological growth and scaling, in addition to corrosion protection. Case studies of similar system operation provided by the manufacturer indicate that bacterial plate counts of < 10,000 CFU/mL and levels of *Legionella* below detectable limits may be achieved by proper system operation [Scappatura, 2002]. Additionally, the systems described in these case studies provided a visible reduction in system scaling, while at the same time reducing mild steel corrosion rates to < 2.5 MPY and copper corrosion rates to < 0.3 MPY [Scappatura, 2002]. As a result, the system was able to reduce blowdown volumes by over 70% [Scappatura, 2002].

The HCD was installed according to the manufacturer's specifications, and a representative from the manufacturer approved the final installation. The sidestream used for the ultrasonic device trial was reused. In order to allow for adequate CO_2 degassing, the outlet end of the sidestream was cut so that water was expelled above the sump water surface level. Plastic sheeting was installed around the sidestream outlet in order to minimize water losses from splashing. A diagram of the HCD reaction chamber provided by the manufacturer is shown in **Figure 8.**

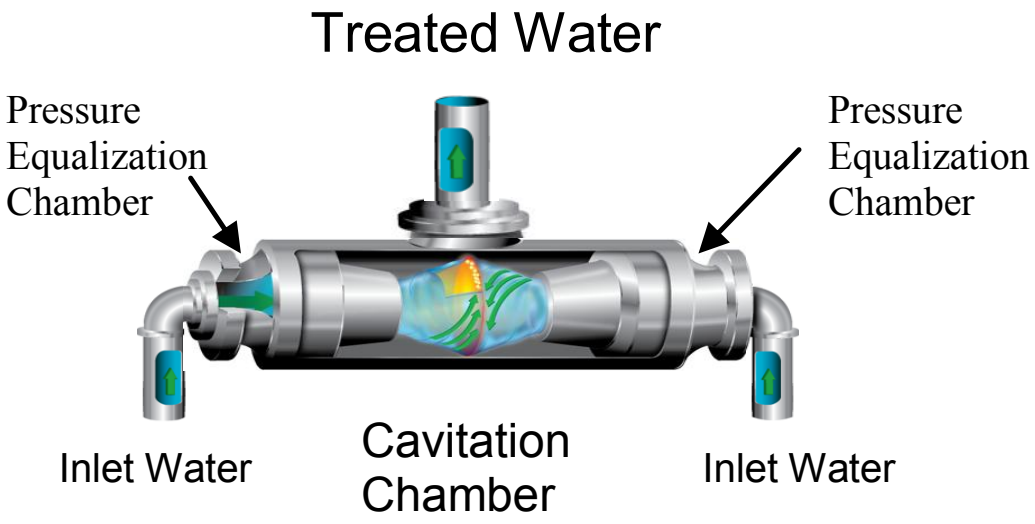


Figure 8 – Hydrodynamic cavitation device (HCD) reaction chamber diagram

3.0 EXPERIMENTAL REPRODUCIBILITY

Each of the five non-chemical water treatment devices was tested for a minimum of 4 weeks. Some tests were repeated due to operational problems. Additionally, chlorination tests were performed between some device trials in order to demonstrate that the microbial population in the towers could be controlled using a traditional approach (i.e. chemical treatment). The chemical parameter values collected during each investigation were used to calculate 3 different scaling indices. The calculation and interpretation of these indices are outlined in Appendix A.

The make-up water quality and performance of T1 (Control) throughout the course of the entire investigation were monitored in order to ensure similar conditions of operation for each individual device trial. These data were compared using statistical analyses to verify that operating conditions were not biased for any of the experimental device trials. This comparison provides evidence of experimental reproducibility by demonstrating consistent and realistic operating conditions over the course of the investigation.

The reproducibility of this investigation is governed by two main factors: make-up water quality and conditions in T1 (Control). In order to verify that each experimental device operated under similar conditions during its respective device trial, it is necessary to analyze the make-up water entering each of the two tower systems for similarity during each device trial. Additionally, it is necessary to demonstrate that control conditions were essentially constant over

the duration of the entire experiment. Data figures and tables demonstrating experimental reproducibility in terms of physical/chemical parameters of make-up water and T1 (Control) operating conditions are included in Appendix B.

3.1 MAKE-UP WATER QUALITY

Both chemical and biological characteristics of the make-up water were monitored regularly over the course of the experiment. Chemical parameters of make-up water quality are shown in Appendix B.1, while the average values for the measured physical and chemical make-up water parameters are shown in **Table 5**. Detailed monthly measurements are shown in **Figure B1 – Figure B4** of Appendix B and summarized in **Table B1**.

Table 5 – Average values for make-up water

Make-up Water Summary Table		
	Average	Standard Deviation
Conductivity (mS/cm)	0.307	0.039
pH	7.33	0.22
Alkalinity (mg/L as $CaCO_3$)	26	5
Calcium Hardness (mg/L as $CaCO_3$)	28	7.3
Magnesium Hardness (mg/L as $CaCO_3$)	24	5.9
Total Hardness (mg/L as $CaCO_3$)	52	11
TDS (mg/L)	208	25
Chloride (mg/L)	37	4
Free Chlorine (mg/L)	0.017	0.009
Copper (mg/L)	ND	-
Iron (mg/L)	ND	-
Sulfate (mg/L)	40.7	6.3
Phosphate (mg/L)	1.15	0.32
Planktonic HPC (CFU/mL)	2.66E+04	6.13E+04

3.1.1 Biological Parameters

Average planktonic HPC in make-up water for each device trial are summarized in **Figure 9.** **Figure 10** shows the variation in planktonic HPC in the make-up water for each trial**.** A comparison of these parameters demonstrates that, while fluctuations in both the biological and chemical composition of the make-up water were observed over the course of the experiment, these fluctuations did not alter conditions to an extent which would have adversely affected experimental reproducibility.

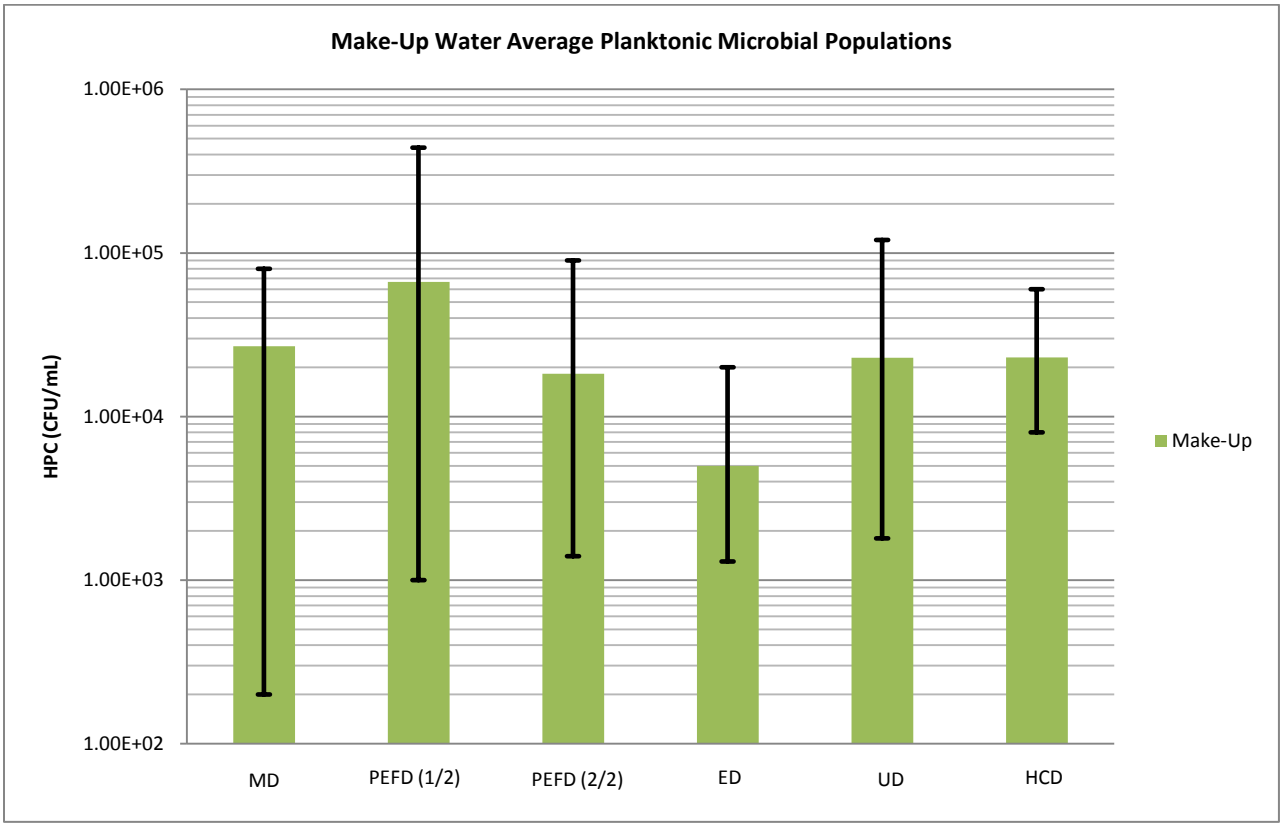


Figure 9 – Average make-up water planktonic microbial population (HPC) during each device trial. Error bars represent range of observed values (maximum and minimum)

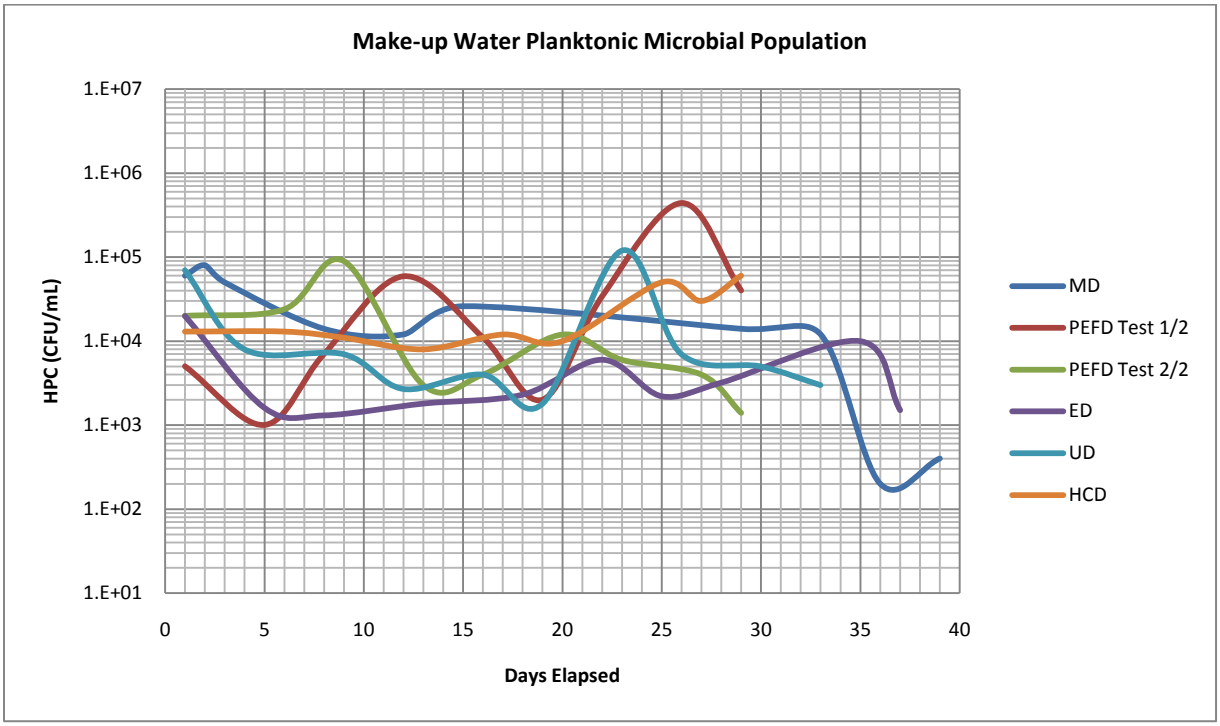


Figure 10 – Make-up water planktonic microbial populations (HPC) for each device trial

3.1.2 Summary

The average heterotrophic plate count for the make-up water over the course of the entire investigation was $10^{4.4}$ CFU/mL. While this value is higher than the EPA mandatory potable water quality standard (< 500 CFU/mL or < $10^{2.7}$ log CFU/mL), it does not demonstrate an excessive or unrealistic level of microbial activity in the make-up water. Planktonic heterotrophic plate count values for a variety of potential make-up water sources are shown in **Table 6**. The water sources highlighted in this table include household taps, which may be used as make-up water sources in small-scale cooling systems, as well as potential natural make-up

water sources for large-scale cooling systems such as lakes and harvested rainwater. **Table 6** indicates that, while the average make-up water planktonic microbial population used in this study exceeds potable water quality standards, it is comparable to other potential make-up water sources.

Table 6 – Concentrations of heterotrophic plate count (HPC) bacteria in various water sources

Water Source	HPC Planktonic Bacteria Count (log CFU/mL)	Reference(s)
Drinking water from household taps (<1 mi. from treatment plant), New Jersey	Range = 1.0 – 4.0+	LeChevallier *et al.*, 1987
Tuscon, AZ household tap	Average = 3.5	Pepper *et al.,* 2004
Tuscon, AZ household tap	Range = 1.0 – 4.7 Average = 4.0	Chaidez & Gerba, 2004
Warm tap water, hospital building	Average = 4.8	Sheffer et al., 2005
Rainwater harvesting system, Seoul, Korea	Range = 3.2 – 3.3	Amin & Han, 2009
Hot water systems, Copenhagen, Denmark	Average = 4.0+	Ovesen *et al.,* 1994; Bagh *et al.*, 2002
Hot water tank, apartment building, Copenhagen, Denmark	Range = 4.8 – 5.0	Bagh *et al.,* 2003
Michigan freshwater lakes (bulk water)	Average = 3.5	Jones *et al.,* 1991
16 community taps and 5 industrial process water basins	Range = 2.7 – 6.0	Jousimies-Somer *et al.*, 1993
Cooling tower water (basins)	Range = 3.0 – 7.0	Bentham, R.H., 1993
Drinking water distribution systems, Durham and Raleigh, NC	Range = <-1.0 – 4.0+	Zhang & DiGiano, 2002
Drinking water distribution systems, Irvine and Garden Grove, CA	Range = <2.0 – 3.4+	Ridgway & Olson, 1982
ASHRAE Project 1361-RP cooling tower make-up water	**Range = 2.3 – 5.6 Average = 4.4**	

3.2 T1 (CONTROL) CONDITIONS

Because the T1 (Control) tower was operated under similar conditions during all device trials except for PEFD Trial 2/2, the data collected from T1 (Control) during each individual device trial may be consolidated and analyzed in order to determine whether or not operating conditions were comparable between the trial. Complete data collected throughout the T1 (Control) tests are shown in Appendix B.2.

3.2.1 T1 (Control) System Operation

Average values for all of the experiments with T1 (Control) are shown in **Table 7**. The target temperature differential throughout the investigation was 10 °F, while a temperature differential of approximately 9-13 °F was maintained for all NCD tests. The only exception was the test conducted with the MD, where operational problems with the heating unit in T1 (Control) resulted in a slightly larger temperature differential.

Table 7 – Average values for T1 (Control)

	T1 (Control)	
	Average	Standard Deviation
Temperature Entering Tower (°F)	99.3	3.1
Sump Temperature (°F)	88.3	3.2
Daily Make-up Water Consumption (gal)	115	7
Daily Blowdown (gal)	17	6
Temperature Differential (°F)	11.0	1.5
Conductivity (mS/cm)	1.174	0.215
pH	8.64	0.10
Alkalinity (mg/L as $CaCO_3$)	113	21
Calcium Hardness (mg/L as $CaCO_3$)	205	88
Magnesium Hardness (mg/L as $CaCO_3$)	122	47
Total Hardness (mg/L as $CaCO_3$)	328	111
TDS (mg/L)	853	165
LSI	1.23	0.29
RSI	6.19	0.52
PSI	7.30	0.56
Planktonic HPC (CFU/mL)	6.77E+05	1.02E+06
Sessile HPC (CFU/cm^2)	2.57E+06	3.66E+06

Throughout each of the trials, the average temperature entering the tower was 99.3 °F and the average sump temperature was 88.3 °F. The temperature entering the tower ranged between 95-105 °F, while the sump temperature ranged between 80-98 °F. This high range of fluctuation observed in the sump was due to large temperature changes caused by make-up water feeding. Since the make-up water was acquired from a cold water faucet and stored at room temperature, it maintained significantly lower temperature than the bulk water. As a result, when make-up water feeding occurred, the most profound impact on tower system temperature was observed in the sump, where the make-up water was discharged.

The average conductivity for all device trials (excluding PEFD Trial 2/2) was 1.174 mS/cm. Conductivity values were higher during PEFD Trial 2/2 in order to achieve a higher

cycles of concentration. The average pH throughout the entire investigation was 8.64, with T1 (Control) maintaining an equilibrium pH in the range of 8.40 – 8.80.

The average alkalinity and hardness in T1 (Control) were 113 and 328 mg/L as $CaCO_3$, respectively. The average LSI for T1 (Control) was 1.23, indicating that conditions were strongly scale forming. The average RSI was 6.19, indicating stable conditions in T1 (Control), while the average PSI was 7.30, indicating moderate scale-forming conditions. A combined analysis of these three indices indicates that conditions in T1 (Control) were mildly to moderately scale-forming for the duration of the investigation.

3.2.2 Biological Parameters

The average planktonic microbial population (enumerated by heterotrophic plate count using a pour plate method) for each device trial is shown in **Figure 11**. Throughout each device trial, a planktonic population of between 10^5 – 10^6 CFU/mL was maintained in the control tower, with the HCD trial generating average microbial counts slightly lower than 10^5 CFU/mL. The average planktonic heterotrophic plate count over the course of the six device trials was 677,000 CFU/mL, fulfilling the goal outlined in the original scope of work of maintaining a control microbial population on the order of magnitude of 10^5 CFU/mL. **Figure 12** depicts variations in planktonic HPC throughout each NCD trial. Since the project scope of work target of 10^5 CFU/mL in the control tower was achieved, no additional microbial seeding of the make-up water was employed for any of the device trials or chlorination tests.

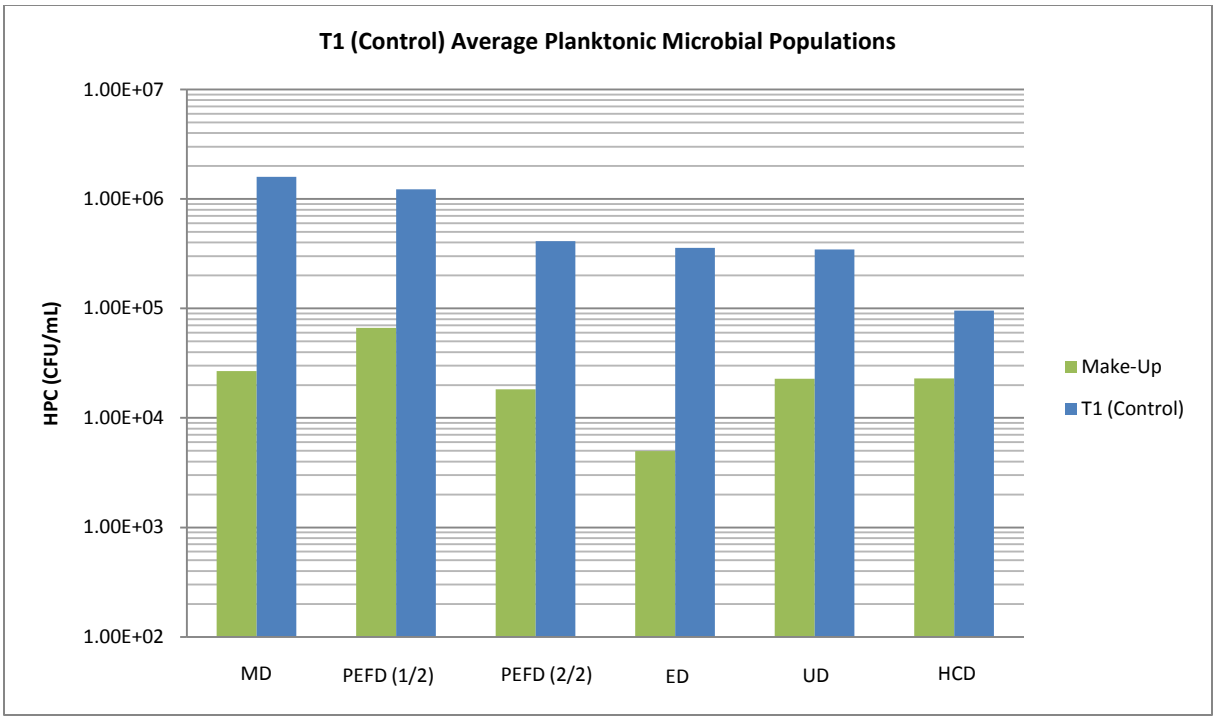


Figure 11 – T1 (Control) average planktonic microbial populations for each device trial

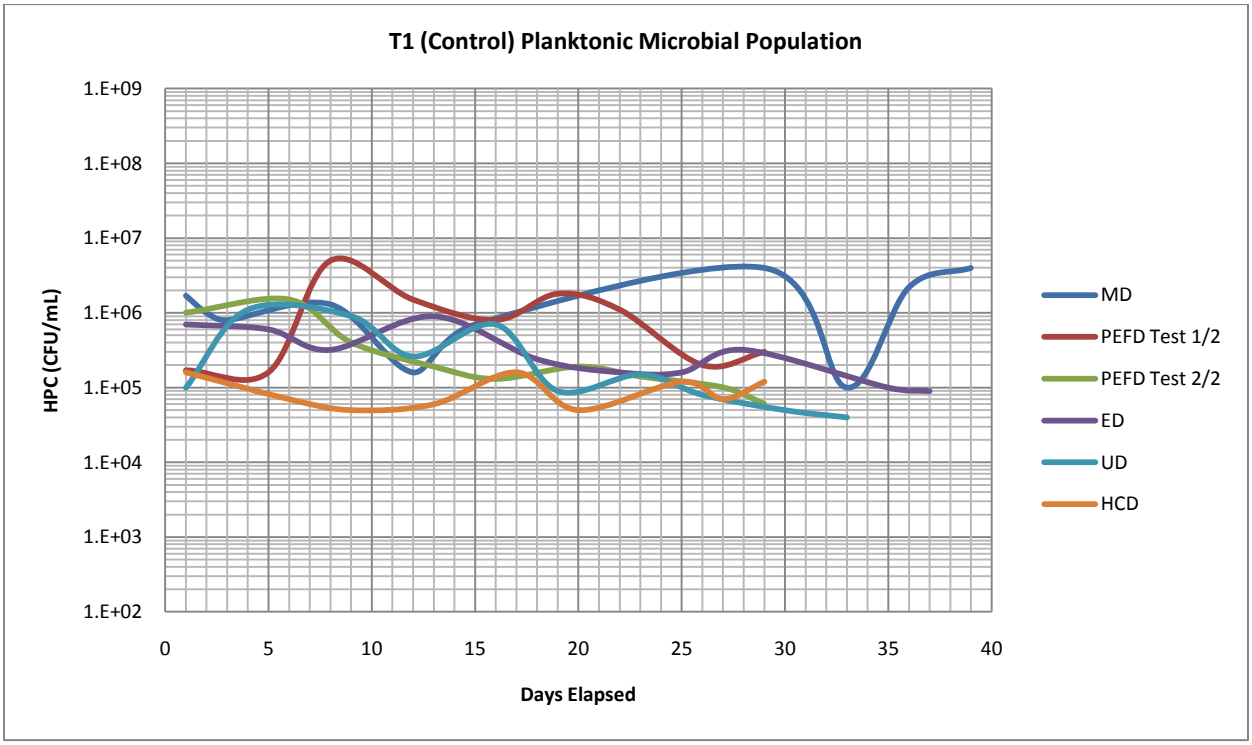


Figure 12 – T1 (Control) planktonic microbial population (HPC) for each device trial

A statistical analysis was used to compare the planktonic heterotrophic plate counts in T1 (Control) for each of the device trials. The results of this statistical analysis are shown in **Table 8**. This analysis indicates that there is a slight difference between HPC data in T1 (Control) for the MD trial and the UD trial. However, the observed differences were less than 1 log value. Ambient temperature changes resulting from changing seasons over the course of the experiment may account for some of the observed difference between early device trials and later ones. During each device trial, T1 (Control) maintained a microbial population in excess of 10^5 CFU/mL, as was outlined in the initial scope of work for this project.

The sessile microbial population in T1 (Control) over the course of the entire study is shown in **Figure 13** with an average sessile heterotrophic plate count of 2.6 x 10^6 CFU/cm^2. A paired t-test was used to compare the T1 (Control) heterotrophic plate count data among different device trials. The resulting p-value of 0.438 indicates that there was no statistically significant difference between the sessile heterotrophic plate counts observed in T1 (Control) during each of the 6 device trials.

Table 8 – Pairwise comparisons of planktonic HPC in T1 (Control)

Level	n	Mean	S.E.
2	7	5.985537	.1613863
3	9	5.809726	.1770747
4	9	5.374876	.1545532
5	8	5.54703	.1012947
6	9	5.332011	.1676016
7	9	5.564345	.0430838

Note:
Level 2 is MD
Level 3 is PEFD (Trial 1/2)
Level 4 is PEFD (Trial 2/2)
Level 5 is ED
Level 6 is UD
Level 7 is HCD

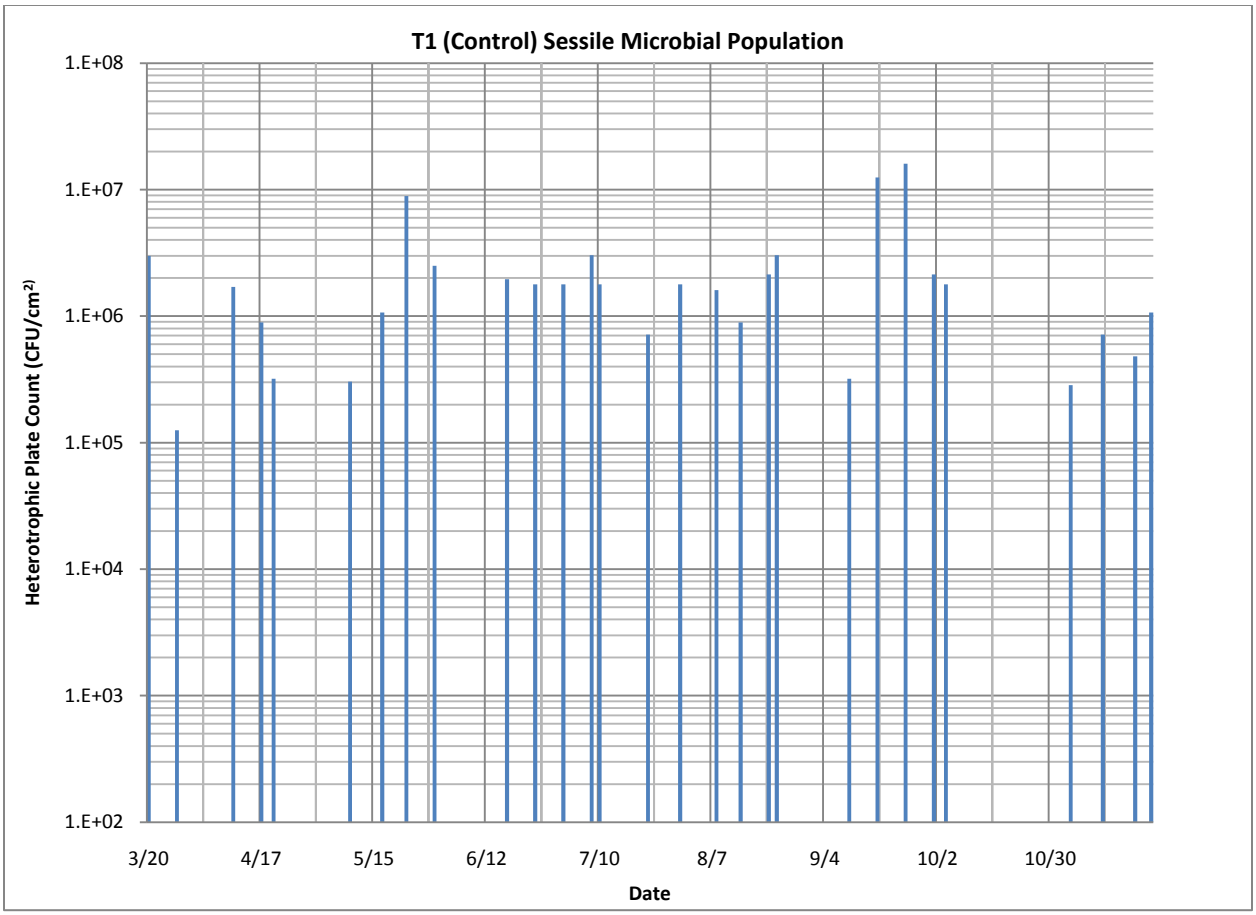


Figure 13 – T1 (Control) combined sessile microbial population

3.2.3 Summary

Conditions in T1 (Control) were approximately the same for each of the six device trials. Temperature fluctuations were the result of changes in the ambient outdoor temperature over the course of the investigation. Fluctuations in chemical parameters, such as conductivity, pH, and alkalinity, were the result of changes in make-up water quality. Total hardness data showed the largest fluctuation over the course of the investigation, which was significantly reduced by the preparation of new standards for hardness measurements. Scaling index calculations indicate

that mild to moderate scaling potential was maintained in T1 (Control) throughout the investigation. Planktonic microbial populations above 10^5 CFU/mL and sessile microbial populations above 10^6 CFU/cm^2 were maintained in T1 (Control) over the course of each device trial.

Another very important measure of reproducibility that was achieved in this study is discussed in Section 4.7, where the results of three separate chlorination tests are presented. It is important to note that the chlorination tests demonstrated not only the reproducibility of the experimental investigations conducted in this study but also the effectiveness of a conventional chemical biofouling control approach for pilot-scale cooling systems.

4.0 DEVICE TRIAL RESULTS AND DISCUSSION

The following section provides the results from each individual device trial. These include operational, chemical, and biological parameters. Biological data are provided in the following sections, while chemical and operational data are provided in the corresponding appendices.

4.1 MAGNETIC DEVICE (MD) TRIAL

The investigation of the magnetic device began on 3/13/09 and lasted until 4/20/09. The heating systems in T1 (Control) and T2 (Device) had to be rewired on 3/29/09 due to electrical problems. Additionally, a solenoid valve malfunction led to a pause in the operation of T1 (Control) from 4/11 – 4/15. Data collected during the test with magnetic device are shown in Appendix C. Operational and chemical data for the make-up water and each of the two system towers are included in Appendix C.1. Statistical analyses of the chemical and water consumption data collected during this investigation are shown in Appendix C.2. This analysis contains data from an earlier trial run which was terminated due to operational problems. Photographs of each of the tower systems before and after the test of the magnetic device are shown in Appendix C.3.

4.1.1 Tower System Operation

Tower system operational data for the MD trial are shown in **Figure C1-C19**. The temperature differential (**Figure C1**) observed for each tower was between 9-15 ºF over the course of the experiment. Operational problems with the heating system of T1 (Control) led to erratic temperature differentials in the second half of the test period. Make-up water consumption rates (**Figure C2**) were similar for the two towers over the course of the experiment (T1 (Control) average = 115 gal/day, T2 (Device) Average = 115 gal/day), while blowdown levels (**Figure C3**) were higher for T2 (Device) than for T1 (Control) (T1 (Control) average = 12 gal/day, T2 (Device) Average = 18 gal/day). The pause in operation experienced by T1 (Control) from 4/11 – 4/15 led to the difference between observed make-up water feed rates and blowdown levels. Based on manufacturer's explanation regarding the mechanism by which the magnetic device decreases scaling, reduced surface bleed and cumulative water consumption are to be expected in the device tower (T2).

Tower temperatures for T1 (Control) remained in the range of 87-105°F, as is shown on **Figure C4**. This temperature range offers neither an advantage nor a disadvantage for the planktonic and sessile microbial populations present in the tower. The slightly higher tower temperatures observed in T1 (Control) towards the end of the trial period may be responsible for the higher planktonic microbial populations observed on 4/10, 4/17, and 4/20. However, during this period the temperature range of T2 (Device) was approximately 95-107 °F, as is shown in **Figure C5**. The behavior of both towers' temperature profiles is comparable, and may be directly linked to changes in ambient temperature (**Figure C6**) and relative humidity (**Figure C7**) during the course of the experiment. Any temperature effects on microbial growth would have been observed in both T1 (Control) and T2 (Device).

Average values and standard deviations for all parameters measured during this investigation are presented in **Table 9**. Additional chemical data collected during this investigation are shown in **Figure C8 – Figure C16**. The fluctuations in these measurements during the beginning portion of the experiment, as well as following periods of non-operation, indicate that several days of tower operation are required in order to achieve steady state operation. Conductivity measurements were somewhat erratic during the experiment, as can be seen in **Figure C8**. The blowdown controllers were reset at various times throughout the course of the experiment to achieve approximately 4-5 cycles of concentration. However, improper calibration of the conductivity data collection probes may have resulted in the collection of seemingly disparate data.

Table 9 – Average values for MD trial

	T1 (Control)		T2 (Device)	
	Average	Standard Deviation	Average	Standard Deviation
Temperature Entering Tower (°F)	93.4	5.5	97.9	3.4
Sump Temperature (°F)	85.0	3.7	84.9	3.1
Daily Make-up Water Consumption (gal)	113	36	115	27
Daily Blowdown (gal)	12	8	18	9
Conductivity (mS/cm)	0.867	0.155	0.819	0.146
pH	8.56	0.08	8.61	0.10
ORP (mV)	257	22	258	36
Alkalinity (mg/L as $CaCO_3$)	134	13	133	16
Calcium Hardness (mg/L as $CaCO_3$)	230	183	257	158
Magnesium Hardness (mg/L as $CaCO_3$)	67	25	78	14
Total Hardness (mg/L as $CaCO_3$)	300	189	349	159
TDS (mg/L)	632	179	672	144
LSI	1.12	0.44	1.19	0.46
RSI	6.31	0.81	6.19	0.84
PSI	7.17	0.76	6.86	0.76
Planktonic HPC (CFU/mL)	1.60E+06	1.42E+06	6.79E+05	6.79E+05
ATP Microbial Equivalents (MEQs/mL)	1.02E+06	1.06E+06	1.02E+06	5.94E+05
Sessile HPC (CFU/cm^2)	1.21E+06	1.17E+06	2.68E+06	1.53E+06

Chloride measurements for each tower are shown in **Table C1**, and these measurements indicate that the cycles of concentration for this run were slightly higher than 5 for each of the towers. Somewhat erratic results for some of the chemical parameters measured towards the end of the experimental run may be the result of the fact that sampling was performed at different times before and after make-up water addition and blowdown, which provided different dilution factors.

Using the chemical data collected during this investigation, three scaling indices were calculated for each of the towers. Both towers produced comparable values for each of the scaling indices analyzed. The Langelier Saturation Index value observed was approximately 1.12 for T1 (Control) and 1.19 for T2 (Device) (**Figure C17**), indicating that the water in each tower had strong scaling potential. The Ryznar Stability Index value observed was approximately 6.31 for T1 (Control) and 6.19 for T2 (Device) (**Figure C18**), indicating that T1 (Control) had a slight tendency to dissolve scale, while T2 (Device) was neutral with regards to scale formation. The average Puckorius Scaling Index value observed for T1 (Control) was 7.17, and for T2 (Device) it was 6.86 (**Figure C19**), indicating that the water in each tower had high scale-dissolving potential. Based on a comparison of the observed values of these three indices, it may be concluded that the water in each tower maintained comparable moderate scaling potential over the course of the experiment.

4.1.2 Biological Parameters

Analysis of the biological data collected during the second evaluation of the magnetic device indicates that the magnetic device offers little to no control of planktonic and sessile microbial populations.

Observations of the biological population present in each tower (**Figure 14** - **Figure 16**) indicate that each tower contained comparable levels of microbial activity both in the bulk water and in the system biofilms throughout the course of the experiment. No planktonic biological sample was taken from T1 on 4/6 since the tower was not operational.

The planktonic heterotrophic plate counts observed in each of the tower systems throughout the MD trial are shown in **Figure 14**. This figure reveals that the heterotrophic plate count in T1 (Control) was higher than that of T2 (Device) on 7 out of the 10 days for which data are available for both tower systems. However, there was less than 1 log value difference on each of these 7 days. A statistical analysis of the heterotrophic plate count data is shown in **Table 10**, and this analysis indicates that the observed difference between the plate counts for T1 (Control) and T2 (Device) was not statistically significant. At no point during the device trial did T2 (Device) maintain a microbial population below the industry standard of 10^4 CFU/mL.

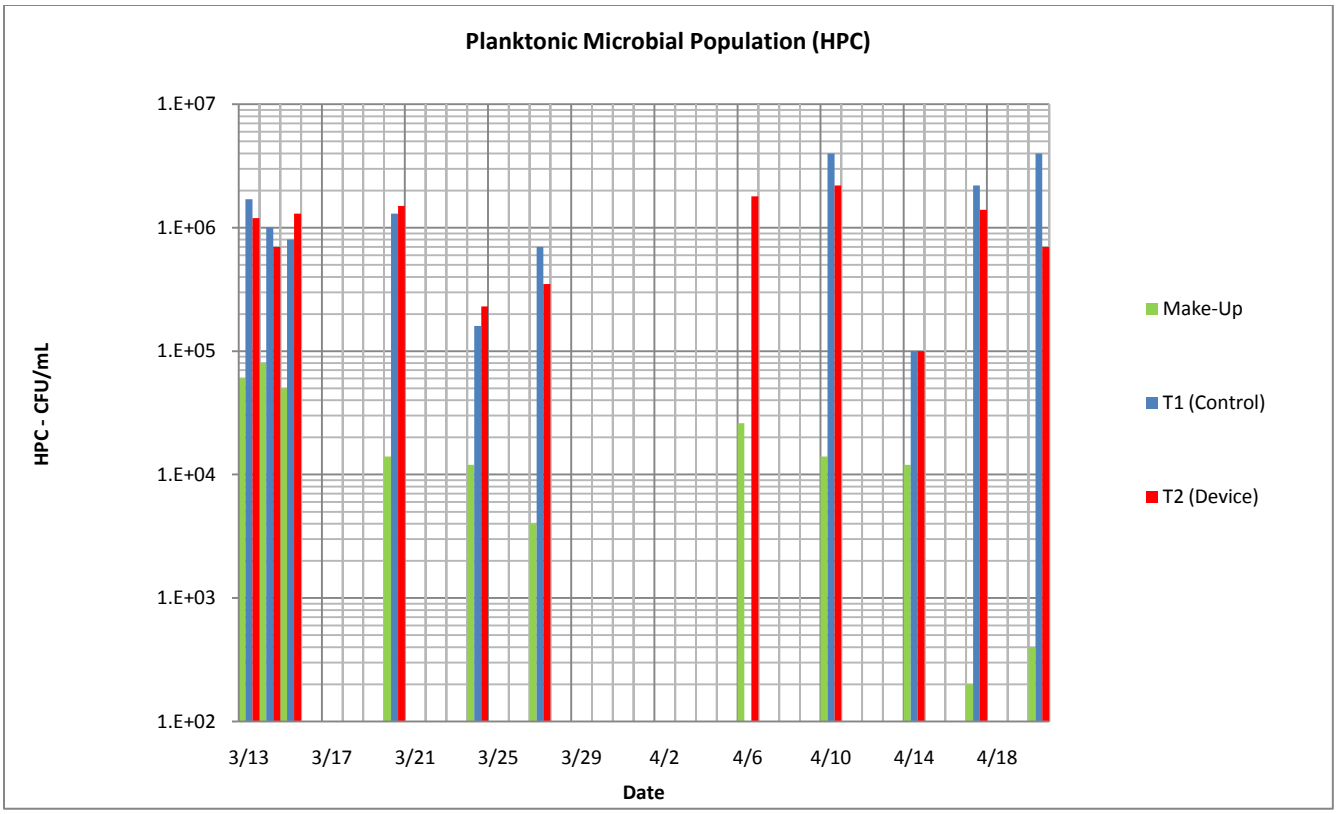


Figure 14 – Planktonic microbial populations (heterotrophic plate count) for MD trial

Table 10 – Planktonic HPC statistical analysis for MD trial

(Planktonic heterotrophic plate counts were transformed to Log 10 data and compared using a paired t-test. Log 10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.15

```
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
   logt1 |      10    5.984324    .1690709    .5346493    5.601859    6.366789
   logt2 |      10    5.845376    .1338754    .4233512    5.542529    6.148223
---------+--------------------------------------------------------------------
    diff |      10    .1389484    .0879725    .2781936   -.0600592    .3379561
------------------------------------------------------------------------------
     mean(diff) = mean(logt1 - logt2)                              t =   1.5795
 Ho: mean(diff) = 0                         degrees of freedom =        9

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.9257         Pr(|T| > |t|) = 0.1487          Pr(T > t) = 0.0743
```

Based on the results of this paired t-test, there was no significant difference in planktonic heterotrophic plate counts between T1 (Control) and T2 (Device).

Relatively high levels of microbial activity were observed in the make-up water throughout the course of this experiment. The make-up water heterotrophic biological population was between 200 – 80,000 CFU/mL, with an average value of 25,000 CFU/mL, which would have resulted in high levels of microbial seeding during periods of make-up water addition to the system. As discussed in Section 3, the make-up water used for the experiment was dechlorinated using an activated carbon column. After passing through the column, the make-up water was then stored in four 125-gallon tanks. While the tanks received fresh water on daily basis, this storage period allowed for microbial growth to occur, resulting in the high levels of make-up water microbial activity.

The planktonic microbial population in each tower system was also monitored by measuring the cellular ATP. Measured ATP concentrations were converted to microbial equivalents as previously described in this report, and these measurements are shown in **Figure 15**. The observed planktonic microbial population was higher in T2 (Device) than in T1 (Control) on 7 of the 10 days for which data are available for both tower systems. In each instance, this difference is less than 1 log value. A statistical analysis of the microbial populations enumerated by ATP measurement is shown in **Table 11**. This analysis indicates that the differences between the microbial populations observed in T1 (Control) and T2 (Device) using ATP measurements are not statistically significant.

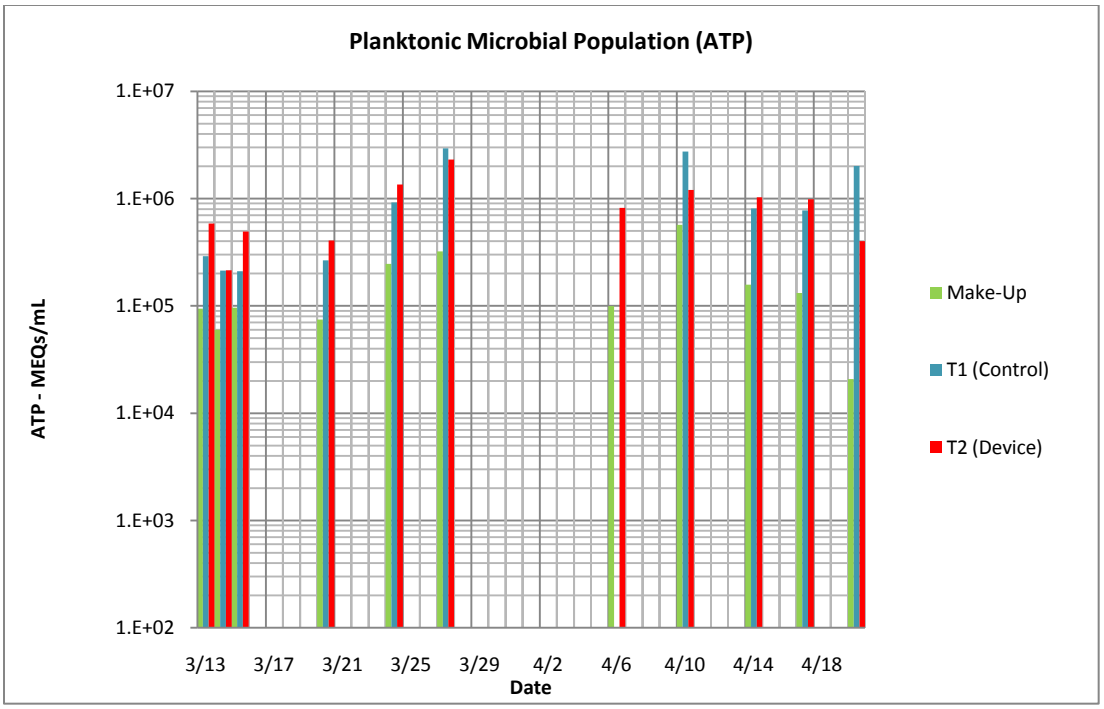


Figure 15 – Planktonic microbial populations (ATP measurement) for MD trial

Table 11 – Planktonic ATP statistical analysis for MD trial

(Planktonic ATP microbial equivalent levels were transformed to Log 10 data and compared using a paired t-test. Log10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.95

```
------------------------------------------------------------------------------
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
  logatp |      10    5.851842    .1426523    .4511063     5.52914    6.174544
 logatp2 |      10    5.858571    .0978048    .3092859    5.637321    6.079821
---------+--------------------------------------------------------------------
    diff |      10    -.006729    .1022969    .3234912   -.2381406    .2246827
------------------------------------------------------------------------------
     mean(diff) = mean(logatp - logatp2)                           t =  -0.0658
 Ho: mean(diff) = 0                           degrees of freedom =        9

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.4745         Pr(|T| > |t|) = 0.9490          Pr(T > t) = 0.5255
```

Based on the results of this paired t-test, there was no significant difference in ATP microbial equivalent levels between T1 (Control) and T2 (Device).

The sessile microbial populations observed in each tower system throughout the course of this device trial are shown in **Figure 16**. This figure reveals that the sessile microbial population was higher in T2 (Device) than in T1 (Control) on 4 out of 5 sampling days. Sessile heterotrophic plate counts for T2 (Device) were in excess of 10^6 CFU/cm^2 on 4 out 5 sampling days. A statistical analysis of the sessile heterotrophic plate counts for each tower system is shown in **Table 12**. This analysis reveals that no statistically significant difference between sessile heterotrophic plate counts in T1 (Control) and T2 (Device) was observed.

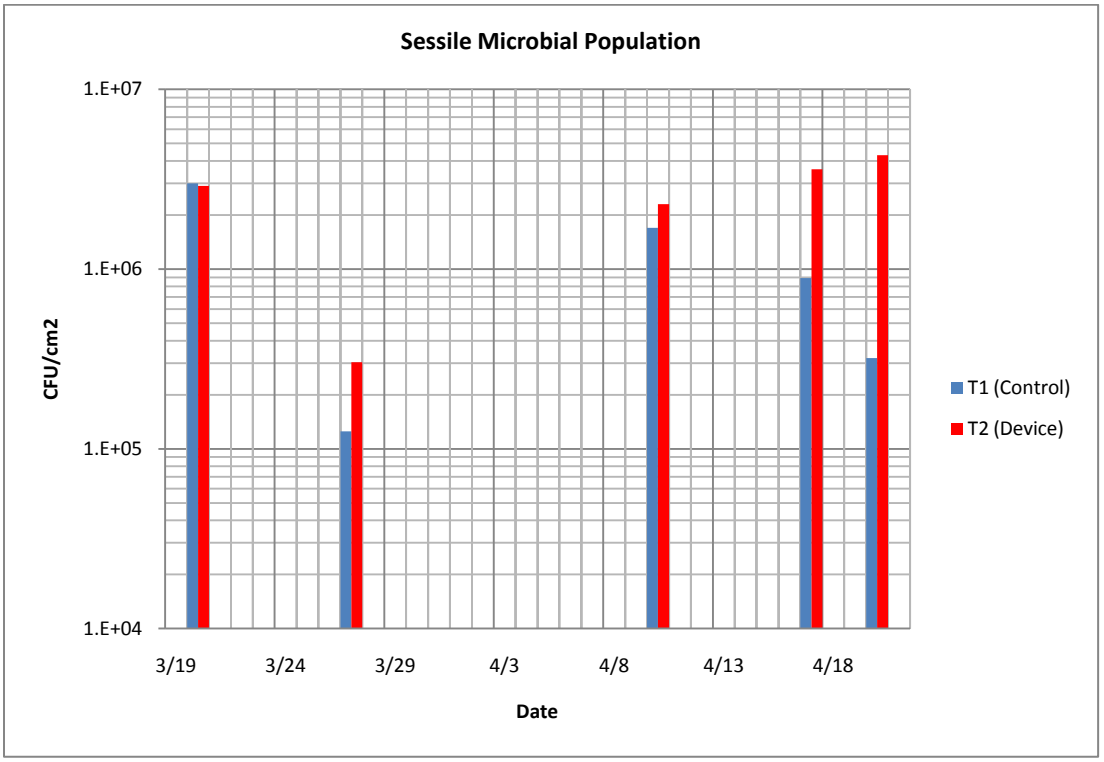


Figure 16 – Sessile microbial population for MD trial

Table 12 – Sessile HPC statistical analysis for MD trial

(Sessile heterotrophic plate counts (**Figure 16**) were transformed to Log 10 data and compared using a paired t-test. Log10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.09

```
------------------------------------------------------------------------------
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
 logbio1 |       5    5.851804    .2483899    .5554167    5.162163    6.541445
 logbio2 |       5    6.299068    .2094168    .4682702    5.717634    6.880502
---------+--------------------------------------------------------------------
    diff |       5   -.4472639    .2008155    .4490371   -1.004817    .1102893
------------------------------------------------------------------------------
     mean(diff) = mean(logbio1 - logbio2)                      t =  -2.2272
 Ho: mean(diff) = 0                               degrees of freedom =        4

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.0449         Pr(|T| > |t|) = 0.0899          Pr(T > t) = 0.9551
```

Based on the results of this paired t-test, there was no significant difference in sessile heterotrophic plate counts between T1 (Control) and T2 (Device).

Since the magnetic device was unable to demonstrate any significant control over the microbial population (both planktonic and sessile) in the experimental towers, the experiment was terminated before conducting the second phase (colonization of the system followed by device installation).

4.1.3 Summary

The results presented in this report demonstrate that the magnetic device did not significantly reduce biological activity compared to the "control" tower. Planktonic heterotrophic plate counts, ATP measurements, and sessile heterotrophic plate counts from T1 (Control) and T2

(Device) showed no significant differences at any point during the investigation. Tower operational conditions were comparable throughout the course of the device trial

4.2 PULSED ELECTRIC FIELD DEVICE (PEFD) TRIAL 1/2

The first investigation of the PEFD began on 5/2/09 and it continued uninterrupted until 5/30/09. Data collected during this investigation are included in Appendix D. Operational data collected during this investigation are shown in Appendix D.1. Statistical analyses of the chemical data analyzed during the investigation are shown in Appendix D.2. Photographs of the system before and after the test are shown in Appendix D.3.

4.2.1 Tower System Operation

Tower system operational data for this device trial are shown in **Figure D1 – Figure D19** . A temperature differential of approximately 9-13 °F was maintained for each tower throughout the majority of the investigation (**Figure D1)**. Since temperature differential calculations were determined using manual temperature readings, it is likely that the low observed temperature differentials (< 10 °F) were the result of measurements being taken soon after the towers received make-up water. Receiving make-up water cools down the sump temperature significantly, and this may have led to the low measured temperature differentials. The quantity of make-up water consumed by each tower throughout the course of the investigation was approximately the same (**Figure D2**). T1 (Control) exhibited higher blowdown volumes initially (**Figure D3)**, and T2 (Device) gradually approached the blowdown rate exhibited by Tower 1 as the experiment progressed. Tower temperature profiles are shown in **Figure D4** (T1) and **Figure D5** (T2), with fluctuations occurring as a result of changing ambient conditions such as temperature (**Figure D6**) and relative humidity (**Figure D7**).

Average values and standard deviations for all parameters measured during this investigation are shown in **Table 13**. Chemical data collected during the course of this experiment are shown in **Figure D8 – Figure D16**. Continuous conductivity data collected during the investigation are shown in **Figure D8**. The conductivity observed in T1 (Control) was slightly higher than that observed in T2 (Device) for the duration of the experiment. Although both blowdown controllers were set at identical values (1.20 mS/cm), the blowdown controller for T2 (Device) was not functioning properly. As a result, the conductivity levels which triggered blowdown in T2 (Device) were lower than the setpoint, resulting in a lower overall conductivity. This trend may also be observed with the continuous pH monitoring of the systems (**Figure D9**). Make-up water conductivity ranged from 0.284 – 0.312 mS/cm, while the make-up water pH ranged from 6.82 – 7.53 (**Figure D11**).

Table 13 – Average values for PEFD (Trial 1/2)

	T1 (Control)		T2 (Device)	
	Average	Standard Deviation	Average	Standard Deviation
Temperature Entering Tower (°F)	102.0	2.1	100.7	3.8
Sump Temperature (°F)	90.7	2.4	88.9	3.5
Daily Make-up Water Consumption (gal)	111	24	109	28
Daily Blowdown (gal)	20	10	15	8
Conductivity (mS/cm)	1.057	0.045	0.948	0.038
pH	8.61	0.09	8.45	0.04
ORP (mV)	228	12	197	64
Alkalinity (mg/L as $CaCO_3$)	132	10	117	10
Calcium Hardness (mg/L as $CaCO_3$)	266	43	250	36
Magnesium Hardness (mg/L as $CaCO_3$)	206	27	185	21
Total Hardness (mg/L as $CaCO_3$)	472	65	436	52
TDS (mg/L)	980	74	934	64
LSI	1.42	0.15	1.18	0.10
RSI	5.71	0.05	6.09	0.19
PSI	6.70	0.16	6.94	0.17
Planktonic HPC (CFU/mL)	1.23E+06	1.54E+06	5.50E+05	4.63E+05
ATP Microbial Equivalents (MEQs/mL)	9.98E+05	6.92E+05	9.65E+05	4.49E+05
Sessile HPC (CFU/cm^2)	3.19E+06	3.91E+06	1.95E+06	1.78E+06

Chloride measurements for each tower are shown in **Table D1**, and these measurements indicate that T1 (Control) was operating at near 6.5 cycles of concentration, while T2 (Device) was operating at near 6 cycles of concentration. These values are higher than the target cycles of concentration (4-5), which is most likely due to the erratic nature of the make-up conductivity and the collection of solids on the conductivity probes which are used to control blowdown.

The chemical parameters measured during this investigation were used to calculate 3 scaling indices commonly used in the water treatment industry: The Langelier Saturation Index (LSI), the Ryznar Stability Index (RSI), and the Puckorius Scaling Index (PSI). At equilibrium, T1 (Control) had an average LSI value of approximately 1.42, while T2 (Device) had an LSI value of 1.18 (**Figure D17**). These values suggest that the water in each tower system had strong scaling potential. The RSI values for T1 (Control) and T2 (Device) at equilibrium were 5.71 and 6.09, respectively, indicating that T1 (Control) had slight scale formation potential and T2 (Device) was neutral with regards to scaling potential (**Figure D18**). The equilibrium PSI values for T1 (Control) and T2 (Device) were 6.70 and 6.94, respectively, also indicating moderate scale-dissolving potential (**Figure D19**). A combined analysis of each of these scaling indices indicates that the towers were operating under very similar conditions.

4.2.2 Biological Parameters

Biological data collected during this investigation are shown in **Figure 17** – **Figure 19.** Analysis of this data indicates that the device did not exhibit any significant biocidal effects. No significant reduction in planktonic or sessile microbial activity was observed in T2 (Device). Biological data

collected during this device trial were combined with data from PEFD Trial 2/2, and the statistical analyses performed using this combined data are discussed in Section 4.3.2.

Heterotrophic plate counts for each of the towers systems were measured throughout the course of the device trial, and these measurements are shown in **Figure 17**. Although T2 (Device) demonstrated lower planktonic heterotrophic plate count values on 7 out of 9 of the biological sampling days, the observed differences between the plate counts from T1 (Control) and T2 (Device) were less than 1 log value. On each sampling day, the microbial population in T2 (Device) was higher than the industry standard of 10^4 CFU/mL.

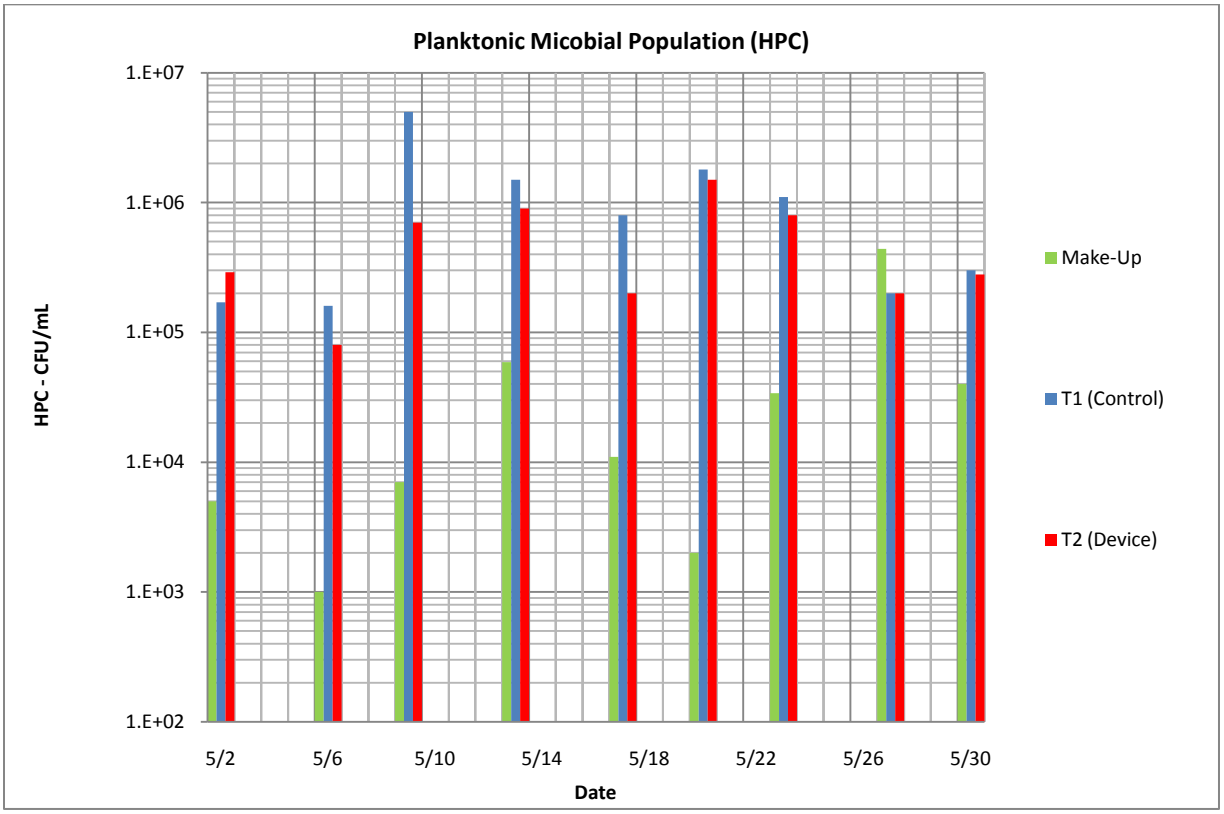


Figure 17 – Planktonic microbial populations (HPC) for PEFD (Trial 1/2)

Microbial populations were also enumerated using measurement of cellular ATP, and the results of these measurements are shown in **Figure 18**. ATP concentrations were converted to microbial equivalents according to the equation previously discussed in this report. Microbial equivalent levels were comparable in each tower system throughout the device trial.

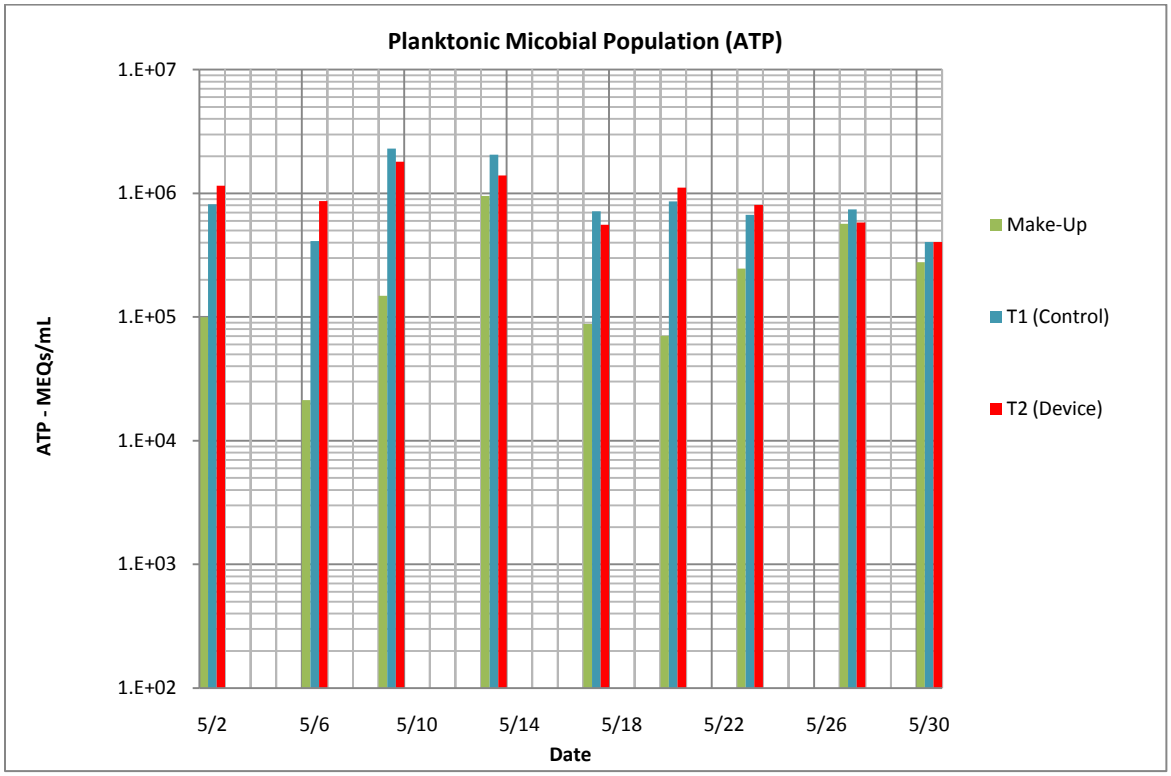


Figure 18 – Planktonic microbial population (ATP measurement) for PEFD (Trial 1/2)

The sessile microbial populations for each tower system over the course of the device trial are shown in **Figure 19**. Each tower system maintained a comparable level of sessile microbial activity, and all measured heterotrophic plate counts were in excess of 10^5 CFU/cm^2. This indicates that the device did not have a significant effect on sessile microbial growth.

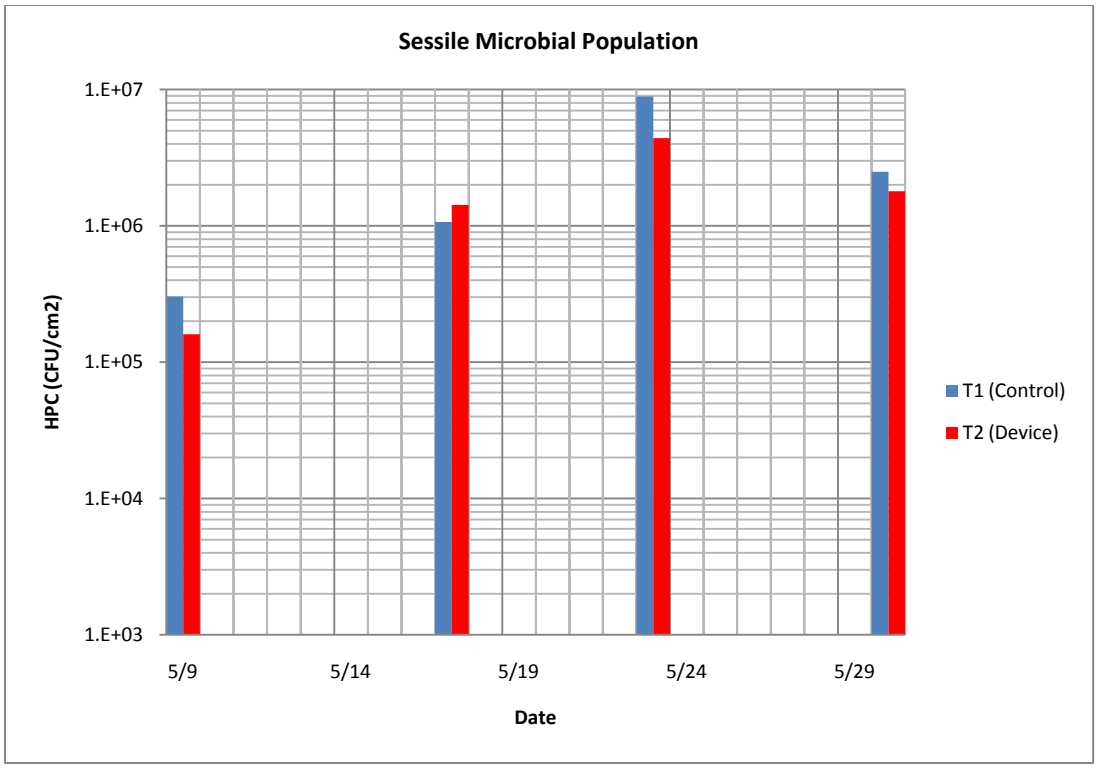


Figure 19 – Sessile microbial population for PEFD (Trial 1/2)

The make-up water contained a heterotrophic biological population between 1,000 – 440,000 CFU/mL during this device trial, with an average value of approximately 67,000 CFU/mL. One sampling date (5/27/09) demonstrated a make-up water microbial population higher than that observed in either of the two tower systems (440,000 CFU/mL). It is likely that this high microbial count was the result of sample contamination since microbial equivalent levels measured using cellular ATP were comparable on this sampling day to other values observed during this device trial. Excluding this outlier, the make-up water contained an observed biological range of 1,000 – 59,000 CFU/mL and an average heterotrophic population of approximately 20,000 CFU/mL. Make-up water biological growth was allowed to occur without any means of biocidal control. No artificial microbial seeding was performed on the make-up water during this or any of the device trials.

Since no significant planktonic or sessile microbial control was observed in this test, Phase II of the device trial (colonization of towers followed by device activation) was not performed. However, an additional Phase I test was performed at a higher cycle of concentration to determine if this would have an effect on biological control using the PEFD. The manufacturer's literature indicates that precipitation of scale helps to remove microorganisms from the system, which may not have occurred to a significant extent during this trial due to the low value of cycles of concentration.

4.2.3 Summary

No biological control resulting from the installation of the PEFD was observed during this trial. According to the manufacturer's literature, this could have been the result of insufficient cycles of concentration employed in this test since the conductivity of the device tower was not as high as is normally encountered in industrial cooling systems. Additional concerns regarding relative humidity and temperature levels in the shower room containing the tower systems and the consistency of the heating units were also addressed prior to the performance of this second device trial. Flexible ducting was installed on the top of each tower system in order to funnel tower exhaust air directly into the ventilation system. Additionally, the fan belt for the roof fan controlling the shower room ventilation system was replaced. These modifications improved ventilation in the shower room, minimizing air recirculation.

4.3 PULSED ELECTRIC FIELD DEVICE (PEFD) TRIAL 2/2

The second investigation of the pulsed electric field device began on 6/12, and it was completed on 7/10. This investigation was performed to determine if higher scaling index would improve biological control in the cooling tower. The primary parameter altered during the second investigation was the blowdown conductivity setpoint. In the previous investigation of the pulsed electric field device, the blowdown conductivity setpoint was chosen to establish a steady state of 4-5 cycles of concentration which results in stable scale forming index. For the second pulsed electric field device investigation, the set point was raised to establish a steady state of 6-7 cycles of concentration. The increased cycles of concentration were verified by total dissolved solids, conductivity, and chloride measurements, although calcium hardness and alkalinity of the make-up water decreased during this device trial in comparison to PEFD Trial 1/2 (See Appendix B.1, **Table B1**).

An electrical failure caused T2 (Device) to go offline on 6/21. T2 (Device) went back online on 6/23 following the installation of a new pump, and it remained online for the duration of the investigation. All data collected during this investigation is included in Appendix E. Operational and chemical data collected during this investigation are shown in Appendix E.1, and statistical analyses of the data collected during both device trials (PEFD Trials 1/2 and 2/2) are shown in Appendix E.2. Photographs of each tower system before and after the device trial are shown in Appendix E.3. Photographs of the last two biofilm sampling coupons taken from each tower during this investigation are shown in Appendix E.4.

4.3.1 Tower System Operation

Tower operational data are shown in **Figure E1 – Figure E19**. The temperature differentials for each tower over the course of the device trial are shown in **Figure E1**, illustrating that a temperature differential of 9-13°F was maintained in each tower throughout the investigation. The differential in T1 (Control) was approximately 1°F lower than that observed in T2 (Device). **Figure E2** demonstrates that make-up water consumption levels were comparable in T1 (Control) and T2 (Device) during this test, as indicated by the similar slopes of make-up consumption volume over time. **Figure E3** demonstrates that T1 (Control) and T2 (Device) maintained comparable cumulative blowdown levels throughout the course of the experiment.

Observation of the temperature profiles for T1 (Control) (**Figure E4**) and T2 (Device) (**Figure E5**) indicate that the two towers were operating under steady and consistent conditions for the majority of the experimental run. Tower operation was halted on 6/22 – 6/23 for T2 (Device) due to operational failure. Fluctuations in the temperature profiles of each tower are the result of changes in ambient conditions such as temperature (**Figure E6**) and relative humidity (**Figure E7**). Ambient relative humidity in the shower room was decreased for this experimental run by the installation of two dehumidifiers and flexible ducting connecting each tower's air outlet directly to the ceiling-mounted exhaust duct.

Average values and standard deviations for the parameters measured during this investigation are shown in **Table 14**. Chemical parameters measured throughout the course of this experiment are shown in **Figure E8 – Figure E16**. Values for these parameters were consistently higher during the second trial than were observed during the first trial. Continuous conductivity data are shown in

Figure E8, and continuous pH data are shown in **Figure E9**. T1 (Control) maintained a pH of approximately 8.71, while T2 (Device) maintained a pH of approximately 8.73. Measurements of conductivity and pH indicate that the operating conditions of each tower were nearly identical for the duration of the experimental trial. The blowdown setpoint on each tower was set to a value of 2.10 mS/cm based on the chemical characteristics of the incoming make-up water (**Figure E11**).

Table 14 – Average values for PEFD (Trial 2/2)

	T1 (Control)		T2 (Device)	
	Average	Standard Deviation	Average	Standard Deviation
Temperature Entering Tower (°F)	96.4	1.0	98.6	1.8
Sump Temperature (°F)	85.6	1.7	86.8	1.6
Daily Make-up Water Consumption (gal)	105	23	96	7
Daily Blowdown (gal)	8	6	8	8
Conductivity (mS/cm)	1.980	0.102	1.908	0.203
pH	8.71	0.03	8.73	0.06
ORP (mV)	196	7	246	22
Alkalinity (mg/L as $CaCO_3$)	123	10	114	16
Calcium Hardness (mg/L as $CaCO_3$)	173	3	167	7
Magnesium Hardness (mg/L as $CaCO_3$)	198	11	192	21
Total Hardness (mg/L as $CaCO_3$)	371	11	361	81
TDS (mg/L)	1411	67	1329	155
LSI	1.24	0.05	1.24	0.13
RSI	6.24	0.08	6.25	0.21
PSI	7.35	0.12	7.40	0.25
Planktonic HPC (CFU/mL)	4.12E+05	5.00E+05	1.17E+06	2.24E+06
ATP Microbial Equivalents (MEQs/mL)	4.59E+05	4.12E+05	6.08E+05	1.96E+05
Sessile HPC (CFU/cm^2)	2.07E+06	5.44E+05	1.89E+06	6.14E+05

Three scaling indices were calculated using the chemical parameters observed during this investigation, and these indices may be used to quantify the scaling potential of each tower's water supply. The Langelier Saturation Index for each tower was approximately 1.24 at equilibrium,

indicating that the water in each tower system had strong scaling potential (**Figure E17**). Equilibrium values of the Ryznar Stability Index for each tower were approximately 6.24 for T1 (Control) and 6.25 for T2 (Device) (**Figure E18**), indicating that the water in each system was stable. However, the Puckorius Scaling Index values were approximately 7.35 for T1 (Control) and 7.40 for T2 (Device) at equilibrium, indicating moderate scale-dissolving potential (**Figure E19**). These indices provide conflicting information regarding whether or not the water in each tower system is scale-forming or scale-dissolving.

4.3.2 Biological Parameters

Observation of the biological population present in each tower (**Figure 20** – **Figure 22**) indicates that each tower maintained comparable levels of microbial activity both in the bulk water and in system biofilms throughout the course of the experiment. No significant reduction in microbial activity was observed in the tower with the device. Both towers maintained planktonic microbial populations in excess of acceptable industry standards (10^4 CFU/mL).

Planktonic heterotrophic plate counts for each tower system over the course of the device trial are shown in **Figure 20**. The microbial population was higher in T2 (device) than in T1 (Control) on 7 of the 9 sampling days. On 6/20, the heterotrophic plate count in T2 (Device) was approximately 1 log value higher than in T1 (Control). This was the largest observed difference between the planktonic microbial populations of the two tower systems. A statistical analysis of the combined planktonic heterotrophic plate count data from PEFD Trial 1/2 and Trial 2/2 is shown in **Table 15**. This analysis indicates that the observed difference between the planktonic heterotrophic plate counts for T1 (Control) and T2 (Device) was not statistically significant.

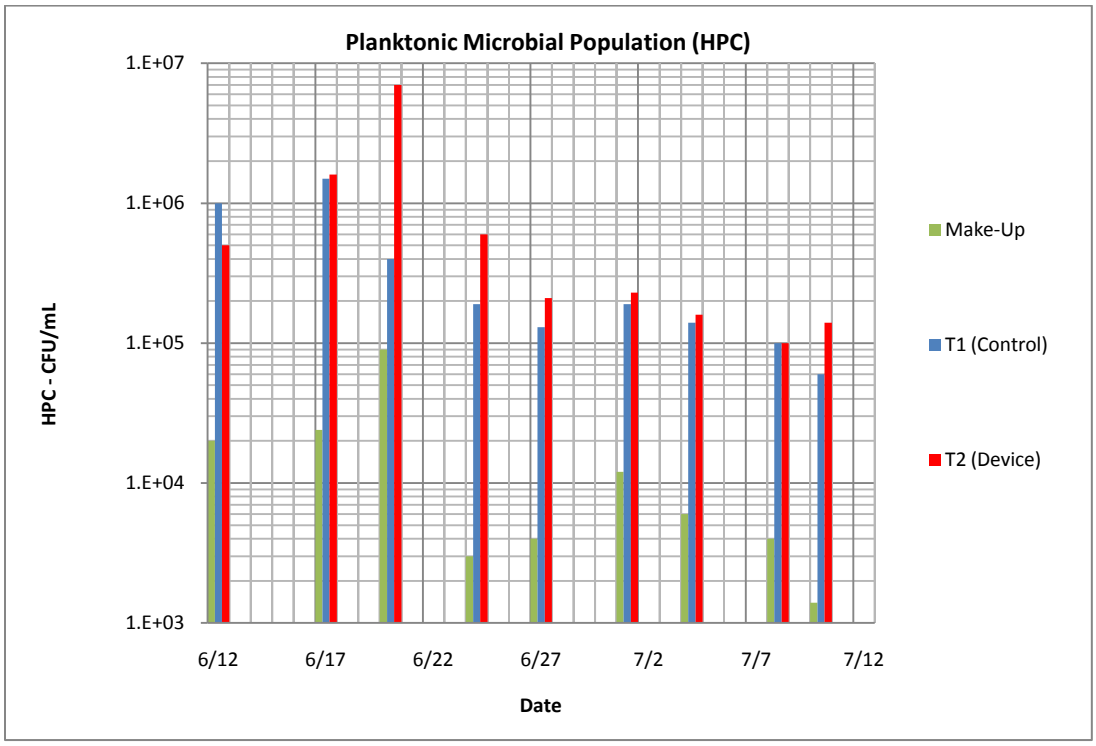


Figure 20 – **Planktonic microbial population enumerated by heterotrophic plate count for PEFD (Trial 2/2)**

Table 15 – Planktonic HPC statistical analysis for PEFD Trial 1/2 and PEFD Trial 2/2

(Planktonic heterotrophic plate counts were transformed to Log 10 data and compared using a paired t-test. Log 10 transformed data was normally distributed, Wilks-Shapiro test for normality)

Combined Runs (p = 0.92)

```
------------------------------------------------------------------------------
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
t1sump10 |      18    5.592301    .1256143    .5329365    5.327278    5.857324
t2sump10 |      18    5.602987    .1161657    .4928494    5.357899    5.848075
---------+--------------------------------------------------------------------
    diff |      18   -.0106858    .1051676    .4461884   -.2325701    .2111985
------------------------------------------------------------------------------
     mean(diff) = mean(t1sump10 - t2sump10)                      t =  -0.1016
 Ho: mean(diff) = 0                               degrees of freedom =       17

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.4601         Pr(|T| > |t|) = 0.9203          Pr(T > t) = 0.5399
```

The results of this t-test indicate that there was no significant difference between planktonic heterotrophic plate counts recorded for each of the tower systems during both device trials.

The concentration of microorganisms present in the make-up water during this investigation was comparable to that observed during the first pulsed electric field device trial. This is a result of the combination of make-up water dechlorination and storage. Although high microbial concentrations in the make-up water most likely led to constant seeding of microorganisms, no difference in planktonic heterotrophic plate counts between T1 (Control) and T2 (Device) was observed, indicating that the non-chemical device did not have an effect on the growth of planktonic microorganisms in the cooling tower.

The microbial population in each of the two tower systems was also enumerated using the measurement of cellular ATP. The concentration of ATP was used to calculate microbial equivalents according to the equation described previously in this report. ATP microbial equivalent measurements are shown in

Figure 21. The observed microbial population was higher in T2 (Device) than in T1 (Control) on 6 of the 8 sampling days. A statistical analysis using ATP data from both PEFD Trial 1/2 and Trial 2/2 is shown in **Table 16**. This analysis shows that the observed difference between the microbial populations of T1 (Control) and T2 (Device) during PEFD Trial 1/2 and Trial 2/2 was not statistically significant.

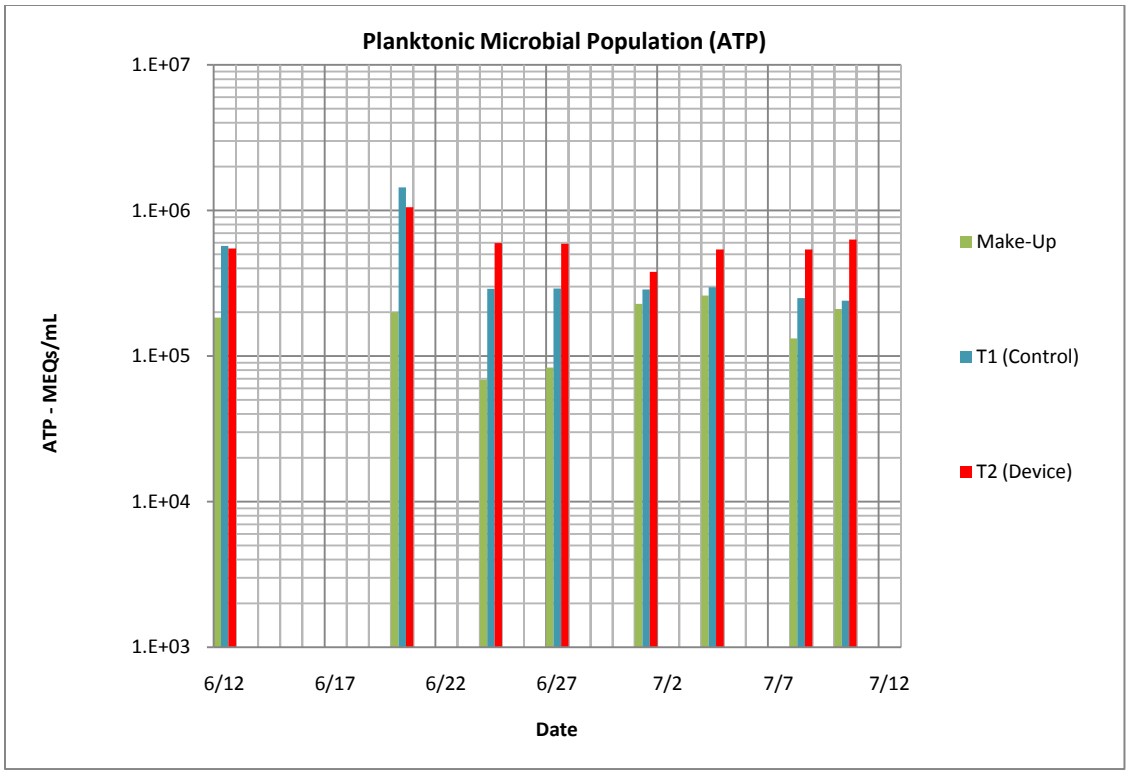


Figure 21 – Planktonic microbial population (ATP measurement) for PEFD (Trial 2/2)

Table 16 – Planktonic ATP statistical analysis for PEFD (Trial 2/2)

(Planktonic ATP microbial equivalent levels were transformed to Log 10 data and compared using a paired t-test. Log10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.041

Variable	Obs	Mean	Std. Err.	Std. Dev.	[95% Conf.	Interval]
atpt110	17	5.755866	.0761582	.3140084	5.594418	5.917315
atpt210	17	5.859539	.0463398	.1910637	5.761303	5.957775
diff	17	-.1036723	.0467444	.1927319	-.2027659	-.0045787

mean(diff) = mean(atpt110 - atpt210) t = -2.2179
Ho: mean(diff) = 0 degrees of freedom = 16

Ha: mean(diff) < 0 Pr(T < t) = 0.0207
Ha: mean(diff) != 0 Pr(|T| > |t|) = 0.0414
Ha: mean(diff) > 0 Pr(T > t) = 0.9793

The results of this t-test indicate that there was no significant difference between planktonic ATP microbial equivalent levels recorded for each of the tower systems during both device trials.

Sessile heterotrophic plate counts for each of the two tower systems over the course of the device trial are shown in **Figure 22**. The sessile microbial populations in the two tower systems were comparable and each tower system maintained a sessile microbial population in excess of 10^6 CFU/cm^2 throughout the trial. A statistical analysis was conducted using the sessile heterotrophic plate counts from both device trials, and the results are shown in **Table 17**. This analysis indicates that the observed difference between the sessile microbial populations in T1 (Control) and T2 (Device) during PEFD Trial 1/2 and Trial 2/2 was not statistically significant.

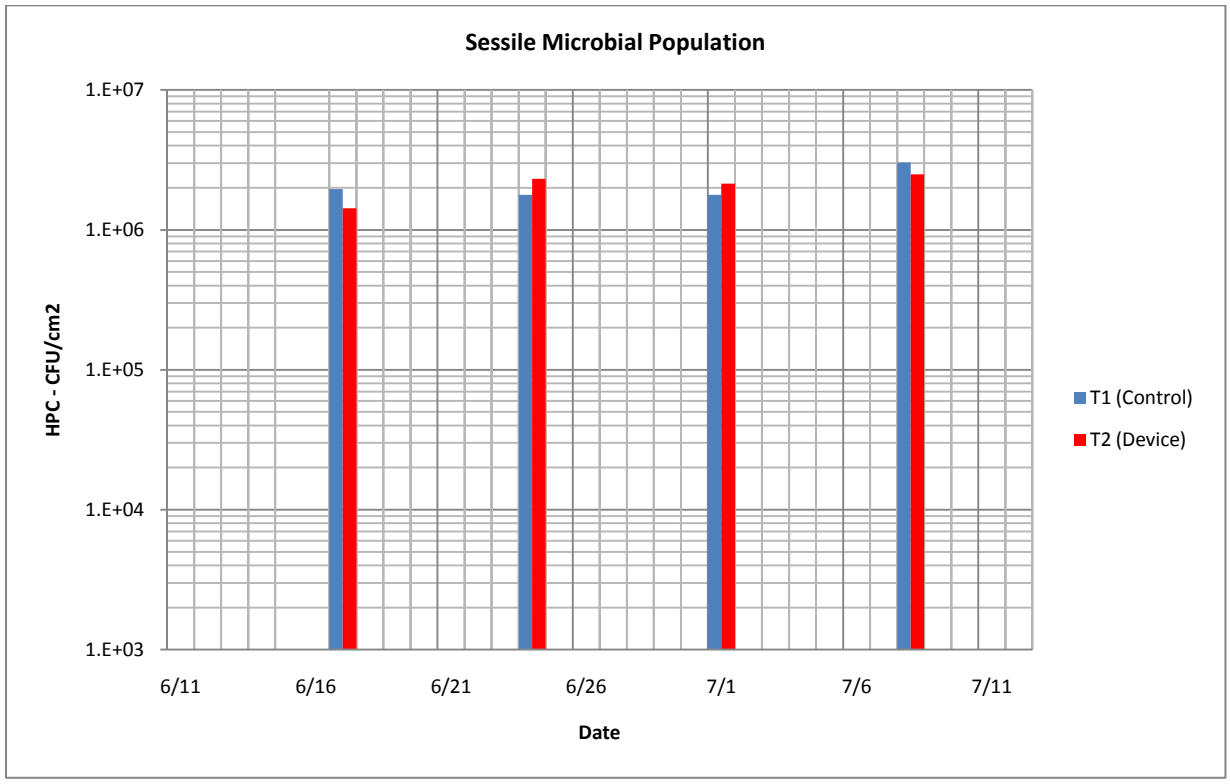


Figure 22 – Sessile microbial population for PEFD (Trial 2/2)

Table 17 – Sessile HPC statistical analysis for PEFD Trial 1/2 and Trial 2/2

(Sessile heterotrophic plate counts were transformed to Log 10 data and compared using a paired t-test. Log 10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.126

```
------------------------------------------------------------------------------
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
 t1bio10 |       9    6.264815    .1293026    .3879078    5.966643    6.562988
 t2bio10 |       9    6.170275    .1344017    .4032052    5.860344    6.480206
---------+--------------------------------------------------------------------
    diff |       9    .0945397    .0552943     .165883   -.0329692    .2220487
------------------------------------------------------------------------------
     mean(diff) = mean(t1bio10 - t2bio10)                      t =   1.7098
 Ho: mean(diff) = 0                     degrees of freedom =        8

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.9372         Pr(|T| > |t|) = 0.1257          Pr(T > t) = 0.0628
```

The results of this t-test indicate that there was no significant difference between sessile heterotrophic plate counts recorded for each of the tower systems during both device trials.

The make-up water contained a heterotrophic biological population between 1,400 – 90,000 CFU/mL during this device trial, with an average value of approximately 18,000 CFU/mL. Make-up water biological growth was allowed to occur without any means of biocidal control since water was dechlorinated prior to usage. No artificial microbial seeding was performed on the make-up water during this or any of the device trials.

4.3.3 Summary

The results presented in this report demonstrate that the pulsed electric field non-chemical device did not significantly reduce biological activity compared to the "control" tower. Two device trials were performed at different cycles of concentration. Planktonic heterotrophic plate counts

and ATP measurements from T1 (Control) and T2 (Device) showed no significant difference at any point during either of the device trials. The same behavior was observed for sessile heterotrophic plate counts in two tower systems during both device trials.

4.4 ELECTROSTATIC DEVICE (ED) TRIAL

The evaluation of the static electric field device began on 7/18 and it continued until 8/15. A malfunction in the make-up feed solenoid valve on T2 (Device) occurred on 8/2. As a result, T2 (Device) was turned off for several hours while the valve was replaced. This malfunction also led to a dilution of the water in T2 (Device). All data collected during this trial is included in Appendix F. Operational and chemical data collected during this investigation are shown in Appendix F.1, and statistical analyses of the chemical data are shown in Appendix F.2. Photographs of the tower systems taken before and after the device trial are shown in Appendix F.3, and photographs of biofilm coupon taken during the investigation are shown in Appendix F.4. After the trial period for the static electric field device had ended, a chlorination test was performed on T2 (Device) and these results are discussed in Section 4.7 of this report.

4.4.1 Tower System Operation

Operational and chemical data collected during this device trial are shown in **Figure F1 – Figure E19**. Throughout the course of the investigation, a temperature differential 9.8 – 11.6 ºF was maintained for T1 (Control), while the temperature differential of 11 – 14 ºF was maintained for T2 (Device) (**Figure F1**). **Figure F2** demonstrates that make-up water consumption levels were comparable in Tower 1 (Control) and Tower 2 (Device) during this test. This is indicated by the similar slopes of the two data sets. **Figure F3** demonstrates that T1 (Control) and T2 (Device) maintained comparable cumulative blowdown levels throughout the course of the experiment. Temperature profiles for each tower are

shown in **Figure F4** (T1) and **Figure F5** (T2). Fluctuations in the temperatures of each tower were the result of changes in ambient temperature (**Figure F6**) and relative humidity (**Figure F7**).

Average values for the parameters measured during this device trial are shown in **Table 18**. Examination of the continuous conductivity data (**Figure F8**) shows evidence of a blowdown valve malfunction in the T2 (Device) system on 7/29, resulting in a sudden spike followed by a dramatic drop in conductivity. Make-up water was unable to enter the tower, and a large amount of water evaporated from the system, thereby increasing the conductivity of system water. Once the valve was replaced, make-up water diluted the system volume, decreasing the overall system conductivity. Continuous pH data collected during the test (**Figure F9**) reveal that the average pH of T1 (Control) was approximately 8.58, while the average pH of T2 (Device) was approximately 8.64. A malfunction in the make-up feed valve of T1 (Control) on 7/30 diluted the system water, decreasing the pH significantly. Make-up water conductivity and pH are shown in **Figure F11.**

Three scaling indices were calculated using the chemical parameters observed during this investigation, and these indices may be used to quantify the scaling potential in each tower. Both towers produced comparable values for each of the scaling indices analyzed. The Langelier Saturation Index for T1 (Control) was approximately 0.96, and for T2 (Device) it was approximately 1.04, indicating that the water in each tower system had moderate scaling potential (**Figure F17**). Equilibrium values of the Ryznar Stability Index (**Figure F18**) were approximately 6.66 and 6.56 for T1 (Control) and T2 (Device), respectively, indicating that the water has slight scale-dissolving potential. Puckorius Scaling Index values were approximately 7.35 for T1 (Control) and 7.40 for T2 (Device) at equilibrium, which indicates moderate scale-dissolving tendency (**Figure F19**). A

combined analysis of these three indices indicates that each tower system contained water with mild scaling potential.

Table 18 – Average parameter values for ED trial

	T1 (Control)		T2 (Device)	
	Average	**Standard Deviation**	**Average**	**Standard Deviation**
Temperature Entering Tower (°F)	100.3	1.6	100.950	2.1
Sump Temperature (°F)	89.3	1.5	88.3	1.9
Daily Make-up Water Consumption (gal)	116	27	102	36
Daily Blowdown (gal)	19	9	18	11
Conductivity (mS/cm)	1.217	0.072	1.184	0.109
pH	8.58	0.08	8.64	0.08
ORP (mV)	190	31	195	34
Alkalinity (mg/L as $CaCO_3$)	86	10	88	11
Calcium Hardness (mg/L as $CaCO_3$)	153	10	152	10
Magnesium Hardness (mg/L as $CaCO_3$)	105	8	99	9
Total Hardness (mg/L as $CaCO_3$)	257	15	251	18
TDS (mg/L)	793	93	773	87
LSI	0.96	0.15	1.04	0.43
RSI	6.66	0.22	6.56	0.72
PSI	7.49	0.24	7.51	0.20
Planktonic HPC (CFU/mL)	3.89E+05	2.79E+05	4.99E+05	3.66E+05
ATP Microbial Equivalents (MEQs/mL)	5.72E+05	1.33E+05	7.44E+05	2.43E+05
Sessile HPC (CFU/cm^2)	1.43E+06	6.05E+05	1.60E+06	5.49E+05

4.4.2 Biological Parameters

Observation of the biological population present in each tower (**Figure 23** – **Figure 25**) indicates that each tower contained comparable levels of microbial activity both in the bulk water and in system biofilms throughout the course of the experiment. No significant reduction in microbial activity was observed in the tower treated with the static electric field device.

A chlorination test was performed on 8/22 in tower T2 (Device), resulting in a significant reduction in planktonic and sessile microbial activity observed in this tower on 8/23. The results of this chlorination test will be discussed together with other chlorination tests conducted throughout this study in Section 4.7 of this report.

Heterotrophic plate counts for each tower system over the course of the device trial are shown in **Figure 23**. The microbial population in T2 (Device) was higher than T1 (Control) on 6 of the 9 sampling days. Both towers maintained planktonic microbial populations in excess of acceptable industry standards (approximately 10^4 CFU/mL). A statistical analysis of the heterotrophic plate counts measured during the device trial is shown in **Table 19**. This analysis indicates that the observed difference between the heterotrophic plate counts in T1 (Control) and T2 (Device) was not statistically significant.

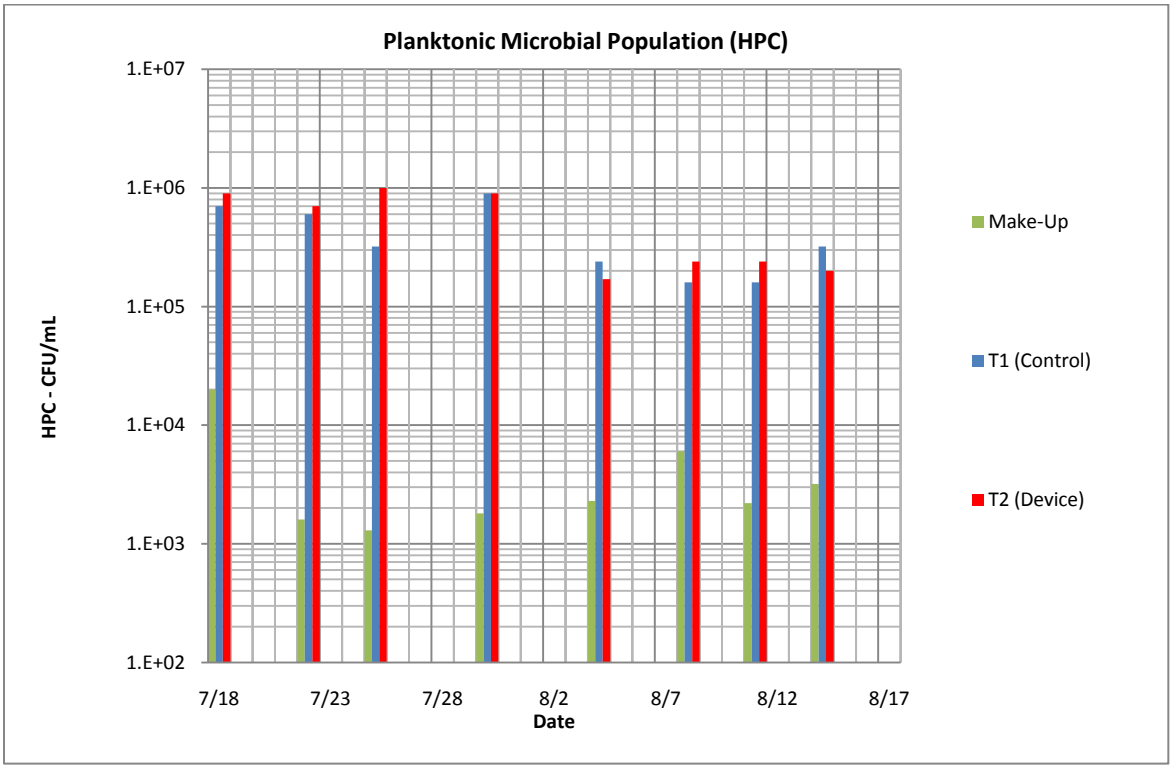


Figure 23 – Planktonic microbial population (HPC) for ED trial

Table 19 – Planktonic HPC statistical analysis for ED trial

(Planktonic heterotrophic plate counts were transformed to Log 10 data and compared using a paired t-test. Log 10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.31

```
------------------------------------------------------------------------------
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
t1sump10 |       8     5.54703    .1012947    .2865046    5.307507    5.786554
t2sump10 |       8    5.630686    .1184785    .3351079    5.350528    5.910843
---------+--------------------------------------------------------------------
    diff |       8   -.0836552    .0769271    .2175826   -.2655589    .0982484
------------------------------------------------------------------------------
     mean(diff) = mean(t1sump10 - t2sump10)                      t =  -1.0875
 Ho: mean(diff) = 0                               degrees of freedom =        7

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.1564         Pr(|T| > |t|) = 0.3129          Pr(T > t) = 0.8436
```

The results of this t-test indicate that there was no significant difference between planktonic heterotrophic plate counts recorded for each of the tower systems during both device trials

The microbial population in each tower was also enumerated by measuring the concentration of cellular ATP present in each tower system and converting this measurement to microbial equivalents using the equation described previously in this report. The microbial equivalents for each tower system during this device trial are shown in **Figure 24**. T2 (Device) maintained higher microbial population than T1 (Control) on 6 of the 8 sampling dates. A statistical analysis of this data is shown in **Table 20**. This analysis shows that the observed difference between the microbial equivalents in T1 (Control) and T2 (Device) was not statistically significant.

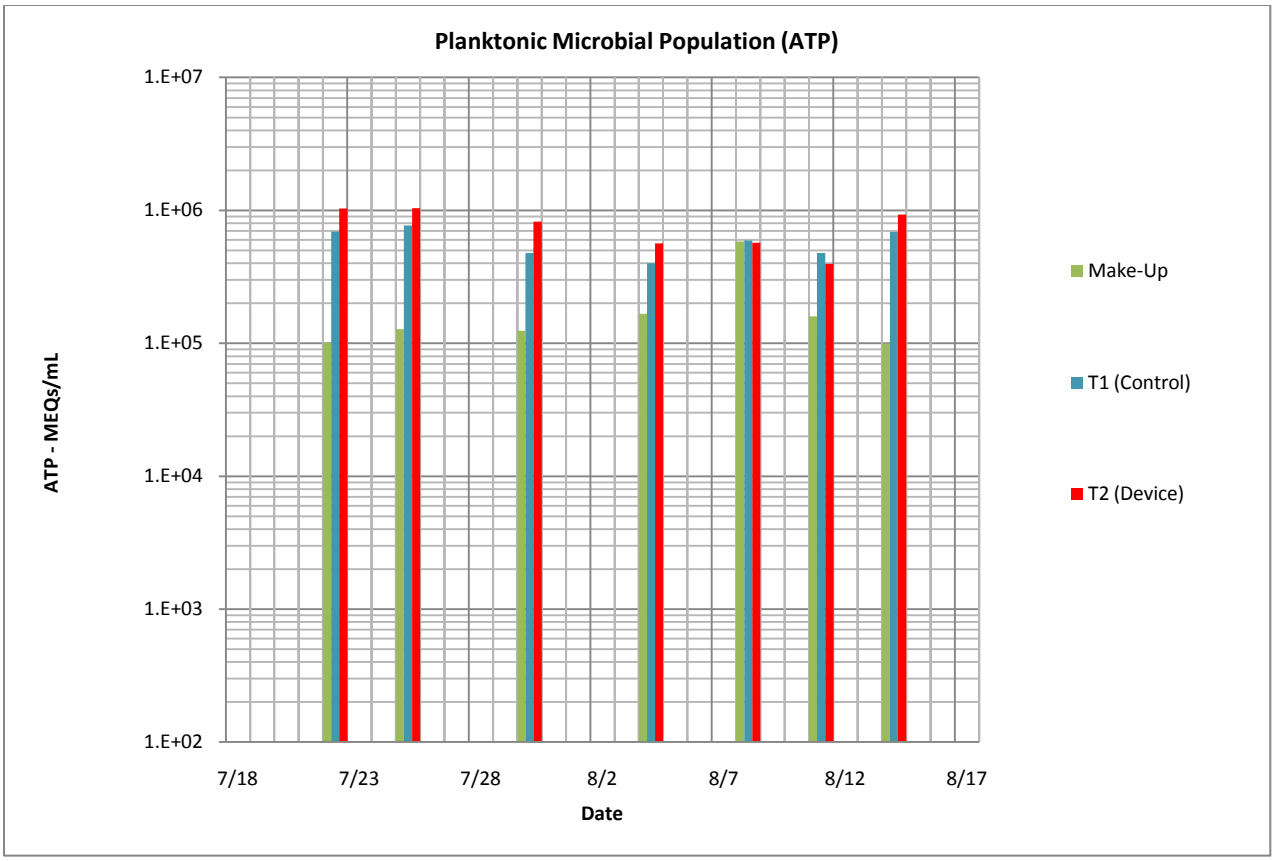


Figure 24 – Planktonic microbial population (ATP measurement) for ED trial

Table 20 – Planktonic ATP statistical analysis for ED trial

(Planktonic ATP microbial equivalent levels were transformed to Log 10 data and compared using a paired t-test. Log 10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.05

```
------------------------------------------------------------------------------
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
 t1atp10 |       7    5.756859    .0397657      .10521    5.659556    5.854162
 t2atp10 |       7     5.85979    .0605656    .1602416    5.711591    6.007989
---------+--------------------------------------------------------------------
    diff |       7   -.1029312    .0420996     .111385   -.2059451    .0000828
------------------------------------------------------------------------------
     mean(diff) = mean(t1atp10 - t2atp10)                          t =  -2.4449
 Ho: mean(diff) = 0                               degrees of freedom =        6

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.0251         Pr(|T| > |t|) = 0.0501          Pr(T > t) = 0.9749
```

The results of this t-test indicate that there was no significant difference between planktonic ATP microbial equivalent levels recorded for each of the tower systems during both device trials.

The sessile heterotrophic plate counts for each tower system throughout the course of the device trial are shown in **Figure 25**. The sessile microbial populations in each tower system were comparable for the duration of the trial. Each tower system maintained a sessile microbial population in excess of 10^5 CFU/cm^2. A statistical analysis of sessile heterotrophic plate counts shown in **Table 21** reveals that the observed difference between the sessile heterotrophic plate counts for T1 (Control) and T2 (Device) was not statistically significant.

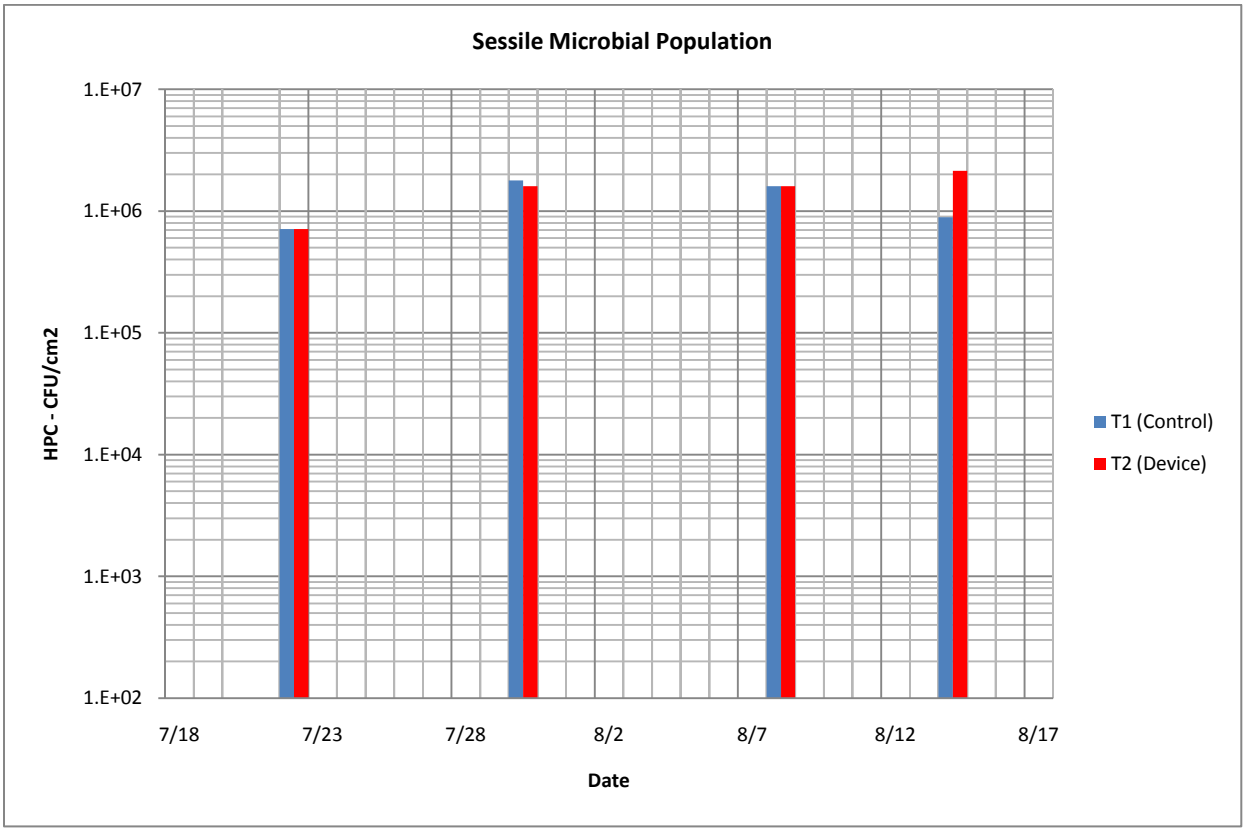


Figure 25 – Sessile microbial population for ED trial

Table 21 – Sessile HPC statistical analysis for ED trial

(Sessile heterotrophic plate counts were transformed to Log 10 data and compared using a paired t-test. Log 10 transformed data was normally distributed, Wilks-Shapiro test for normality)

p = 0.46

```
------------------------------------------------------------------------------
Variable |     Obs        Mean    Std. Err.   Std. Dev.   [95% Conf. Interval]
---------+--------------------------------------------------------------------
 t1bio10 |       4    6.064831    .0968112    .1936224    5.756734    6.372927
 t2bio10 |       4    6.148427    .1027606    .2055213    5.821397    6.475457
---------+--------------------------------------------------------------------
    diff |       4   -.0835965    .0995033    .1990065   -.4002603    .2330673
------------------------------------------------------------------------------
     mean(diff) = mean(t1bio10 - t2bio10)                      t =  -0.8401
 Ho: mean(diff) = 0                         degrees of freedom =        3

 Ha: mean(diff) < 0           Ha: mean(diff) != 0           Ha: mean(diff) > 0
 Pr(T < t) = 0.2313         Pr(|T| > |t|) = 0.4625          Pr(T > t) = 0.7687
```

The results of this t-test indicate that there was no significant difference between sessile heterotrophic plate counts recorded for each of the tower systems during both device trials.

The make-up water contained a heterotrophic biological population between 1,300 – 20,000 CFU/mL during this device trial, with an average value of approximately 4,400 CFU/mL. Make-up water biological growth was allowed to occur without any means of biocidal control since water was dechlorinated prior to usage. No artificial microbial seeding was performed on the make-up water during this or any of the device trials.

Phase I consisted of operating both the control and device towers for 4 weeks, with the non-chemical treatment device activated at the beginning of the investigation. This phase was completed for the static electric field treatment device. Since no microbial control was demonstrated during Phase I, Phase II was not initiated for this device. Phase II was to be conducted after a 2-week period during which each tower was allowed to operate with no treatment to achieve significant biological activity. The device would then be activated to determine whether or not any existing biological

growth in the device tower may be removed through the application of this non-chemical treatment device.

4.4.3 Summary

The results presented in this report demonstrate that the static electric field non-chemical device did not significantly reduce biological activity compared to the "control" tower. Planktonic heterotrophic plate counts and ATP measurements from T1 (Control) and T2 (Device) showed no significant difference at any point during the investigation. The same trend was observed for sessile heterotrophic plate counts in the two tower systems.

4.5 ULTRASONIC DEVICE (UD) TRIAL

The investigation of the ultrasonic non-chemical treatment device began on 9/2 and continued uninterrupted until 9/30. Data collected during this investigation is shown in Appendix G. Operational and chemical data from this investigation are included in Appendix G.1, and statistical analyses of the chemical parameters are included in Appendix G.2. Appendix G.3 contains photographs of each tower system before and after the device trial. Appendix G.4 contains photographs of biofilm sampling coupons taken from each tower system throughout the course of the investigation.

4.5.1 Tower System Operation

Tower operational and chemical data is shown in **Figure G1 – Figure G19**. Average values for the parameters measured during this device trial are contained in **Table 22**. The temperature differential for each tower system over the course of the experiment is shown in **Figure G1**. Calculation of the temperature differential was performed by subtracting the manual sump temperature reading from the manual reading of the temperature entering each tower system. The tower systems were designed with a target temperature differential of approximately 10 °F. **Figure G1** indicates that T1 (Control) operated with a temperature differential of 9.0 – 11.3 ºF, while T2 (Device) operated with a temperature differential of 10.2 – 13.2 ºF. The difference in temperature differentials between the two tower systems was due to differing performance efficiencies of the heating elements and axial cooling fans installed in each system.

Table 22 – Average parameter values for UD trial

	T1 (Control)		T2 (Device)	
	Average	Standard Deviation	Average	Standard Deviation
Temperature Entering Tower (°F)	100.6	2.4	102.5	3.0
Sump Temperature (°F)	90.1	2.8	90.3	3.3
Daily Make-up Water Consumption (gal)	123	27	121	27
Daily Blowdown (gal)	19	9	25	12
Conductivity (mS/cm)	1.334	0.149	1.335	0.167
pH	8.71	0.06	8.78	0.05
ORP (mV)	197	33	185	27
Alkalinity (mg/L as $CaCO_3$)	102	14	98	11
Calcium Hardness (mg/L as $CaCO_3$)	175	15	176	17
Magnesium Hardness (mg/L as $CaCO_3$)	124	21	126	22
Total Hardness (mg/L as $CaCO_3$)	299	36	302	39
TDS (mg/L)	915	115	914	128
LSI	1.22	0.13	1.30	0.11
RSI	6.27	0.21	6.19	0.17
PSI	7.49	0.24	7.51	0.20
Planktonic HPC (CFU/mL)	3.81E+05	4.06E+05	2.04E+05	1.98E+05
ATP Microbial Equivalents (MEQs/mL)	9.00E+05	7.71E+05	5.52E+05	7.33E+05
Sessile HPC (CFU/cm^2)	7.74E+06	7.69E+06	1.43E+07	2.27E+07

Both towers were supplied with make-up water from the same source. Make-up water consumption rates for each tower system (**Figure G2)** were nearly identical throughout the test, as indicated by the similar slopes produced by the cumulative consumption curves. Cumulative blowdown volumes for each tower system (**Figure G3**) indicate that T2 (Device) had more frequent blowdowns than T1 (Control). This is most likely because temperatures in T1 (Control) (**Figure G4**) were lower than those in T2 (Device) (**Figure G5**), resulting in more evaporation and more frequent blowdowns in T2 (Device) due to increased conductivity. Fluctuations in tower temperatures were the result of changes in ambient temperature (**Figure G6**) and relative humidity (**Figure G7**).

Chemical data collected during this investigation are shown in Appendix G (**Figure G8** – **Figure G16**). The continuous conductivity data (**Figure G8**) shows two significant increases in the tower system conductivities. On 9/14, the blowdown conductivity setpoint was changed from 1200 µS/cm to 1400 µS/cm, increasing peak conductivity levels in both tower systems. On 9/22, the blowdown conductivity setpoint was raised from 1400 µS/cm to 1500 µS/cm, again increasing peak conductivity levels for each system. Changes in the blowdown conductivity setpoint were made based on the increasing conductivity of the make-up water used in this trial (**Figure G11**). The blowdown conductivity setpoint was adjusted to be approximately 4 times the conductivity of the incoming make-up water in order to establish 4-5 cycles of concentration. Continuous pH data for the tower system are shown in **Figure G10**, indicating that the average pH in T1 (Control) at equilibrium was approximately 8.71, while the average equilibrium pH for T2 (Device) was approximately 8.78. Inflows of make-up water were associated with steep drops in tower pH. Following the periods of make-up water feeding, the pH steadily increased as system water evaporated.

Chloride concentration measurements collected in this investigation (**Table G1**) indicate that each system was operating at 5-6 cycles of concentration throughout the course of the testing. This is slightly higher than the target of 4-5 cycles of concentration. Conductivity was used to establish the cycles of concentration in each tower. The chloride ion is a more sensitive indicator of cycles of concentration due to its relative inertness, and as a result the cycles of concentration determined using chloride measurements tend to be higher than those determined using conductivity.

Values for three scaling indices were calculated using the chemical data collected during this investigation. The Langelier Saturation Index (**Figure G17**) was approximately 1.22 for T1 (Control) and 1.30 for T2 (Device), indicating strong scaling potential. The equilibrium Ryznar Stability Index

(**Figure G18**) for T1 (Control) was 6.27, while for T2 (Device) it was approximately 6.19, indicating little to no scaling potential in either of the tower systems. The Puckorius Scaling Index (**Figure G19**) was approximately 7.49 for T1 (Control) and 7.51 for T2 (Device), indicating moderate scale-dissolving potential.

4.5.2 Biological Parameters

Biological data collected during this trial with ultrasonic device are shown in **Figure 26** – **Figure 28**. The planktonic and sessile microbial populations were comparable in each of the two towers for the duration of this trial. There was no observed reduction in microbial activity in T2 (Device). A chlorination test was performed beginning on 10/2. The last data point (10/4) is excluded from the discussion of the data collected in this trial and the results of this chlorination test will be discussed together with other chlorination tests conducted throughout this study in Section 4.7 of this report.

The planktonic heterotrophic plate counts for each tower system during this device trial are shown in **Figure 26**. Each tower system had heterotrophic plate counts higher than the industrial standard of 10^4 CFU/mL. T2 (Device) maintained a lower microbial population on 6 of the 9 sampling dates. If the non-chemical device reduced the average tower microbial concentrations, it should have been reflected in a significant difference between microbial counts for T1 (Control) and T2 (Device). However, no statistically significant difference between the microbial counts was observed in the two towers. A paired t-test was used to compare log 10 planktonic heterotrophic plate

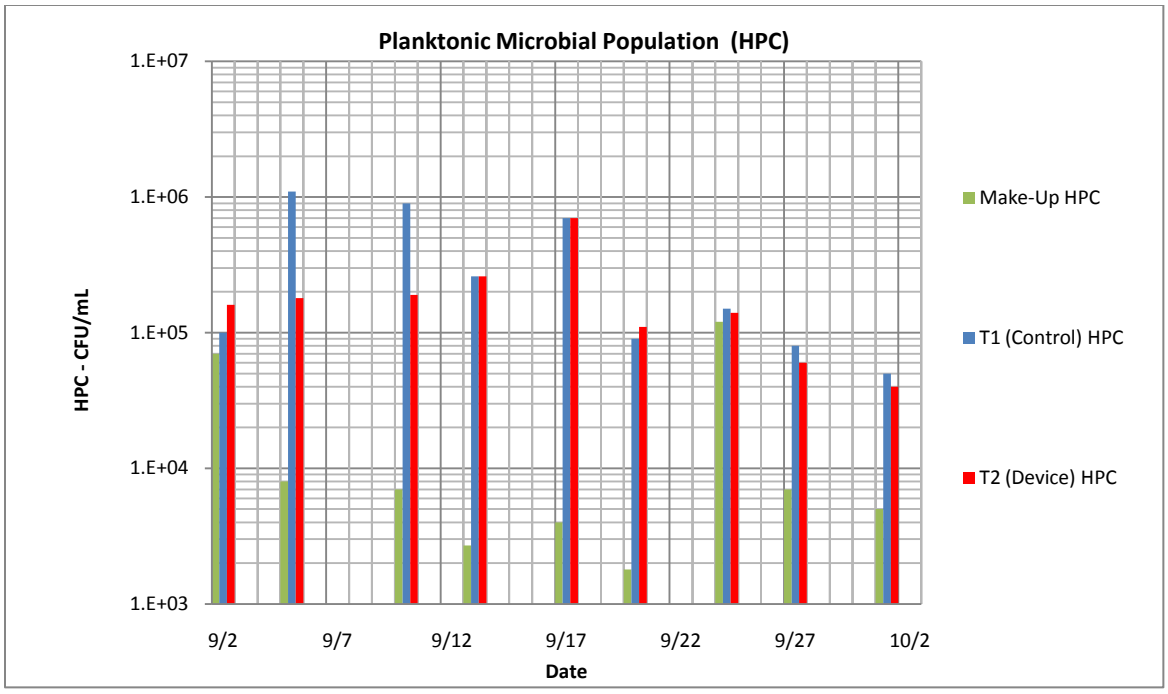


Figure 26 – Planktonic microbial population (HPC) for UD trial

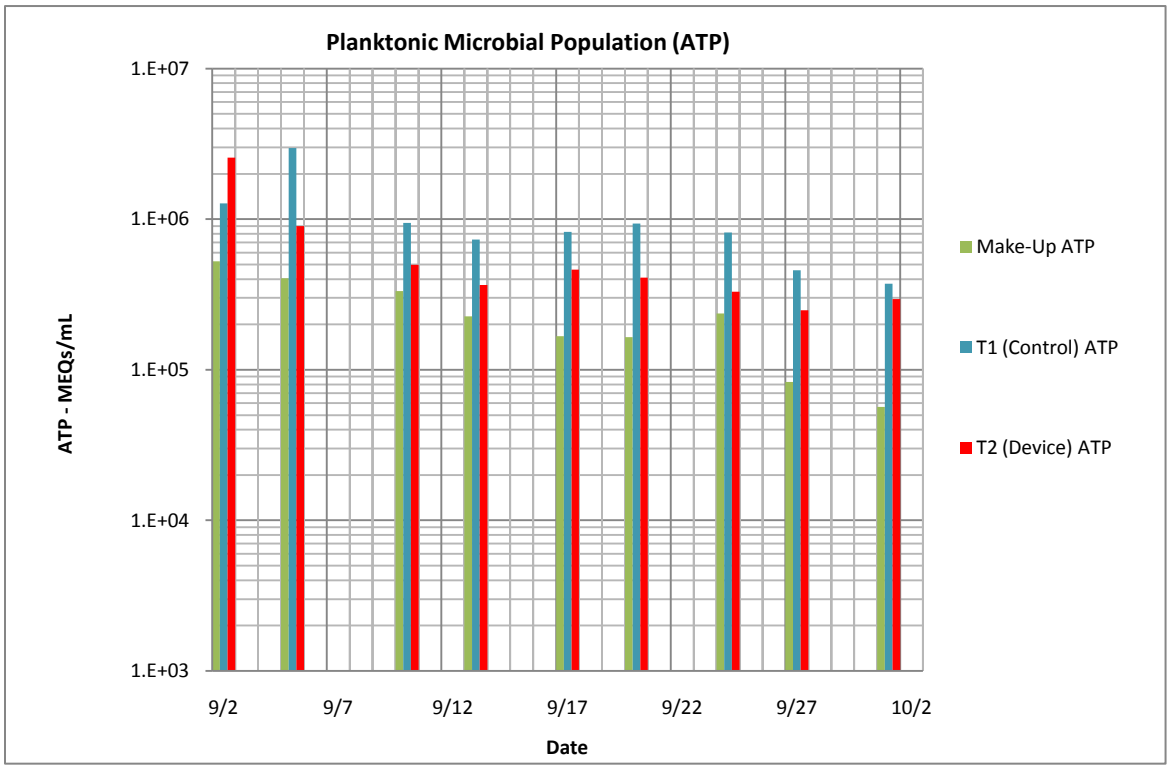


Figure 27 – Planktonic microbial population (ATP measurement) for UD trial

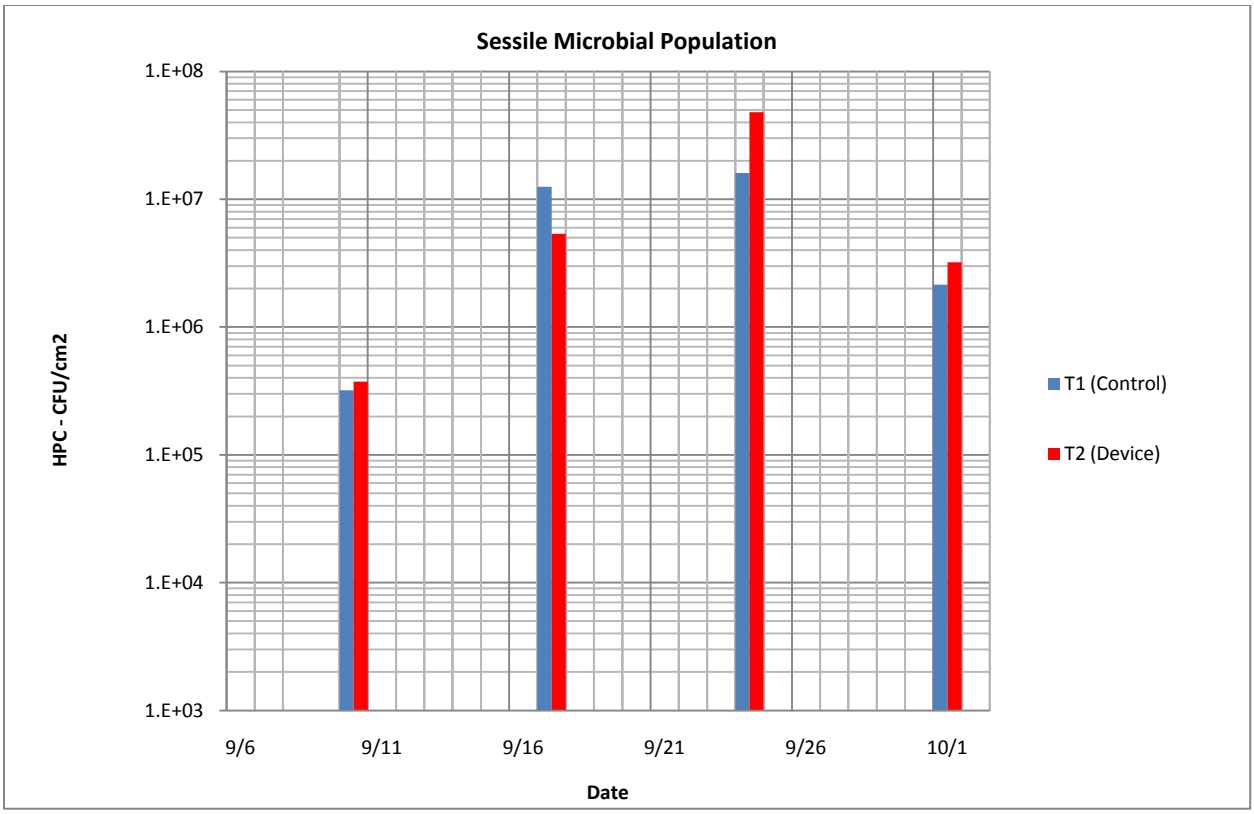


Figure 28 – Sessile microbial population for UD trial

counts for each tower system, resulting in a p value of 0.449. This indicates that the observed difference between the planktonic heterotrophic plate counts in T1 (Control) and T2 (Device) was not statistically significant.

Planktonic microbial populations in each tower were also enumerated by converting measured cellular ATP concentrations to microbial equivalents using the equation previously discussed in this report. The planktonic microbial equivalent values determined in this fashion are shown in **Figure 27**. While T2 (Device) maintained a lower microbial population throughout the device trial, the difference between T1 (Control) and T2 (Device) was less than 1 log value. A paired t-test was performed using the log 10 microbial equivalent data from this device trial, resulting in a p-value of $p = 0.063$. This

indicates that the observed difference between the microbial populations in T1 (Control) and T2 (Device) was not statistically significant.

Sessile heterotrophic plate counts from T1 (Control) and T2 (Device) were also comparable throughout the course of this investigation (**Figure 28**). A paired t-test was performed using the log 10 sessile heterotrophic plate counts, resulting in a p-value of $p = 0.469$. This indicates that the observed difference between the sessile heterotrophic plate counts in T1 (Control) and T2 (Device) was not statistically significant.

The make-up water contained a heterotrophic biological population between 1,800 – 120,000 CFU/mL during this device trial, with an average value of approximately 23,000 CFU/mL. Make-up water biological growth was allowed to occur without any means of biocidal control since water was dechlorinated prior to usage. No artificial microbial seeding was performed on the make-up water during this or any of the device trials.

Since no sessile or planktonic microbial control was demonstrated during Phase I, the device investigation was terminated without performing the Phase II testing. Phase I consisted of operating both the control and device towers for 4 weeks, with the non-chemical treatment device activated at the beginning of the investigation. This phase was completed for the ultrasonic treatment device. Phase II was to be conducted after a 2 week period during which each tower was allowed to operate with no treatment. The device would then be activated to determine whether or not any existing biological growth in the device tower may be removed through the application of non-chemical treatment.

4.5.3 Summary

The results presented in this report demonstrate that the ultrasonic non-chemical device did not significantly reduce biological activity compared to the "control" tower. Planktonic heterotrophic plate counts and ATP measurements from T1 (Control) and T2 (Device) showed no statistically significant difference at any point during the investigation. The same trend was observed for sessile heterotrophic plate counts from each tower system.

4.6 HYDRODYNAMIC CAVITATION DEVICE (HCD) TRIAL

The evaluation of the hydrodynamic cavitation treatment device began on 10/27, and it continued uninterrupted until 11/24. All data collected during this investigation is included in Appendix G. Operational and chemical data figures and tables for this investigation are shown in Appendix G.1, and statistical analyses of the chemical data are included in Appendix G.2. Photographs of each of the two tower systems before and after the device trial are included in Appendix G.3, and photographs of biofilm sampling coupons are provided in Appendix G.4.

4.6.1 Tower System Operation

Tower operational and chemical data are shown in **Figure G1 - Figure G19**. After three days of operation, the temperature differential for each tower system was 10 – 11.6 ºF (**Figure G1**), which was near the target differential of 10 ºF. T2 (Device) consumed 443 gal. more make-up water than T1 (Control) over the course of the experiment (**Figure G2**), but cumulative blowdown volumes for the two towers were comparable (**Figure G3**). T2 (Device) most likely consumed more make-up water than T1 (Control) due to increased splashing within the system holding tank caused by the discharge from the non-chemical device recirculation loop.

Tower temperature profiles are shown in **Figure G4** (T1) and **Figure G5** (T2). The heater installed in the T2 (Device) tower system was not operating at full capacity from 10/31 to 11/19. As a result, average tower temperatures were lower in T2 (Device) than in T1 (Control) during this period. Over the course of the experiment, the average temperature entering T1 (Control) was 99.8 °F, while the average temperature entering T2 (Device) was 95.5 °F. The

difference in temperature between the two towers was not severe enough to have a significant impact on microbial growth rates.

Average values and standard deviations for all parameters measured during this investigation are shown in **Table 23**. These values were calculated using the data collected after the first blowdown occurred in each of the towers. Chemical data collected during this investigation are shown in **Figure G8 - Figure G16**.

Table 23 – Average values and standard deviations for HCD trial

	T1 (Control)		T2 (Device)	
	Average	Standard Deviation	Average	Standard Deviation
Temperature Entering Tower (°F)	99.8	1.1	95.5	3.2
Sump Temperature (°F)	89.1	1.3	84.9	3.3
Daily Make-up Water Consumption (gal)	124	27	139	32
Daily Blowdown	23	10	23	10
Conductivity (mS/cm)	1.364	0.091	1.385	0.080
pH	8.75	0.04	8.67	0.05
ORP (mV)	180	22	181	23
Alkalinity (mg/L as $CaCO_3$)	115	8	115	6
Calcium Hardness (mg/L as $CaCO_3$)	201	10	191	14
Magnesium Hardness (mg/L as $CaCO_3$)	102	11	99	10
Total Hardness (mg/L as $CaCO_3$)	303	16	290	21
TDS (mg/L)	929	66	920	51
LSI	1.36	0.07	1.21	0.10
RSI	6.02	0.10	6.24	0.15
PSI	7.21	0.11	7.35	0.14
Planktonic HPC (CFU/mL)	9.56E+04	4.50E+04	1.24E+05	9.03E+04
ATP Microbial Equivalents (MEQs/mL)	3.81E+05	1.12E+05	5.65E+05	3.03E+05
Sessile HPC (CFU/cm^2)	6.37E+05	3.37E+05	2.10E+06	9.48E+05

Three scaling indices were calculated using the chemical parameters measured during this device trial. The Langelier Saturation Index (**Figure G17**) was approximately 1.36 for T1 (Control) at equilibrium, and the equilibrium value for T2 (Device) was approximately 1.21. These values indicate that each system had mild scaling potential. The Ryznar Stability Index (**Figure G18**) was 6.02 for T1 (Control) and 6.24 for T2 (Device) at equilibrium, while the equilibrium values of the Puckorius Scaling Index (**Figure G19**) for T1 (Control) and T2 (Device) were 7.21 and 7.35, respectively. All of these scaling index values indicate mild scaling potential in both T1 (Control) and T2 (Device).

4.6.2 Biological Parameters

Biological data collected during this investigation are included in **Figure 29** – **Figure 31**. Based on these data, there was no observed biocidal effect as a result of the device installation. T1 (Control) and T2 (Device) demonstrated comparable sessile and planktonic microbial growth throughout the device trial.

Planktonic heterotrophic plate counts for each tower system over the course of the device trial are shown in **Figure 29**. Neither T1 (Control) nor T2 (Device) maintained planktonic microbial levels below the industry standard (10^4 CFU/mL). The average planktonic heterotrophic plate count for T1 (Control) was 9.56×10^4 CFU/mL with a standard deviation of 4.50×10^4 CFU/mL, while the average value for T2 (Device) was 1.24×10^5 CFU/mL with a standard deviation of 9.03×10^4 CFU/mL. A paired t-test was performed using the log 10 heterotrophic plate count data from this device trial, resulting in a p-value of 0.513. This

indicates that the observed difference between the planktonic microbial populations of T1 (Control) and T2 (Device) was not statistically significant.

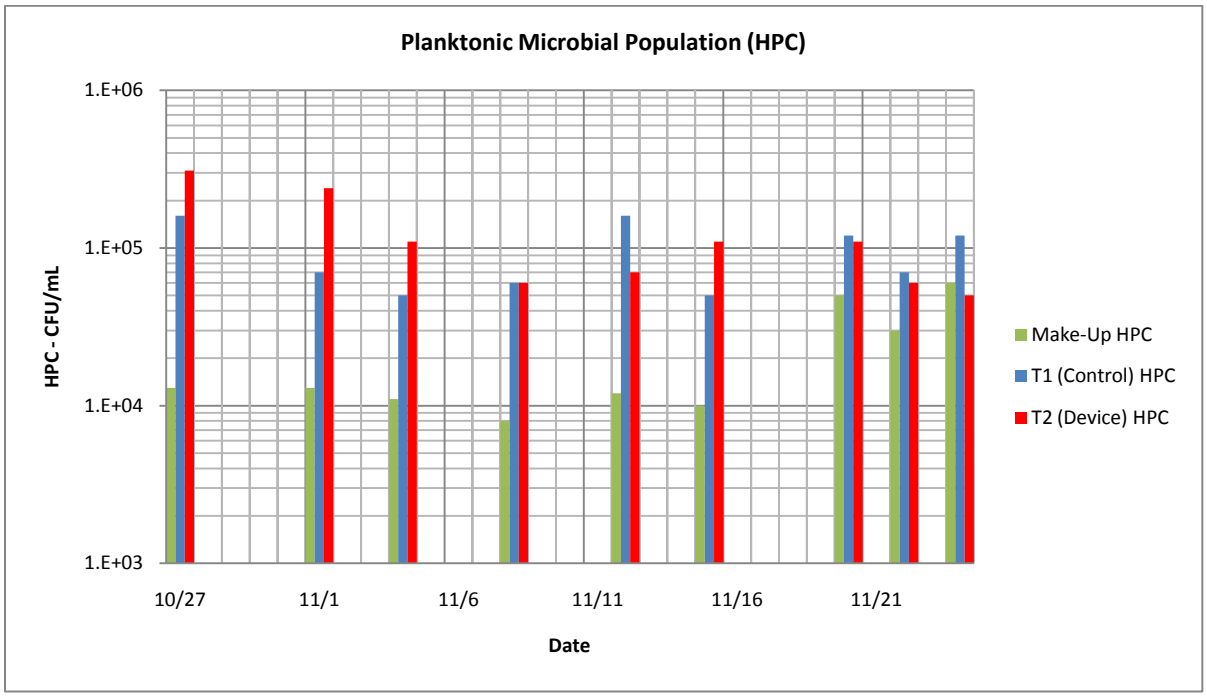


Figure 29 – Planktonic microbial population enumerated by heterotrophic plate count for HCD trial

The planktonic microbial population in each tower system was also enumerated by converting cellular ATP measurements to microbial equivalents using the equation described previously in this report. The planktonic microbial equivalents for each tower system during the course of this device trial are shown in **Figure 30**. The microbial population was higher in T2 (Device) than T1 (Control) on 7 of the 9 sampling dates. A paired t-test was performed using the log 10 microbial equivalent data, resulting in a p-value of $p = 0.058$. This indicates that the observed difference between the microbial populations observed in T1 (Control) and T2 (Device) was not statistically significant.

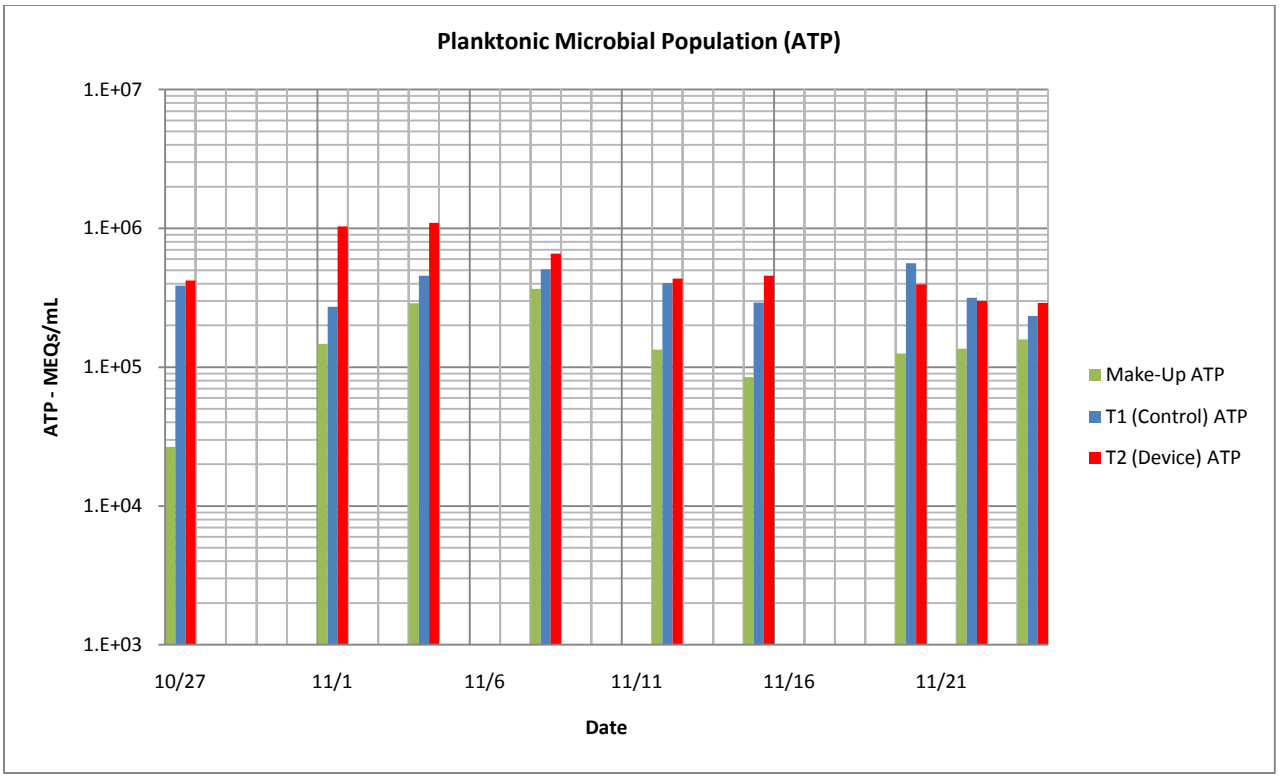


Figure 30 – Planktonic microbial population enumerated by ATP measurement for HCD trial

The average sessile heterotrophic plate count (**Figure 31**) for T1 (Control) was 6.37 x 10^5 CFU/cm^2 with a standard deviation of 3.37 x 10^5 CFU/cm^2, while for T2 (Device) the average was 2.10 x 10^6 CFU/cm^2 with a standard deviation of 9.48 x 10^5 CFU/cm^2. T2 (Device) maintained a higher sessile microbial population than T1 (Control) on 3 of the 4 sampling dates. A paired t-test was performed using the log 10 sessile heterotrophic plate count data from this device trial, resulting in a p-value of 0.058. This indicates that the observed difference between the sessile microbial populations in T1 (Control) and T2 (Device) was not statistically significant. Since no significant difference in the sessile and planktonic microbial populations was observed between T1 (Control) and T2 (Device), the investigation was terminated following the completion of Phase I. Phase II was not completed for this treatment device.

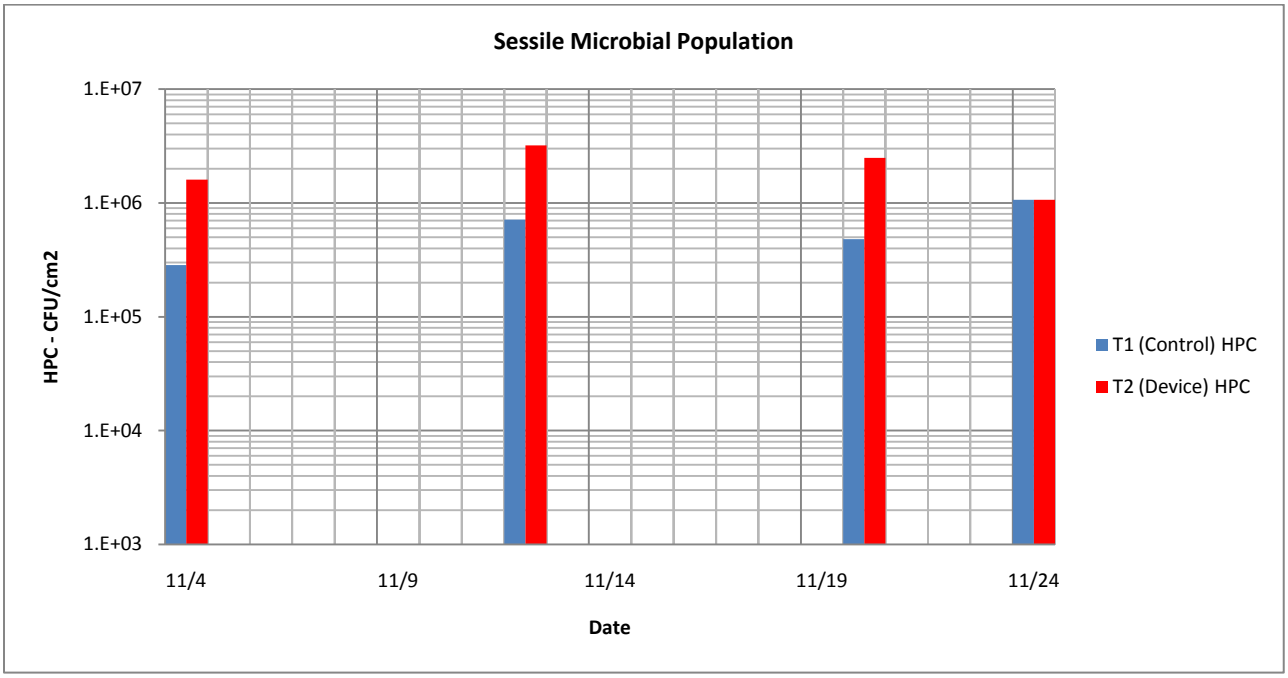


Figure 31 – Sessile microbial population for HCD trial

The make-up water contained a heterotrophic biological population between 8,000 – 60,000 CFU/mL during this device trial, with an average value of approximately 23,000 CFU/mL. Make-up water biological growth was allowed to occur without any means of biocidal control since water was dechlorinated prior to usage. No artificial microbial seeding was performed on the make-up water during this or any of the device trials.

4.6.3 Summary

The results presented in this report demonstrate that the hydrodynamic cavitation non-chemical device did not reduce planktonic or sessile microbial populations compared to the "control" tower. Heterotrophic plate counts for both planktonic and sessile microbial populations were comparable for T1 (Control) and T2 (Device). In addition, ATP measurements showed no significant microbial reduction in the device tower system when compared to the control tower system.

4.7 CHLORINATION TEST RESULTS

Over the course of the investigation, 3 separate chlorination tests were performed to demonstrate effective chemical microbial control. The first chlorination test was performed from 1/15/09 – 1/26/09. The second chlorination was performed immediately following the ED trial (8/14/09 – 8/23/09), and the third was performed immediately following the UD trial (9/27/09 – 10/4/09).

The selection of free chlorine as a positive control was based on common practice in cooling water treatment and a previous study where several chemical biocides, including free chlorine, were evaluated in model cooling towers that simulated real-world cooling tower operational conditions (Thomas et al., 1999). A detailed description of both the results of the Thomas et al. study and the protocol used for each chlorination test during this investigation is included in Section 2.2.3 of this report.

4.7.1 Pre-Device Trial Chlorination Test

The first chlorination test was performed prior to the beginning of the device trials. During this test, both T1 (Control) and T2 (Device) operated untreated from 1/15 – 1/22. After samples were taken on 1/22, a spike dose of chlorine was added to each of the towers. Following this spike dose, chlorine stock solution was pumped into each tower system to maintain a chlorine concentration of approximately 1 mg/L. The planktonic microbial population enumerated by heterotrophic plate count is shown in **Figure 32**, while ATP enumeration is shown in **Figure 33**. Each tower demonstrated a 2-3 log reduction in planktonic microbial activity within 3 days from the beginning of chlorination. The sessile microbial population is shown in **Figure 34**.

Chlorination produced a 4-5 log reduction in sessile microbial activity in each of the tower systems. Make-up water heterotrophic plate counts observed during this chlorination test were comparable to those observed during the device trials which followed.

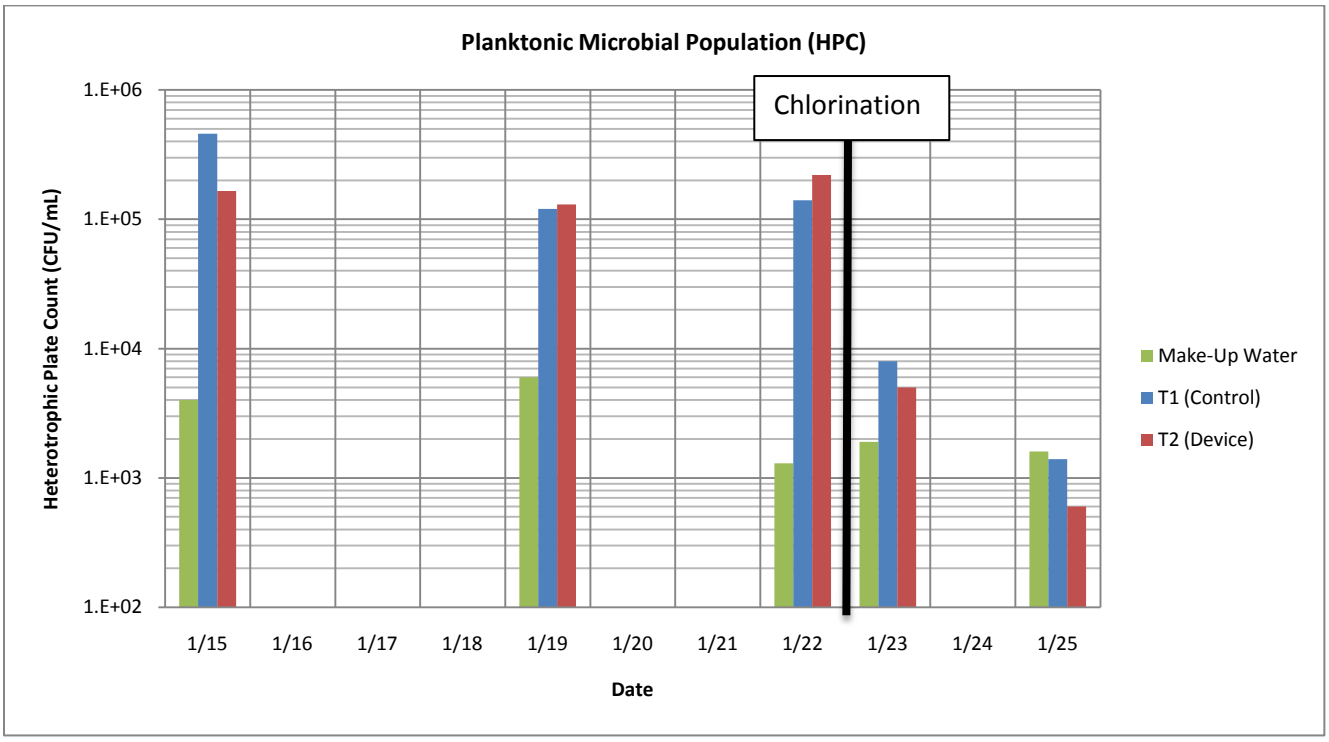


Figure 32 – Pre-device trial chlorination test planktonic microbial population enumerated by HPC

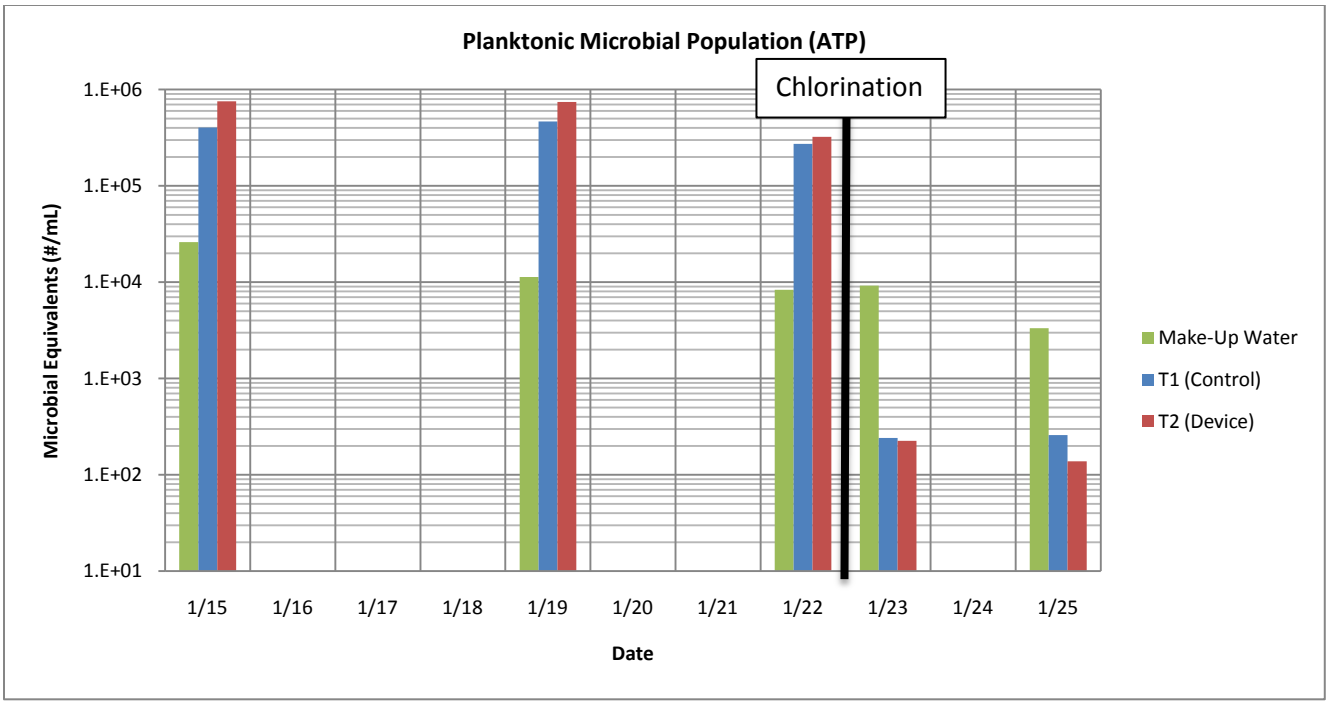


Figure 33 – Pre-device trial chlorination test planktonic microbial population enumerated by ATP

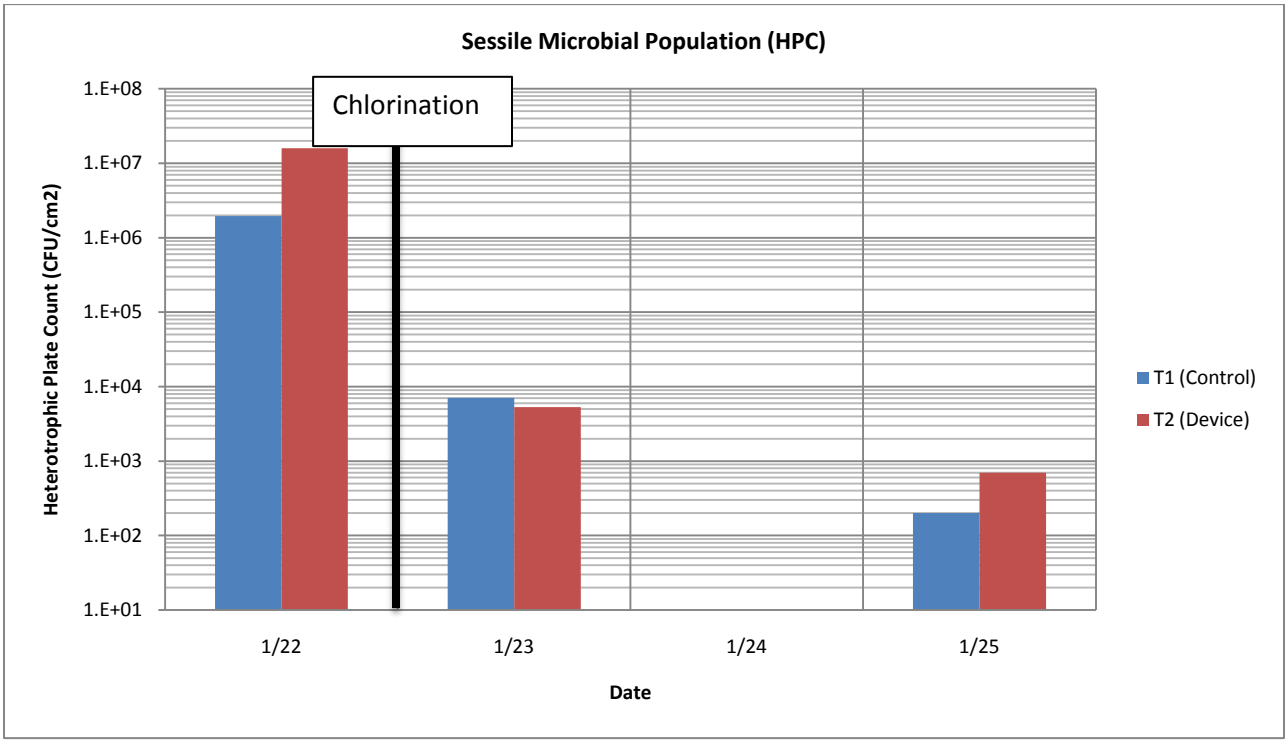


Figure 34 – Pre-device trial chlorination test sessile microbial population

4.7.2 ED/UD Chlorination Test

The second chlorination test was performed immediately after the ED trial and before the towers were prepared for the UD trial. During this chlorination test, T2 (Device) received chemical treatment, while T1 (Control) remained untreated. Chlorination began on 8/21 after biological samples were taken. The chlorination process began with a spike dose of chlorine, followed by a steady flow of chlorine stock solution in order to maintain a free chlorine concentration of approximately 1 mg/L. The planktonic microbial population is shown enumerated by HPC in **Figure 35** and enumerated by ATP in **Figure 36**. Make-up water heterotrophic plate counts observed during this chlorination test were comparable to those observed during each device trial.

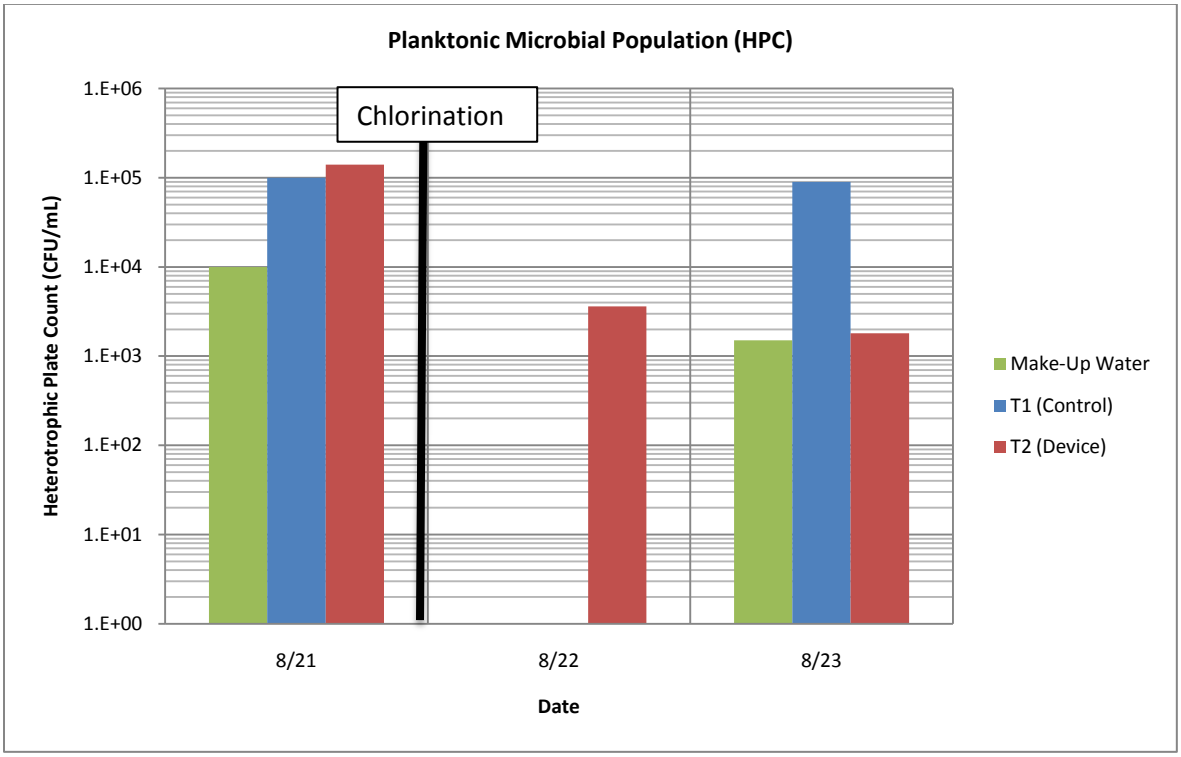


Figure 35 – ED/UD chlorination test planktonic microbial population enumerated by HPC

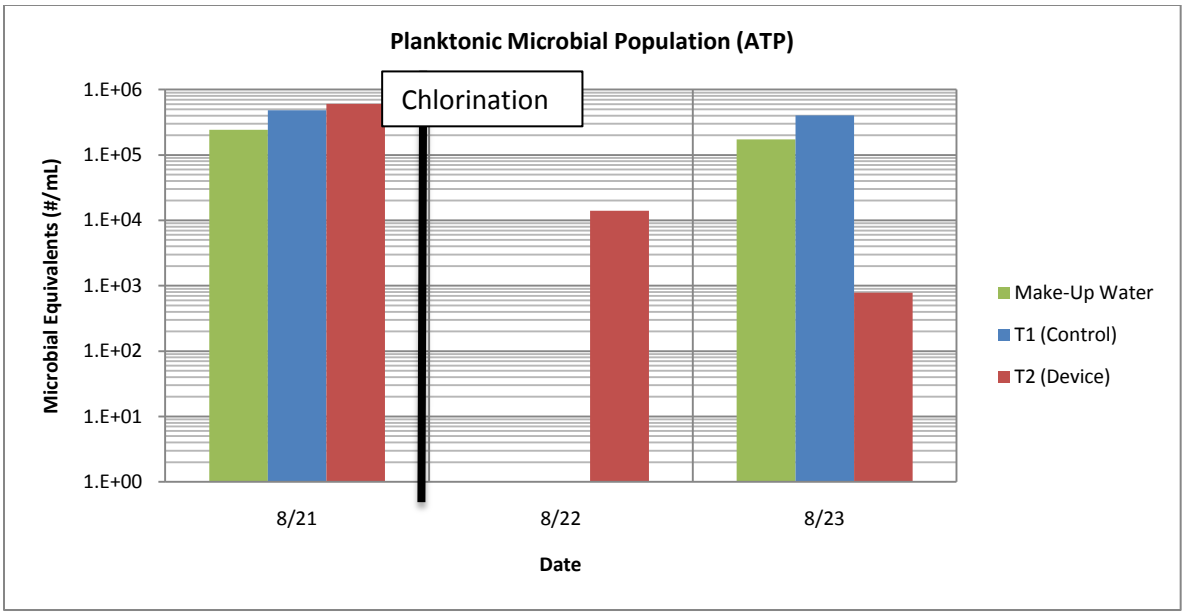


Figure 36 – ED/UD chlorination test planktonic microbial population enumerated by ATP

The planktonic microbial population experienced a 2 log reduction within 2 days of the beginning of chlorination. The sessile microbial population is shown in **Figure 37**. Chlorination resulted in a 4 log reduction in the sessile microbial population.

4.7.3 UD/HCD Chlorination Test

The third chlorination test was performed immediately following the UD trial and immediately before the towers were prepared for the HCD trial. Chlorination of T2 (Device) began on 10/1 following biological sampling, and it was performed using a steady dose of chlorine stock solution. No shock dose of chlorine was used for this test, but instead a gradual increase in chlorine concentration was performed over a period of 5 days until the concentration reached 1 mg/L. The planktonic microbial population enumerated by HPC is shown in **Figure 38** and

enumerated by ATP in **Figure 39**. Make-up water heterotrophic plate counts observed during this chlorination tests were comparable to those observed during all other device trials.

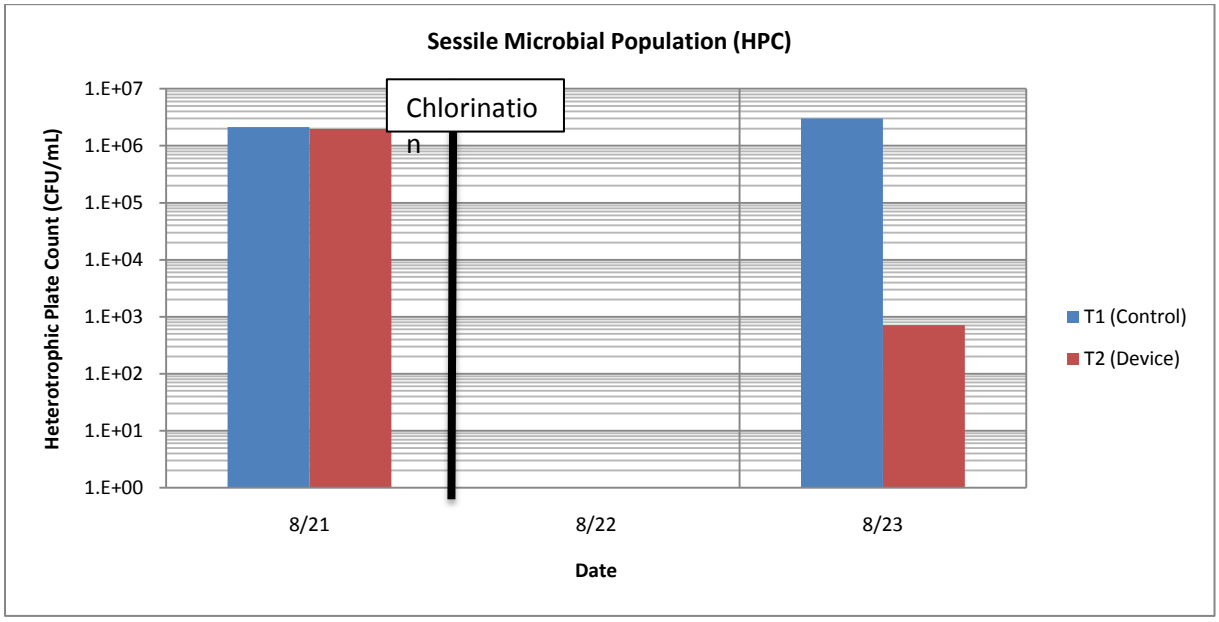


Figure 37 – ED/UD chlorination test sessile microbial population

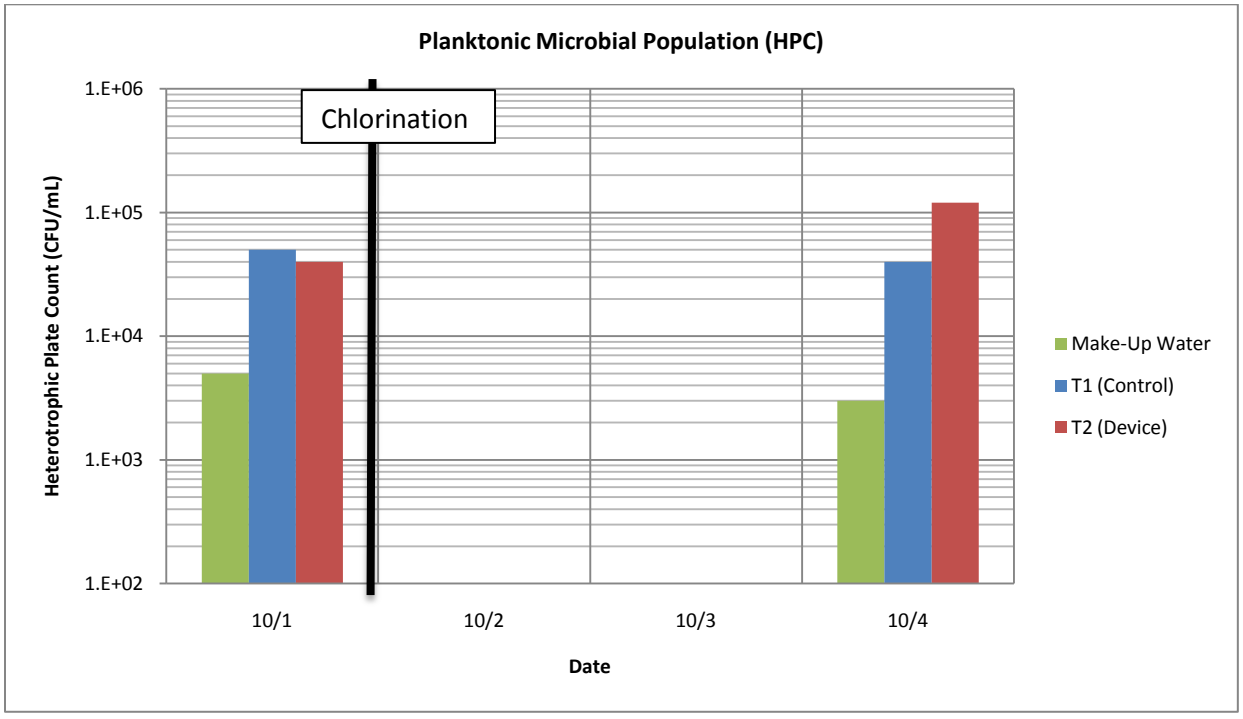


Figure 38 – UD/HCD chlorination test planktonic microbial population enumerated by HPC

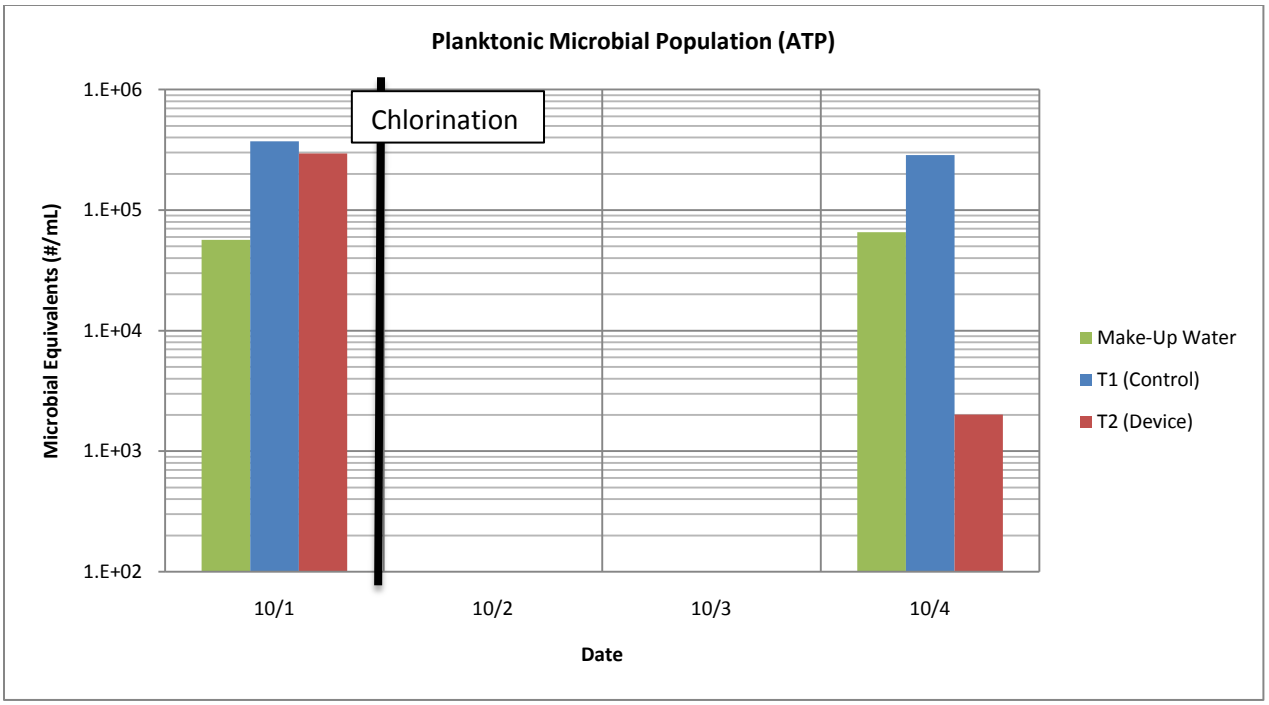


Figure 39 – UD/HCD chlorination test planktonic microbial population enumerated by ATP

The HPC enumeration of the planktonic microbial population showed an increase in microbial activity following chlorination, rather than a decrease. This result is contradictory to the results of the previous chlorination tests. Tower planktonic heterotrophic plate count increased from 4.0 x 10^4 CFU/mL to 1.2 x 10^5 CFU/mL. Sessile heterotrophic plate counts decreased from 3.2 x 10^6 CFU/cm^2 to 6.2 x 10^3 CFU/cm^2 (**Figure 40**). The observed 3 log reduction in the sessile microbial population of T2 (Device) following chlorination indicates that biomass was effectively removed from surfaces within the system. The results in Fig 38 could be due to entrainment of biomass from the surfaces as a result of removal during chlorination. This effect is often observed in field situation: an effective biocide treatment causes release of biomass from surfaces into the recirculating water which results in very high but transient planktonic counts. In the previous chlorination tests, the initial shock dose of chlorine would

have effectively oxidized this material, but the lower chlorine dose used during this particular test were unable to effectively oxidize the majority of this material during three days of operation.

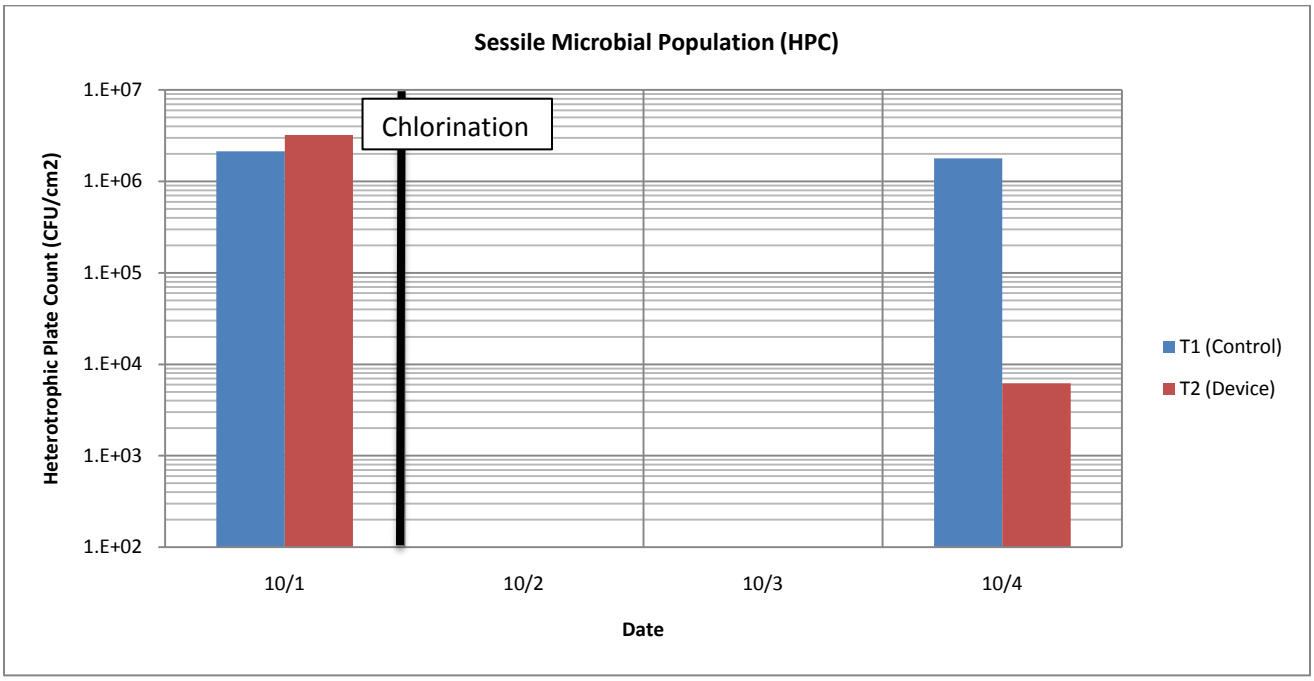


Figure 40 – UD/HCD chlorination test sessile microbial population

4.8 HPC ANALYSES USING ATP MICROBIAL EQUIVALENTS AND POUR PLATE METHOD

While standard plating practices allow for accurate enumeration of planktonic microbial populations in water samples, these methods generally require a minimum of three days in order to process the sample. Microbial population enumeration via ATP measurement, however, is a simple process which may be completed in approximately one hour. As a result, it is highly desirable to establish the practical utility of the ATP method in terms of its ability to accurately quantify the planktonic microbial population in cooling water samples.

In order to determine whether the ATP measurement offers a valid alternative to traditional plating techniques, heterotrophic plate count data and ATP microbial equivalent data from the entire investigation were compared. This comparison indicated that all heterotrophic plate counts and ATP microbial equivalent measurements collected in this study had a coefficient of correlation of $R^2 = 0.37$.

The observed correlation between ATP microbial equivalents and HPC CFU/mL values is quite low. This weak correlation is most likely due to the inclusion of all experimental data, even those that were later discarded. For example, a device trial that was performed with MD at the beginning of this study had to be interrupted due to numerous operational problems. The ATP measurements taken from this period were included in this comparative analysis although they demonstrate a far weaker correlation with HPC measurements than was observed during the six device trials discussed in this report. As a result, these data were removed and the comparative analysis was performed a second time. Excluding the data from this failed device trial, a correlation of $R^2 = 0.77$ was observed between ATP microbial equivalents and HPC

measurements taken from the tower systems. These results indicates that ATP measurement offers a fast means of estimating planktonic activity in a given water sample for microbial populations in excess of 10^5 CFU/mL. However, traditional plating techniques should be used when accurate measurements are required.

The correlation between ATP microbial equivalents and HPC measurements in the system make-up water was also analyzed. A graph comparing these data is shown in **Figure 41**. The correlation observed between the two data sets was relatively low, and make-up water microbial equivalent levels were generally on the order of 1 log value higher than those determined using heterotrophic plate counts. Based on this comparison, it may be concluded that ATP measurement does not offer a reliable estimation of microbial populations when the HPC values are below 10^4 CFU/mL. However, it is possible that water samples with lower microbial populations require larger sample volume during analysis in order to produce accurate results (the sample volume for both the systems water and the make-up water was 50 mL).

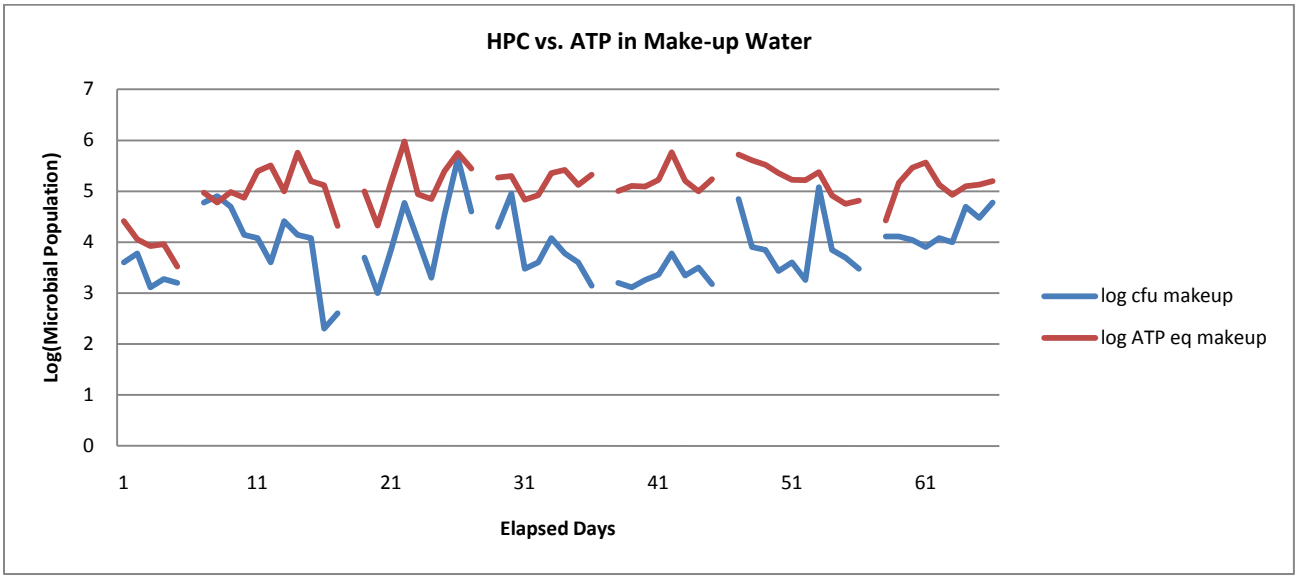


Figure 41 –HPC vs. ATP measurement of planktonic microbial population in make-up water

5.0 SUMMARY AND CONCLUSIONS

Many facilities managers are faced with increasing pressure to utilize "green" technologies for the operation of cooling systems. The goal is to increase efficiency and reduce water consumption. One choice they often face is whether to continue to use conventional chemical water treatment or to use non-chemical water treatment devices as an alternative. There are many different non-chemical devices commercially available today. In order to make an informed decision, objective evidence of the efficacy of these non-chemical devices for cooling water management is needed.

Very limited objective and relevant information is available to verify the efficacy of these non-chemical devices to control microbial growth in cooling towers in the field, which is an essential element in the efficient operation of a cooling tower. Review of the literature reveals few controlled studies on the efficacy of non-chemical devices, and none that were performed under conditions that simulate normal cooling tower operation. Therefore, this study was designed to provide a controlled, independent, scientific evaluation of a variety of classes of non-chemical devices in a model cooling tower operated to simulate typical cooling tower operation.

This study examined the efficacy of five (5) NCDs to control the planktonic and sessile microbial populations in a pilot-scale cooling tower system. The model towers were operated according to specifications from ASHRAE and were representative of typical operation of a

cooling tower, including cycles of concentration, temperature differential, and water chemistry. The make-up water was dechlorinated using an activated carbon filter, a method that has been used previously to produce dechlorinated water for disinfection studies. The use of dechlorinated city tap water was specified to eliminate any antimicrobial effects of chlorine in the make-up water. It would be expected that without chlorine the microbial concentrations in this water would be higher than chlorinated water. However, the concentrations of HPC bacteria that we observered in the make-up water were not unlike the average microbial counts in tap water observed by other investigators.

Under the controlled experimental conditions used in this study, none of the devices were shown to control microbial growth. There was no statistically significant difference in the concentration of HPC observed between the control tower and a tower treated by any of the five NCDs evaluated in this study (i.e., biological and chemical parameters were comparable in T1 and T2 for all device trials). The p-values calculated from t-tests comparing planktonic HPC results for the experimental and control towers were 0.81, 0.92, 0.60, 0.45, and 0.51 for the magnetic, pulsed electric field, electrostatic, ultrasonic, and hydrodynamic cavitation device trials, respectively. The p-values from t-tests comparing sessile HPC results for the experimental and control towers were 0.04, 0.13, 0.43, 0.47, and 0.06 for the magnetic, pulsed electric field, electrostatic, ultrasonic, and hydrodynamic cavitation device trials, respectively.

Repeated chemical treatment of the pilot-scale cooling towers using an industry accepted chemical biocide of known efficacy (chlorine) achieved significant reduction (i.e., three orders of magnitude) in microbial growth in these towers. These “positive control” experiments demonstrated that the model system, when treated with an active biocide, was capable of reflecting this antimicrobial effect.

In addition to not being able to achieve significant reduction in planktonic or sessile microbial activity in the experimental device tower system (T2 (Device)) when compared to the control tower system (T1 (Control)), planktonic microbial levels in the experimental device tower (T2 (Device)) were consistently` higher than recommended industry standard of 10^4 CFU/mL. Furthermore, microbial levels in the Device tower (T-2) were never lower than those observed in the incoming make-up water, further demonstrating that no biological control was demonstrated by these five NCDs under the experimental conditions used in this study.

The findings of this study are not in agreement with previous research published by non-chemical device manufacturers and some reports from independent researchers on these same devices. These studies reported reductions in microbial populations, which were not observed in this study. Therefore, it is important to review the reasons for differing outcomes.

The scientific peer-reviewed literature offers little in the way of reports that verify the antimicrobial effects of many non-chemical devices. For example, electroporation is one of the mechanisms that has been suggested as the basis for an antimicrobial effect from devices such as those using static or pulsed power. While electroporation is effectively used in molecular biology to disrupt cell membranes, it should be noted that the electromagnetic fields used in these applications include high energy (volts to kilovolts), are applied across very short distances (centimeters), and for long exposure times. The conditions applied in the field application of static or pulsed power devices and in the present study produce exposures of considerably less energy and are applied over greater distances and shorter exposure time.

Comparison between HPC analysis by pour plate method and ATP measurements revealed that the ATP measurement offers a fast means of estimating planktonic activity in a water sample containing microbial populations in excess of 10^5 CFU/mL. On the other hand,

ATP measurement does not offer a reliable estimation of microbial populations when the HPC values are below 10^4 CFU/mL. It is possible that water samples with lower microbial populations require larger sample volume during analysis in order to produce accurate results.

In conclusion, none of the NCDs evaluated in this study demonstrated significant biological control in model cooling tower system operated under realistic process conditions that may be encountered in the field. However, this study still offers several opportunities for continued investigation of non-chemical treatment devices. The effects of residual chlorine in the incoming make-up water were not analyzed during this investigation, and the effect of some non-chemical water treatment devices may be augmented by the presence of chlorine. In addition, the combined effects of chemical (e.g., oxidizing and non-oxidizing biocides) and physical treatment technologies on the control of biological growth in cooling towers may offer significant advantages. Combining hydrodynamic cavitation as well as ultrasonic cavitation with chemical disinfection has been shown to achieve significant kill of different microorganisms and the use of such hybrid systems (both chemical and physical) certainly warrants further investigation.

The results from this study show that effective microbial control in cooling water systems may not be achieved using a non-chemical device as the sole method of water treatment. Consequently, equipment operators, building owners and engineers should consider taking more frequent water sample tests for their systems that rely on NCD devices for biological control. If the testing shows an issue, one possible measure is to add chemical treatment capability to their system.

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