Fate and Transport of Endocrine Disrupting Compounds during Wastewater Treatment:
The Role of Colloidal and Particulate Material

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Fate and Transport of Endocrine Disrupting Compounds during Wastewater Treatment: The Role of Colloidal and Particulate Material

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(ABSTRACT)

The presence of biologically-active estrogenic endocrine disrupting compounds (EDCs) in treated effluents from biological wastewater treatment facilities has prompted wide-spread interest in the behavior of these contaminants during the activated sludge process. The yeast-estrogen screen (YES) was used to quantify the estrogenic activity of samples taken from different areas of three wastewater treatment facilities. An estrogenic mass-balance around these facilities revealed that the majority of influent estrogenic activity was removed in the activated sludge process, but the main route for EDC discharge to the natural environment was via the treated effluent. The estrogenic activity in the effluent from a membrane bioreactor (MBR) was lower compared to a fully aerobic activated sludge process using secondary clarification, suggesting that enhanced removal of particulate and colloidal material may improve EDC removal efficiency.

Colloidal material was obtained from settled mixed liquor suspended solids (MLSS) collected from a pilot MBR and a full-scale activated sludge process that included anoxic and aerobic zones. The MLSS was sized fractionated by filtration, and used to quantify the sorption coefficients for pyrene, 17β-estradiol (E2), and 17α-ethinylestradiol (EE2) by fluorescence quenching. The MLSS-derived colloidal organic carbon (COC) sorption coefficient (K_{coc}) for pyrene ranged from (< 1 to 80) L/kg_{coc}, indicating a similar affinity for pyrene compared to natural organic matter. K_{coc} coefficients for E2 ranged between (< 1 to 158) L/kg_{coc} for E2 and (< 1 to 228) L/kg_{coc} for EE2, and are the highest E2 and EE2 sorption coefficients reported in the literature to date. There was a strong correlation between the K_{coc} coefficients and molar extinction coefficient at 280 nm (e_{280}) for pyrene and E2, suggesting that the interaction of the π-electrons is an important factor in determining overall sorption behavior. There was no such correlation for EE2. Based on the K_{coc} coefficients and COC concentrations of the samples, between 1 and 50% of the aqueous E2 and EE2 concentrations were associated with colloidal material.

In a novel application of the YES bioassay, the bioavailability of colloid-associated E2 was quantified by comparing the EC_{50} values of the dose-response curves generated in the presence and absence of size fractionated COC. An increase in EC_{50} values as a function of COC concentration was attributed to a reduction in bioavailability of E2, suggesting that MLSS-derived COC can reduce, but not eliminate, the biological impact of EDCs. However, there was a high degree of variability in the EC_{50} values, and estimates of the colloid-associated E2 fraction based on the K_{coc}-e_{280} correlation were unsuccessful in accurately predicting increases in EC_{50} values. Nevertheless, the YES bioassay may represent a powerful tool in determining the bioavailability of EDCs in complex environmental samples.

Results from this research effort suggest that the colloidal phase derived from activated sludge systems represents an important transport vehicle whereby EDCs and other trace organic compounds can enter into the natural environment. Consequently, wastewater treatment plants discharging to sensitive ecosystems or involved with direct water reuse programs should optimize the treatment process to remove colloidal material.

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DEDICATION

This work is dedicated to my family (both related and adopted), whose steadfast support and encouragement made the often arduous process of discovery more enjoyable, to the memory of my grandparents, Herb and Ruth Holbrook and Floyd and Dorothy Kirkham, who were environmentalists in their own right, and to the memory of Tracy Lynn Hobbs, who began to teach me the value and power of community.

It is my sincere hope that the results of this research will contribute to the field of Environmental Engineering by enlightening both scientists and practicing engineers to the complexity of pollutant transport from biological wastewater treatment systems. The need for a better understanding of trace contaminant behavior in the natural environment and wastewater treatment systems is substantial, and it would give me a great sense of satisfaction if this work encourages others to pursue a similar line of research.

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Chapter 1. Introduction

There has been increasing concern about the presence and effect of estrogenic compounds released into the natural environment from wastewater treatment facilities. The linkage between estrogenic compounds and sewage began in the late 1980s and early 1990s with reports that fisherman in the United Kingdom were finding an abnormally large number of wastewater treatment lagoon reared male fish with both male and female sexual organs (1). This initial report generated a flurry of research activity to determine the cause of the sexual disruption in fish and, consequently, many other reports have shown a severe impact of sewage discharge on wildlife. There have been two well documented impacts of treated effluent on fish in the natural environment, including vitellogenin production (2-4) and widespread sexual disruption of male fish (5,6). Vitellogenin is a protein which is produced in the liver as a precursor to egg yolk development (3,7), and is typically found in high concentrations only in spawning females. Consequently, high concentrations of vitellogenin in male fish indicate exposure and response to an estrogenic compound.

These environmental observations led to more controlled studies using caged fish (e.g., 1,8). These caged fish were placed upstream and downstream of wastewater treatment plants, and invariably increased levels of vitellogenin were found in the male fish maintained in the downstream locations. The environmental observations and controlled studies using fish as sentinel species demonstrated that estrogenic endocrine disrupting compounds are present in treated effluents. In this context, an endocrine disrupting compound is any exogenous substance that alters the function of the endocrine system (9) and, consequently, causes adverse health effects such as protein synthesis or abnormal organ development.

Estrogenic endocrine disrupting compounds (EDCs) can be categorized into four main categories: reproductive hormones and steroids (2,9,10); detergent metabolites (11,12); plasticizers (1,8,13,14); and biocides (15,16). In 2002, Dana Kolpin and associates at the United States Geological Service (USGS) performed a reconnaissance of the US streams in regards to organic contaminants (17). These researchers sampled 139 streams from various parts of the country, and concluded that there were detected levels of hormones, detergent metabolites, and plasticizers in over 80, 70, and 60% of the samples, respectively. Furthermore, the majority of these compounds are believed to originate from treated wastewater effluent. Based on the results of this study (17) and the nearly ubiquitous presence of EDCs in the natural environment, there is a clear need to understand the behavior of these trace organic contaminants during wastewater treatment processes in order to mitigate their release into receiving streams.

A literature review (Chapter 2) was conducted prior to evaluate the current state of knowledge regarding the behavior of EDCs in the activated sludge process. The literature revealed that the majority of investigations were focused on monitoring EDCs in the influent and effluent of wastewater treatment plants (e.g., 18-22) rather than focusing on mechanisms that define the fate of these chemicals. Subsequently, a field investigation (Chapter 3) was conducted with two main objectives: i) to investigate the behavior of EDCs during different wastewater treatment and solids handling processes; and ii) to develop hypothesis regarding the EDC transport mechanisms during the activated sludge process. In order to quantify the estrogenic activity of the environmental samples, the yeast estrogen screen (YES) bioassay was used (23). One of the conclusions of the field study was that between 26 to 43% of the influent estrogenic activity was discharged from the activated sludge process as treated effluent while the waste activated sludge contained only 2 – 14% of the influent estrogenic activity. Consequently, between 51 – 67% of the influent estrogenic activity was either removed (i.e., mineralized) or not detected by the YES bioassay protocol.

The unique aspect of the study was the side-by-side comparison of a conventional activated sludge facilities (CAS) and membrane bioreactors (MBR). The CAS system uses secondary clarification for liquid solid separation whereas the MBR system uses a membrane filter. Although the two biological wastewater treatment systems were operated with similar conditions (i.e., sludge age) and treated the same influent wastewater, the estrogenic activity distribution was different. For example, the MBR system removed 67% of the influent estrogenic activity whereas the CAS system removed 51%. The difference in estrogenic activity removal was attributed to increase in suspended solids (particulate) and colloid removed by the MBR system compared with the CAS system.

The sorption of organic contaminants to colloidal material is important in both natural environments and engineered systems for three main reasons. First, the organic colloids can increase the apparent aqueous solubility of organic contaminants (24-27), signifying that organic contaminants can be found above their maximum aqueous solubility if colloids are present. Secondly, the sorption of organic contaminants to colloidal material reduces, but does not eliminate, the bioavailability, or toxicity, of organic contaminants (28-30). Lastly, colloidal material reduces the biodegradability of pollutants (e.g., 31). Subsequently, several hypothesis addressing the impact of colloidal material on the behavior of EDCs during the activated sludge process were considered:

1. EDCs in from biological wastewater treatment plants will be bound to colloidal organic material.
2. Sorption of EDCs to colloids is related to structural characteristics of the colloidal organic carbon.
3. Colloid-bound EDCs will have lower but still measurable bioavailability compared to dissolved EDCs.

In order to test these hypotheses, two objectives were developed and addressed:

1. Calculate the sorption coefficients between model EDCs and colloidal material derived from different wastewater treatment processes; and
2. Determine the bioavailability of colloid-associated E2 to the hER.

Specific experiments that address these objectives are outlined in each chapter. Initially, it was necessary to select and validate a method for detecting binding between contaminants and colloidal organic carbon that was recovered from wastewater treatment plant samples. Fluorescence quenching was selected as the method. Although not considered an EDC, pyrene is commonly used in fluorescence quenching experiments and was used to validate the method and compare the sorption behavior of mixed liquor-derived colloidal material to natural organic matter (NOM). The results are presented in Chapter 4 and show that colloidal material from activated sludge processes has a similar affinity for pyrene compared to NOM and demonstrated a strong correlation between the sorption coefficient and aromatic content of the colloidal material. However, the affinity of pyrene for colloids derived from an activated sludge facility using secondary clarifiers was substantially greater than colloids derived from an MBR system.

Next, the sorption coefficients between colloidal material derived from activated sludge processes and two EDCs, E2 and 17α-ethinylestradiol (EE2), were quantified using the fluorescence quenching method. The results are presented in Chapter 5 and indicate that simple partitioning models underestimate the affinity of E2 and EE2 for activated sludge-derived colloidal material. The correlation between colloidal aromatic content and sorption coefficients was fair ($r^2 = 0.50$) for E2 but not significant for EE2. However, between 1 and 50% of the aqueous E2 and EE2 was found to be associated with colloidal material. It was concluded that treatment processes that enhance the capture of colloidal material (like MBRs) will have a lower estrogenic activity in the treated effluent compared with facilities that do not promote colloidal removal.

It became apparent that sorption of EDCs to activated sludge-derived colloidal material was occurring in the systems studied during this project; however, the protocol used to detect those compounds that were both dissolved and less readily available in solution relied on a solvent extraction step. Additional experiments were conducted that involved a novel and unique use of the YES bioassay in an effort to determine how bioavailable EDCs were that had been incubated with activated sludge-derived colloidal material. Chapter 6 presents a novel and unique use for the YES bioassay to test for the bioavailability of EDCs, and one that may be further developed for future applications in risk assessment and modeling. The results of these experiments indicate that the colloidal organic carbon concentration is an important factor in reducing the bioavailability of E2.

Finally, the engineering significance of this work is discussed in Chapter 7. In summary, the affinity of EDCs for colloidal material derived from a biological wastewater treatment process is dependent on concentration and nature of the colloidal organic material. EDCs that are associated with colloidal material will have a different fate within the treatment system and in the receiving water compared to EDCs in the dissolved phase. Wastewater treatment processes that reduce the concentration of colloidal material, such as filtration and/or coagulation, will reduce the effluent EDC concentration.

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Chapter 2. Literature Review

Estrogenic Endocrine Disrupting Compounds and Wastewater Treatment

The nearly ubiquitous presence of anthropogenic pollutants in the aquatic environments (1) has ignited interest in the ability of biological wastewater treatment facilities to effectively remove trace organic contaminants from treated effluent. Trace organic contaminants include antibiotics, personal use products, plasticizers, stabilizers, detergent metabolites and hormones. Many of these compounds are referred to as endocrine disruptors due to their ability to interfere with the biochemical cycles of hormone regulation (2). Kavlock *et al*. (3) defines an environmental endocrine disruptor as "an exogenous agent that interferes with the production, release, transport, metabolism, binding, action or elimination of natural hormones in the body responsible for the maintenance of homeostasis and the regulation of developmental processes". Estrogenic endocrine disrupting compounds (EDCs) have the ability to directly or indirectly affect the reproductive system, and a long list of EDCs found in the natural environment include DDT (4), PCB (5,6), phenols (7,8), organochlorines (9), phthalates (10-12), alkylphenolic ethoxylates (APEOs) (13-18), and natural and synthetic estrogenic hormones (1,19-28). APEOs and estrogenic hormones have received the greatest attention from environmental scientists due to their presence in wastewater effluents (e.g., 14,23-25) and demonstrated impact on wildlife (e.g,, 29,30). Of these two groups, it appears that the estrogenic hormones represent the predominant form of estrogenic activity in wastewater effluents (24,25), although high levels of APEOs have been reported for some treatment facilities (31). The literature pertaining to APEOs and estrogenic hormones will be discussed separately.

Alkylphenol Ethoxylates. APEOs are nonionic surfactants that have been used in cleaning products for more than 40 years (32). The most common derivatives used in industry are nonylphenol ethoxylate (NPEO) and octylphenol ethoxylate (OPEO). The majority of literature available on APEOs utilizes NPEO and NP as a representative family. Of the NP isomers, the para-substituted group (4-NP) is the most common (33). Wastewater discharges have been identified as being a significant source of APEO and associated metabolites, and both are found in the natural environment (31,34).

Ahel *et al.* (35-38) and Giger *et al*. (39) have thoroughly investigated the fate of alkylphenol ethoxylates (APEOs) during the different processes of wastewater treatment. During degradation, higher oligomers (nEO > 8) are eliminated and form metabolites, including alkylphenol polyethoxycarboxylic acid derivatives (AP1EC and AP2EC, respectively), alkylphenol di- and mono-oxylates (AP1EO and AP2EO, respectively), and alkylphenols. Anaerobic degradation produces AP1EO and AP2EO in addition to alkylphenols. Hydrophobicity, toxicity, and estrogenicity increase as the ethoxylate chain is reduced. The metabolic products have been shown to resist further degradation in activated sludge facilities and are therefore discharged into the aquatic environment.

The removal of APEO oligomers and AP is approximately 40% (molar basis) during the activated sludge process (35). This is in fair agreement with the removal efficiency reported by Di Corcia *et al.* (40) of 53 (±19%) from their survey of activated sludge facilities in Rome. Ahel *et al.* (35,36) reported a strong correlation between nitrification efficiency with NPEO and associated metabolite elimination during secondary treatment. They concluded that a longer SRT, lower F/M ratio and low ammonium concentration would favor APEO and AP removal. A survey of both domestic and industrial wastewater facilities in the United States determined a high removal (> 90%) of NPEO from the aqueous phase (17,41)with correspondingly high levels in final biosolids (13). The range of total APEC and AP compounds found in the treated effluents are between 10 and 300 μg/L in Switzerland (42) and between 20 and 50 μg/L in the US (40). Ahel *et al.* (36,37) estimated that up to 95% of AP (contributing 40% of the total effluent APEO load) was bound to digested sludge. NP, NP1EO and NP2EO were detected at concentrations greater than 50 mg/kg in treated biosolids (43). Knudsen *et al.* (44) present an interesting case study in the use of post-aerobic processes following anaerobic digestion. They found that post-aeration reduced the concentration of total NP compounds to approximately 15 mg/kg.

Nonylphenol comprised approximately 90% of the total NPEO concentration found in sediment with concentrations ranging from 170 – 3,000 μg/L (45). A triphasic degradation process was observed in sludge-amended soils (32). The first degradation phase consisted of a fast decline for 3 weeks, followed by a slower rate for the next 9 weeks, with a plateau over the following 7 months. After a one-year period, there was some persistence of all parental compounds. Marcomini *et al.* (46) noted that biodegradation of NP and NP1EO was much faster under aerobic than anaerobic conditions.

Estrogenic Hormones. The estrogenic hormones include the natural hormones estrone (E1), 17β-estradiol (E2), and estriol (E3) as well as the synthetic hormone 17α-ethinylestradiol (EE2). Between 10 and 100 μg of E1, E2, E3 and EE2 are excreted daily by women, while pregnant women may excrete up to 30,000 μg of estrogen, primarily in the form of E3 (47-49). Estrogenic hormones are mainly excreted from the human body as non-active conjugates of sulfuric and glucuronic acids (49). However, many microorganisms (e.g., *Escherichia coli*) exhibit sulphatase and glucuronidase activity (50), and are therefore able to convert the non-active conjugates into biologically active unconjugated estrogens. Baronti *et al.* (49) suggested that deconjugation occurs preferentially in the sewers, although Nasu *et al.* (51) suggested that deconjugation may also take place in the primary clarifier.

Reported influent and effluent concentrations of E1, E2, E3, and EE2 cover a fairly broad range of values, and may be affected by factors such as infiltration/inflow and per capita water usage. For example, Johnson *et al.* (52) developed an empirical relationship to determine the influent concentrations of E1, E2 and EE2 based on the populations served by the treatment works and the influent flow rate. Based on the measured data from Baronti *et al.* (49), average influent concentrations of E1, E2, E3, and EE2 are 80, 11.5, 3, and 52 ng/L, respectively, although there is a wide range of reported values from other countries (Table 2-1). While these concentrations are expected to vary between treatment works, it is important to note that the concentrations of these compounds are much lower (low to mid ng/L) than other organic contaminants that are routinely monitored in wastewater effluents, thereby indicating the potential of these compounds to be of environmental concern at extremely low concentrations.

Table 2-1 – Summary of Influent and Effluent concentrations of estrogenic hormones (ng/L)[a].

Facility	Estriol (E1)		Estradiol (E2)		Estrone (E3)		Ethinylestradiol (EE2)		Ref.
	Influent	Effluent	Influent	Effluent	Influent	Effluent	Influent	Effluent	
Cobis, Italy	84 (59)	3.7 (3)	16 (8)	1.5 (1)	71 (35)	10.1 (5)	3.9 (5)	0.65 (0.3)	(49)
Fregene, Italy	71 (37)	1.1 (0.7)	9.2 (5)	1 (0.7)	67 (17)	4.1 (2)	3.4 (2)	0.68 (0.7)	(49)
Ostia, Italy	130 (48)	1.1 (0.4)	15 (7)	2.4 (1)	51 (13)	45 (25)	2.5 (2)	0.8 (0.3)	(49)
Roma Sud, Italy	54 (42)	8.7 (6)	8.6 (2)	1.9 (0.9)	35 (10)	30 (16)	2.9 (2)	0.7 (0.4)	(49)
Roma Est, Italy	66 (46)	0.8 (4)	9.3 (2)	0.7 (0.1)	50 (14)	7.7 (3)	2.3 (2)	0.4 (0.2)	(49)
Roma Nord, Italy	79 (42)	2.6 (3)	12 (3)	1 (0.6)	37 (8)	14 (15)	2.9 (2)	0.48 (0.1)	(49)
Southwest Germany (18 WWTPs)	na[b]	na	na	2.9 (4)	na	5.1 (7)	na	1.8 (3)	(53)
California, USA - WWTP 1	na	na	na	3.8 (0.7)	na	na	na	1.9 (0.6)	(23)
California, USA - WWTP 2	na	na	na	0.77 (0.4)	na	na	na	0.33 (0.2)	(23)
California, USA - WWTP 3 (MF)	na	na	na	1.4 (1)	na	na	na	0.14 (0.03)	(23)
California, USA - WWTP 3 (RO)	na	na	na	0.24 (0.2)	na	na	na	< 0.1	(23)

[a]Values listed are average (standard deviation). [b] na = not applicable.

Reported effluent concentrations for the estrogenic hormones range from 3 to 9 ng/L for E1 (20,49), 0.1 to 5 ng/L for E2 (23,25), 1 to 8 ng/L for E3 (49) and 0.1 to 9 ng/L for EE2 (20,49,53). A summary of effluent estrogenic hormone concentrations is found in Table 1. Removal of E1, E2, E3, and EE2 were calculated to be 96, 88, 65, and 79%, respectively, for the Bartoni *et al.* investigation (49). Similarly, Johnson *et al.* (52) calculated E1, E2, and EE2 removal of 72, 87, and 85%, respectively. Lower E2 and E3 removal efficiencies (64 and 14%, respectively) were reported by Ternes *et al.* (22) for the Frankfurt, Germany WWTP, suggesting that variations in effluent quality can be expected from biological treatment systems. Indeed, Williams *et al.* (54) observed a 2- to 3-fold difference in E1 and E2 concentrations from composite effluent samples taken on consecutive days. However, the literature suggests that greater than 75% of individual estrogenic hormones are typically removed by the activated sludge process (e.g., (55)). Batch microcosm experiments using ^{14}C-E2 and ^{14}C-EE2 by Layton *et al.* (56) demonstrated a fairly rapid mineralization of ^{14}C-E2 by mixed liquor suspended solids (MLSS) derived from municipal activated sludge processes while the mineralization rate of ^{14}C-EE2 was much slower. Similarly, the ^{14}C-E2 mineralization rate by MLSS from an industrial activated sludge process was slower compared to a MLSS from a municipal wastewater treatment facility suggested that the composition of bacterial populations are an important factor in E2 removal efficiency. Ternes *et al.* (50) also demonstrated that E2 was rapidly oxidized to E1, and that EE2 was fairly recalcitrant to biodegradation. However, as noted by these two research groups (50,56), the concentrations of E2 and EE2 used in these experiments were on the order to 10^2 to 10^3 times greater than normally found in sewage, and therefore may not represent “real-world” conditions. Consequently, the impact of biodegradation on determining effluent concentrations of estrogenic hormones may be overstated.

Sorption of estrogenic hormones by wastewater-derived particles and colloids should be expected due to the lipophilic/hydrophobic nature of these contaminants. For example, the octanol-water partition coefficients (K_{ow}) for the estrogenic hormones are 3.13 for E1 (57), 4.01 for E2 (57), 2.7 for E3 (58), and 3.67 for EE2 (57), indicating a moderate level of relative hydrophobicity. Huang and Sedlak (23) suggested that approximately 70% of E2 should be associated with the organic particles during the activated sludge process (assuming a MLVSS concentration of 2 g/L). Ternes *et al.* (59) detected up to 37, 49, and 17 ng/g of E1, E2, and EE2, respectively, in activated and anaerobically digested sludges, indicating that estrogenic hormones are present in the particulate phase and also persist during the anaerobic digestion process. To our knowledge, the study by Ternes *et al.* (59) is the only investigation to quantify individual estrogenic hormones associated with the particulate phase of wastewater treatment processes. The affinity of estrogenic hormones for colloidal material has been demonstrated indirectly by Huang and Sedlak (23), who reported lower effluent concentrations of E2 and EE2 from treatment facilities that employed advanced treatment unit processes such as sand filtration, membrane filtration, and reverse osmosis.

The sorption behavior of estrogenic hormones with respect to MLSS has not been well studied. However, several research groups have measured the affinity of selected estrogenic hormones for natural organic matter (NOM). Lai *et al.* (58) demonstrated that sorption of E1, E2, E3, and EE2 by river and estuarine sediments were non-linear, indicating that adsorption rather than partitioning was a critical sorption mechanism. Lai *et al.* (58) also demonstrated sorption competition and greater sorption of estrogenic hormones by sediments in high salinity environments. Holthaus *et al.* (60) performed E2 and EE2 sorption experiments using both bed and suspended sediments taken from five British Rivers. These researchers calculated distribution coefficients (K_D) ranging from 4 to 74 L/kg for E2 and 8 to 121 L/kg for EE2 for the bed sediments, and 21 to 122 L/kg for E2 and 19 to 260 L/kg for EE2 for the suspended sediments. Furthermore, they reported a significant increase in bed sediment K_D coefficients with smaller particle sizes and higher organic carbon concentrations. In comparison, Bowman *et al.* (61) calculated K_D coefficients of 141 and 102 L/kg for E2 and E3, respectively, using estuarine sediments, and determined that the distribution coefficients for colloids were two orders of magnitude greater than the K_D coefficients from the sediment. Bowman *et al.* (61) also found a strong correlation between organic carbon concentration and specific surface area on the distribution coefficients, which is in agreement with Holthaus *et al.* (60) .

More recently, Yamamoto *et al*. (62) conducted sorption experiments between DOM-surrogates, colloidal material, and selected estrogenic compounds. These investigators concluded that the sorption coefficients (K_{oc}) were relatively independent of the compound's K_{ow} coefficient. However, strong correlations between K_{oc} coefficients and both ultra-violet adsorption at 272 nm and the phenolic group concentration of the DOM suggest that the sorption behavior is related to π-electron interaction and hydrogen bonding.

Methods for Detection Estrogenic Compounds. Two approaches are employed to detect the presence of estrogenic compounds in environmental matrices. The first is qualification and quantification of individual compounds using various analytical methods. Typically, these methods consist of (i) extraction using either liquid-liquid extraction (e.g., 63) or solid-phase extraction with various stationary phases (e.g., 22,64), (ii) concentration by evaporating the extraction solvent (e.g., 23), (iii) derivitization (e.g., 25,65), and (iv) detection by an instrument. These instruments have included gas chromatography-mass spectroscopy (GC-MS) (e.g., 20), GC-MS-MS (e.g., 23,59), and high performance liquid chromatography-mass spectroscopy (LC-MS) (e.g., 52). The reader is directed to a recent review article detailing the different analytical detection procedures by Lopez de Alda and Barcelo (66) and the references therein for further information.

The second method quantifies the estrogenic activity of an environmental matrix be measuring the positive response of a bioassay without identifying the individual estrogenic contaminant. The two most common bioassays are the yeast estrogen screen (YES) (8,67) and MCF-7 breast cancer cell assay (E-Screen) (12). Similar to detection by an analytical instrument, this method also typically employs a liquid-liquid or solid-phase extraction and concentration procedure. The concentrated extract is then introduced to the bioassay, and the estrogenic activity of the concentrated sample is then quantified. The YES method is the most commonly used bioassay for wastewater applications (e.g., (7,20,28,68-71)) although this method of quantifying estrogenic activity does have limitations (72).

Wastewater-Derived Material – Components and Composition

Mixed Liquor Suspended Solids. MLSS found in activated sludge systems are a complex, heterogeneous consortium containing microorganisms, exopolymeric substances (EPS), inorganic cations, soluble microbial products and debris (73-77). Based on electronic microscopic analysis, Jorand (78) reported that biological flocs are composed of microbial cells that are completely surrounded by and imbedded in a heterogenous EPS mixture. EPS are arguably the most complex and important component of activated sludge floc. Due to the location of EPS, their chemical composition affects the surface properties of bacterial flocs. EPS is composed primarily of proteins, polysaccharides, lipids, humics and fulvics, and nucleic acids (79-85), are important in floc structure and strength (86,87), and have significant sorption properties (85,88-92). Although there is still a great deal of research in determining operating parameters that influence the quantity and relative components of EPS, several researchers have shown that feed composition (93), sludge retention time (88), environmental conditions (82) and bacterial species (77) are all important factors in EPS formation.

Due to the importance of EPS in activated sludge systems, several researchers have tried to determine what individual component dominates surface properties. The majority of literature on this subject is in the context of flocculation (e.g., 94-96). However, because sorption of organic contaminants onto activated sludge-derived material will undoubtedly be related to the surface properties of the undissolved particles solids, these experiments are very relevant. Liao *et al.* (88) showed an inverse relationship between the polysaccharide content of EPS and SRT and a direct relationship between the protein content of EPS and SRT; the DNA content of the EPS did not change significantly as a function of SRT. Liao *et al.* (88) concluded that the proportions of EPS components were more important in controlling surface properties (hydrophobicity and surface charge) than the quantity of individual components. Morgan *et al.* (82) also suggested that the concentration of biopolymers was not important for different surface properties but rather the arrangement of the function groups. This suggests that hydrophobic amino acids may be an important parameter in controlling sorption potential. Jorand *et al.* (97) reported that the hydrophobic fraction of EPS consisted only of proteins. This is consistent with Liao *et al.* (88) who found a weak correlation with hydrophobicity and protein content. A negative correlation between the protein concentration and C/N ratio of EPS was demonstrated (98).

Colloidal and Dissolved Material. It is generally believed that the majority of colloidal and dissolved organic materials discharged from biological wastewater facilities are microbially derived (99-107), and therefore the organic carbon contained in treated effluent possess unique characteristics compared to NOM (108). The distinction between the colloidal phase and dissolved phase is arbitrarily defined, and different investigators have used a range of sizes in their experiments. For example, the dissolved phase has been defined as any material less than 500 daltons, (109), less than 1,000 daltons (1 kD) (110,111), less than 3 kD (112), and less than 0.22 μm (102,113). Based on thermodynamic properties, Gustafsson and Gschwend (114) suggest that aquatic colloids should be any material which is unaffected by gravity and provides an environment where organic contaminants can escape from the aqueous environment. Based on this definition, the dissolved phase should be considered to lie between 1 and 10 kD; therefore, for wastewater applications the colloidal phase should be defined as material between 1 kD and 1.5 μm in size (115).

The wastewater treatment literature does not use such a rigorous distinction between colloidal and dissolved material and instead uses a comprehensive term called soluble microbial products (SMPs). According to Barker and Stuckey (116), SMPs are defined as "the pool of organic compounds that are released into solution from substrate metabolism (usually with biomass growth) and biomass decay". Most of the literature involving SMPs is related to the molecular weight distribution and composition of organic material.

However, there are differences within the literature suggesting that all the variables related to SMP production are still unknown. For example, Saunders and Dick (117) reported a decrease in high molecular weight (> 100 kD) COD fractions at increasing SRTs, whereas Barker and Stuckey (101) observed that higher molecular weight organic material fractions became dominant at higher SRTs. Manka and Rebhun (118) found that the largest fractions of organic matter in domestic secondary effluents were found in the highest molecular weight fraction (> 20 kD) and lowest molecular weight fraction (< 500 daltons). Amy *et al*. (102) reported a transformation of relatively simple wastewater organic material (< 500 daltons) in primary effluent into more complex microbial byproducts (0.5 – 5 kD). Several researchers have observed a bimodal molecular weight distribution of effluent organics from activated sludge facilities (119-122).

Organic matter composition of wastewater effluents is also discussed in the literature (101,123-127), and include proteins, polysaccharides, humic and fulvic acids, nucleic acids, enzymes, and structural components of cells. However, the dominant molecular group among effluent organic matter varies widely in the literature. For example, Manka and Rebhun (118) reported that proteins and humic acids were the major effluent organic carbon contributors, while Ma *et al*. (128) found that wastewater effluents were enriched in fulvic acids compared to humic acid.

Two investigators have examined the metal chelating properties of SMP. Kuo and Parkin (6) utilized anaerobic chemostats and found the production of some moderately strong nickel chelating compounds. The maximum nickel chelating production was found to occur at an SRT of 40 days. Bender *et al.* (129) concluded that high molecular weight compounds (> 100 kD) had substantial copper binding capacity.

Sorption of Hydrophobic Compounds by Individual Wastewater Organic Components

Proteins. The types and primary structure of amino acids influences the secondary and tertiary structure of proteins and thus serves as an important mechanism for sorption properties. The hydrophobicity of individual amino acids was found to have a weak correlation ($R^2 = 0.49$) with sorption of nonylphenol hexaethoxylate, a nonionic surfactant, during investigations that used thin layer chromatography (130). However, this does not explain the strong binding affinity of this compound with proteins (35,36,131-133). The binding of nonionic surfactants and proteins is assumed to involve more than one amino acid functional group (130) as well as hydrophobic forces.

Dunigan and McIntosh (134) studied sorption of atrazine on a model protein (egg albumin) and theorized that the high binding affinity was due to a number of functional groups on the protein. Amide groups have been shown to influence sorption behavior of aromatic groups and may therefore be important in sorption of specific organic micropollutants (135). Protein-associated nitrogen comprised the majority of total nitrogen in the influent wastewater and activated sludge of a domestic treatment facility (136). Amide groups were hypothesized to be protected from biological and chemical degradation within complex, highly cross-linked organic structures (136).

*Humic and Fulvic Acids***.** The most studied and reactive fractions in organic matter are the humic and fulvic acids (137). The number and types of functional groups present, including members of the carboxyl and phenolic families, diversify these organic polyelectrolytes. As a result, hydroxybenzenecarboxylates have been used as models for organic matter (138). Fulvic acids typically have more carboxylic and slightly more phenolic carbon than humic acids (139). Carboxylate groups are believed to be responsible for sorption at circumneutral and low pH with phenolate groups dominating at high pH (140). The surface chemistry of humic and fulvic acids are controlled by the quantity and number of functional groups with the physical properties varying with ambient conditions such as pH and ionic strength (141). Proton binding (charging behavior) has been shown to be a function of type and size of acid. The total concentration of proton binding sites is greatest in fulvic acid and has an inverse relationship with molecular size in humic acids (139). The high charge density, as well as the small size and lack of void space, can partially explain the relatively small binding affinity of organic pollutants in fulvic species (137).

Nanny and Maza (142) explored the binding affinity of two monoaromatic hydrocarbons and several humic acids with varying aromaticity. They concluded that the distribution coefficient for both benzene and pyridine increased with an increase in humic acid aromatic content. However, they also found that the nature of each interaction was different. While both benzene and pyridine interactions increased at decreasing pH, the bonding affinity for pyridine was higher than benzene due to the lone pair of electrons on pyridine's amine group and π- π interactions between the aromatic ring of pyridine and aromatic components of humic acids (142). This was in agreement with several research groups (111,143-146), who concluded that the quantity of aromatic groups was a good predictor of binding affinity for nonionic pollutants. A strong correlation between binding affinity and aromatic carbon together with aromatic oxygen or carboxylate carbon content was also demonstrated (147). Chiou *et al.* (148) suggested that the higher partitioning of nonionic compounds to aromatic rather than aliphatic sorbents is synonymous with their higher solubility in an aromatic (benzene) vs. aliphatic (n-hexane) solvent. However, Chefetz *et al.* (149) reported a negative correlation between aromatic content of the sorbent and distribution coefficient, maintaining that aliphatic carbon-containing groups are at least as important in the binding of nonionic compounds.

*Colloidal Material***.** As defined in the previous section, colloids are defined as any material within the size range of 1 kD (approximately 1 nm (150)) and 1.5 μm. To our knowledge, there has been no investigation regarding the affinity of activated sludge-derived colloidal material for organic contaminants. However, the association of hydrophobic organic compounds (HOCs) and colloidal material derived from the natural environment has been well documented. One of the earliest investigations on the role of colloidal material and HOC sorption was by Poirrier *et al.* (151), who demonstrated that colloids from lake systems were able to concentrate C^{14}-DDT by a factor of 10^4 greater than water when compared on a mass basis. Chin and Gschwend (152) reported substantial affinity of marine-derived organic colloids for both pyrene and phenanthrene and concluded that the magnitude of the binding was dependent on the nature of the colloidal organic matter. Chin *et al*. (146) suggested that the affinity of humic acid for pyrene was directly related to the aromatic content and molecular weight of the humic acid. Additionally, the origin of colloidal material plays a large role in the affinity for HOCs (111,153-158). Similarly, White *et al.* (159) reported that a major fraction of HOCs with K_{ow} coefficients greater than 10^4 can be bound to colloidal material.

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Chapter 3. Estrogen Receptor Agonist Fate during Wastewater and Biosolids Treatment Processes: A Mass Balance Analysis

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Chapter 4. Fluorescence Quenching of Pyrene by Colloidal Organic Carbon From Activated Sludge Systems

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ABSTRACT

The fate of hydrophobic organic compounds (HOCs) is strongly influenced by their ability to bind to organic material. The partitioning behavior of pyrene for different size fractions of colloidal organic carbon (COC) originating from two biological wastewater treatment facilities (a full-scale activated sludge system (CAS) and membrane bioreactor (MBR)) was investigated by fluorescence quenching. Fluorescence lifetime measurements demonstrated a measurable dynamic quenching component in all samples, including the colloidal-free permeates. COC partition coefficients (K_{coc}) for pyrene ranged from < 1 to 80×10^3 L/kg_{coc} and were comparable to values obtained in the literature for NOM. The aromatic content of COC was quantified by the molar extinction coefficient at 280 nm (e_{280}). A strong correlation was observed between the aromatic content and K_{coc} coefficient for the FSAS samples but was much weaker for the MBR samples, suggesting an important difference in the COC from the two treatment systems. Subsequently, e_{280} coefficients may be a useful tool for predicting colloidal transport of HOCs at FSAS facilities. The removal of COC from treated effluents may significantly reduce the concentrations of HOCs to the receiving streams.

Introduction

The ultimate fate of hydrophobic organic compounds (HOCs) in the environment is highly dependent upon the nature and concentration of organic carbon (OC) available for sorption. In aqueous solutions, sorption is controlled primarily by the partitioning (hydrophobic interactions) of HOCs into OC, since adsorption by inorganic materials (i.e., minerals) is suppressed by competing reactions with water (1,2). It is well established that HOC partitioning to OC depends on physical and chemical properties of both components, including the hydrophobicity and water solubility of the HOC (3-5) and the structure and composition of the OC (6-10).

Colloids often constitute a relatively small fraction of the total waterborne particle mass (< 10%) (11) but possess large surface areas that facilitate covalent, electrostatic and/or hydrophobic binding of other colloids and dissolved species. Partitioning of HOCs to colloids has received particular interest in natural systems since increased transport and decreased toxicity of HOCs have been observed in both field and laboratory investigations (12-16). OC that passes through filters with pore sizes of 1.5 µm but are retained by a 1,000 dalton ultrafilter (1 kD) are referred to colloidal organic carbon (COC) in this paper.

Although the importance of HOC-colloid partitioning has been documented in the natural environment (6,14,17-19), to our knowledge no researchers have investigated the partioning behavior of colloids originating from biological wastewater treatment systems. Removal of colloids from wastewater effluents, once an afterthought for operators and engineers alike, has become a goal for many advanced wastewater treatment (AWT) processes that are being used to subsidize agricultural, industrial, and potable water demands. Colloid-facilitated transport of toxic compounds from wastewater treatment facilities may potentially constitute a significant public and ecosystem health risk and, therefore, warrants investigation.

One technique that has been used to quantify colloid and fluorescent HOC partition coefficients (K_{coc}) is fluorescence quenching (FQ) (6,9,10,20,21). This method relies on a measurable decrease in fluorescence intensity once the fluorophore becomes associated with the COC (i.e., static quenching). The advantages of this method include high precision and reproducibility (10,21) and minimal artifact introduction caused by changes to the colloid structure (i.e., fast equilibration times). However, FQ is susceptible to dynamic quenching, which occurs when non-static quenchers such as dissolved oxygen (22) and other non-colloidal material (6,14) contact the fluorophore during excitation and cause a loss of fluorescence intensity (23). The most definite method of distinguishing between static and dynamic quenching components is measuring the HOC's fluorescence lifetime as a function of COC concentration (23). If quenching is primarily static, the ratio of the fluorescence lifetime in colloid-free and colloid-present aqueous solutions should not be statistically different than zero (24,25).

Most colloid partitioning studies have used commercially available or isolated/purified organic matter to understand the behavior of polycyclic aromatic hydrocarbons (PAHs) in natural systems (9,12,21,22,24-27). However, the structure and composition of wastewater-derived COC may be different compared to natural organic matter (NOM) since they are generated from different sources (28). For example, the majority of effluent COC is believed to originate from soluble microbial products (SMPs) (29) while NOM is often derived from terrestrial sources. Consequently, the OC fractions of NOM and wastewater-derived COC may be uniquely different (30).

In this study, the partitioning between wastewater-derived COC and pyrene, a fluorescent and environmentally relevant pollutant that is often used to compare the sorption capacity of different sorbents for HOCs, was investigated. The partitioning behavior between pyrene and non-settleable organic material from two wastewater treatment facilities was quantified by FQ. Fluorescence lifetime measurements were conducted to distinguish between quenching mechanisms and UV-spectroscopy was used to provide quantitative measurements of colloid aromaticity. The objectives of this study were: (1) to determine if fluorescence quenching can be used to study the partitioning behavior of colloids originating from biological wastewater treatment facilities; (2) to quantify K_{coc} for different size fractions of wastewater-derived COC; and (3) to compare the results for pyrene found in this study with other investigations using naturally- or commercially-derived COC.

Materials and Methods

Chemicals. Both pyrene (Aldrich, 97% pure) and sodium azide (Aldrich, 98% pure) were used without further purification. A 20 mg/L stock solution of pyrene was prepared in methanol and stored in an amber bottle. A 30 g/L aqueous solution of sodium azide was prepared and used in subsequent experiments.

Sample Collection, Preparation, and Filtration Protocols. A total of three separate experiments were performed. Two experiments (June and August I 2002) used samples from a full-scale activated sludge (FSAS) system while the third experiment (August II 2002) used samples from a pilot-scale membrane bioreactor (MBR). Process configurations and operational details of the MBR and FSAS systems can be found elsewhere (31). Briefly, a ZW-10 ZeeWeed® unit (Zenon Environmental, Inc., Burlington, Canada) was used for the MBR system. This membrane unit has a nominal pore size of 0.04 µm and is comprised of an inner reinforcing structure covered with a non-ionic, hydrophilic polymeric material. The membrane is approximately 0.8 m long and provides 0.93 m^2 of surface area, has a permeate header located at the top of the unit, and is continuously aerated at the base to provide turbulence at the fiber surface. The FSAS system contained both anoxic and aerobic zones to facilitate total nitrogen removal. For each experiment, approximately 5 L of mixed liquor suspended solids (MLSS) were collected in a glass bottle from the aerobic zone of the specific treatment system and allowed to settle for 1 hour. The resulting supernatant was filtered through pre-combusted 1.5 µm glass fiber filters (Whatman 934-AH). Aliquots of the < 1.5 µm filtrate were used for further size fractionation by employing 0.22 µm cellulose nitrate disc filters (Fisher) and ultrafilter membranes in parallel.

The 0.22 µm disc filters were conditioned with approximately 10 mL of < 1.5 µm filtrate prior to sample collection. Amicon YM100, YM30 and YM1 ultrafilters with nominal molecular weight unit cut-offs (MWCO) of 100, 30, and 1kD, respectively, were used for this experiment. Prior to using the ultrafilters, each membrane was soaked overnight in distilled water to remove the glycerine preservative added by the manufacturer. Each membrane was then flushed with 200 mL of Milli-Q water. All ultrafiltration experiments were conducted in magnetically stirred batch cells (200 mL volume), pressurized with nitrogen gas (350 kPa), stirred at 300 rpm, and operated at room temperature (23 ± 1° C).

For both August 2002 experiments, approximately 140 mL of each size fraction were collected (as filtrate or permeate) and used in the following experiments. For the June 2002 experiment, the filtrates/permeates from each colloid size fraction sample (except the < 1 kD permeate) were used as the feed source for a 1 kD ultrafilter. The June samples were concentrated by a factor of 2 to 3 and both retentate (1 kD to 1.5 µm, 1 kD to 0.22 µm, 1 to 100 kD and 1 to 30 kD) and < 1 kD permeate from each reactor was separately collected and used in the colloid partitioning experiments described below.

All FQ experiments were conducted within 12 hours of sample collection to minimize polymerization of the organic material. Sodium azide was added immediately after filtration was completed at a final concentration of 100 mg/L in all collected samples to inhibit microbial activity.

Fluorescence Quenching Measurements. Fluorescent quenching (FQ) experiments were conducted according to Gustaffson *et al.* (10). Borosilicate glass tubes (15 mL) containing samples of each COC filtrate were diluted, in parallel, with < 1 kDa permeate to yield a series of five to seven solutions of varying carbon concentrations (F_{spike}). The pyrene stock solution was used to spike individual tubes to final concentrations less than 30% of the maximum aqueous solubility (0.135 mg/L (17)), and the fluorescence intensity (FI) was recorded (F_{spike}) The final concentration of methanol was below 0.5% (v/v) and not expected to interfere with pyrene partitioning behavior (14). Solutions of equal carbon concentrations that were not spiked with pyrene but received equal concentrations of methanol were used to measure the background FI (F_{back}). The same procedure was followed using only < 1 kDa permeate to determine the fluorescence intensity of pyrene in the absence of organic colloids ($F_{o,spike}$ and $F_{o,back}$). All tubes were capped with aluminum foil, rotated end-over end at 30 rpm for 2 minutes, and allowed to equilibrate in the dark for an additional 20 minutes before measuring the fluorescence intensity. Preliminary results found that apparent equilibrium was reached within 5 minutes (data not shown). FI measurements were performed on a Perkin-Elmer 650-10S fluorescence spectrophotometer using a 1-cm quartz cuvette. The excitation/emission wavelengths were 334/394 and a 5 nm band-pass filter was used for both excitation/emission beams. Absorbance measurements were made at 334 and 394 nm and used to correct the fluorescence measurements for both primary and secondary inner filter effects (IFE) (20). Fluorescence intensity measurements were adjusted for background interference by the following equations:

$$F_o = (F_{o,spike})_{\text{IFE corrected}} - (F_{o,back})_{\text{IFE corrected}} \qquad (1)$$

$$F = (F_{spike})_{\text{IFE corrected}} - (F_{back})_{\text{IFE corrected}} \qquad (2)$$

Stern-Volmer (S-V) plots were constructed from the F_o/F ratios and the colloid organic concentration of the different solutions. COC binding coefficients (K_{coc}) were calculated from the best-fit slope of the S-V plots from linear regression. Statistically significant partitioning was evaluated from the p-value of the slope from the linear regression and was considered significantly different than zero if $p < 0.05$. Each experiment was conducted with a minimum of duplicate samples.

Fluorescence Lifetime Measurements. Fluorescence decay curves for the August I and II 2002 samples were obtained for both pyrene-free and pyrene-spiked solutions (prepared in the same manner as previously described) containing different size fractions and concentrations of COC using a time-correlated single-photon counting instrument located at the National Institute of Standards and Technology, Gaithersburg, MD. A GL-3300 nitrogen laser (337 nm, 50 ps pulse duration) (Photon Technology International, Inc., Canada) was used and fluorescence emission was recorded using a photomultiplier tube and charge-coupled device detector. Non-linear regression parameters obtained for samples without pyrene using a single, two-parameter exponential decay model were used to determine the fluorescence decay contribution from COC. These parameters were then held constant in a double, four-parameter non-linear regression model that was subsequently fitted to the fluorescence decay data obtained for samples containing pyrene, both in the absence and presence of COC. This method considers the contributions of background fluorescence decay from COC in calculating the fluorescence lifetime of pyrene. All non-linear regression parameters were obtained using SigmaPlot (Version 8.0, Chicago, IL).

Fluorescence lifetime measurements were conducted within 48 hours of sample collection due to unavoidable travel time from the sample location. A portion of the colloidal material may have undergone polymerization within this time frame but should not impact the efficiency or role of dynamic quenchers. Each experiment was conducted with duplicate samples.

Organic Carbon Measurements. Aliquots from each sample were acidified with H_3PO_4 and bubbled with nitrogen gas to remove inorganic carbon prior to quantification (Sievers 800 TOC Analyzer). Each sample was run in triplicate and the arithmetic average was used in subsequent calculations.

DOC, COC, and Molar Extinction Coefficient Calculations. Dissolved organic carbon (DOC) is defined as any organic carbon material that can pass through a 1 kD ultrafilter. COC concentrations were calculated by subtracting the organic carbon concentration contained in the < 1 kD permeate from the organic carbon concentration measured in each specific size fraction ($COC = OC_{size\ fraction} - DOC_{<1\ kD}$). Molar extinction coefficients (e_{280}) for each size fraction were calculated by dividing the absorbance at 280 nm by the COC concentration.

Results and Discussion

Fluorescence Lifetime Measurements. Fluorescence lifetime measurements were conducted to quantify the extent of dynamic quenching with August I (FSAS) and II (MBR) 2002 samples, which are described in detail in the following section. Samples containing only COC revealed a fairly constant fluorescence lifetime component of 9.0 ± 0.4 and 8.9 ± 0.2 ns for the FSAS and MBR samples. Surprisingly, the dissolved phase (< 1 kDa permeate) had a similar background fluorescence lifetime component (8.5 ± 0.5 and 8.3 ± 0.3 ns), suggesting the presence of humic or fulvic material in all samples (32). The presence of humic or fulvic-like material in the dissolved phase would not be expected to contribute to HOC partitioning because of physical size limitations; the organic material must be sufficiently large enough to accommodate a HOC molecule (10,19).

For the FSAS and MBR dissolved samples, the fluorescence lifetime of pyrene decreased with increasing dilutions of COC-free permeate (< 1 kD) indicating dynamic quenching (Figure 4-1). The lifetime values ranged between 74 to 98 and 82 to 92 ns for the two samples, well below the air-saturated value reported by Danielsen *et al.* (25) of 144 ns. It is well established that pyrene is dynamically quenched by oxygen (22,23,25), nitrated compounds (33) and nitrite (34) with the decrease in fluorescence lifetime calculated by:

$$\tau_o/\tau = (1 + k_o\ \tau_o[O_2])\ (1 + k_{no3}\ \tau_o[NO_3^-])(1 + k_{no2}\ \tau_o[NO_2^-]) \qquad (3)$$

where τ and τ_o are the fluorescence lifetimes in the absence and presence of a dynamic quencher, respectively (35), and k_o, k_{no3}, and k_{no2} are the diffusion rates of oxygen, nitrate, and nitrite, respectively. Both wastewater treatment facilities were operated to achieve nitrification and samples contained nitrate $[NO_3^-]$ and nitrite $[NO_2^-]$

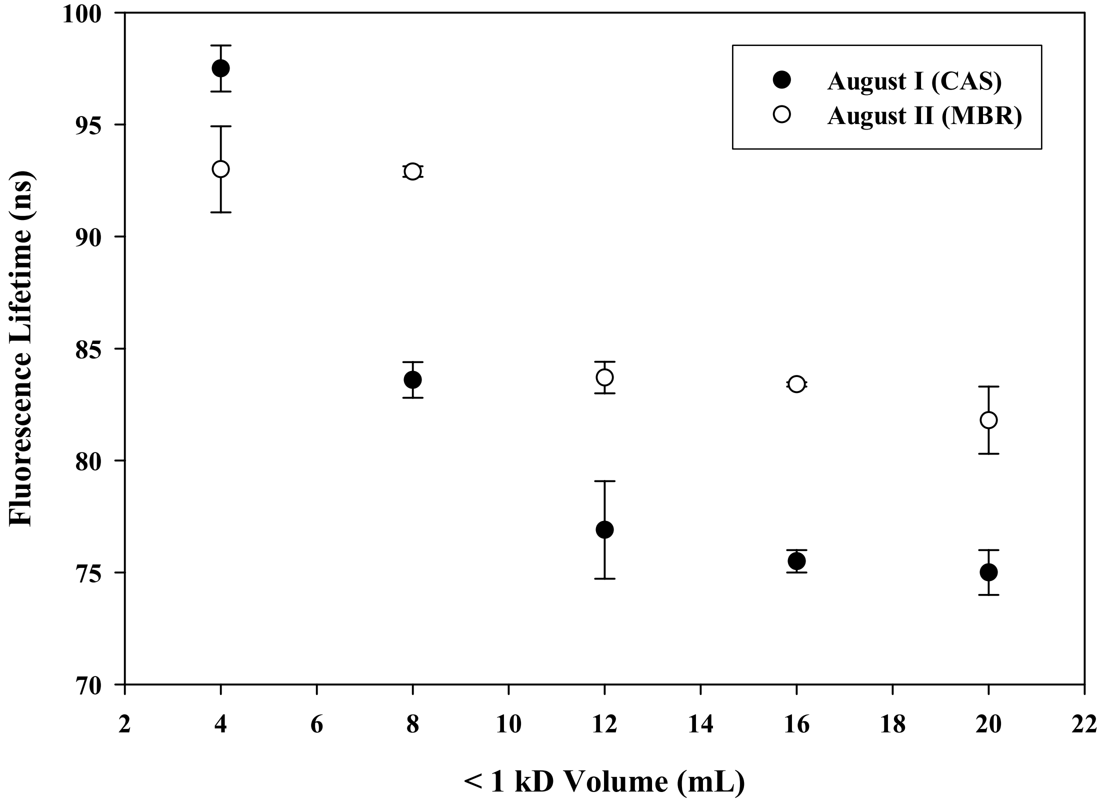

Figure 4-1: Fluorescence lifetime of colloid-free (< 1 kD) permeate from August I and II samples. Error bars are ranges of duplicate measurements.

concentrations of approximately 190 and 10 μM, respectively (FSAS full-scale data). Using equation 3 with $\tau_o = 190$ ns (25), $k_o = 1 \times 10^{10}\ M^{-1}\ S^{-1}$ (35), $[O_2] = 266\ \mu M$, $k_{no3} = 5 \times 10^9\ M^{-1}\ S^{-1}$ (33), and $k_{no2} = 5.7 \times 10^9\ M^{-1}\ S^{-1}$ (34), τ is calculated to be 105 ns, which is slightly higher than the experimental values. The small difference between calculated and experimentally derived τ values suggest that dissolved oxygen and inorganic nitrogen species constitute the major source for dynamic quenching, but that additional dynamic quenching compounds may have been present in solution. For example, inorganic anions, halogenated compounds, and amines have been shown to reduce the fluorescence intensity of PAHs through dynamic quenching (36). Many of these compounds are likely to be present in treated wastewater.

Similar to the dissolved phase, there was a decrease in τ values with an increase in COC concentration (data not shown). However, τ values decreased at the same rate as τ_o so that the τ_o/τ ratio vs. COC concentration yielded slopes indistinguishable from zero (p

> 0.10) for all size fractions (e.g., Figure 4-2). Therefore, any loss of fluorescence intensity observed during addition of COC can be attributed to static quenching since the dynamic quenching component was statistically equivalent in all samples. Consequently, colloid partitioning coefficients (K_{coc}) for pyrene can be determined with the linear Stern-Volmer equation.

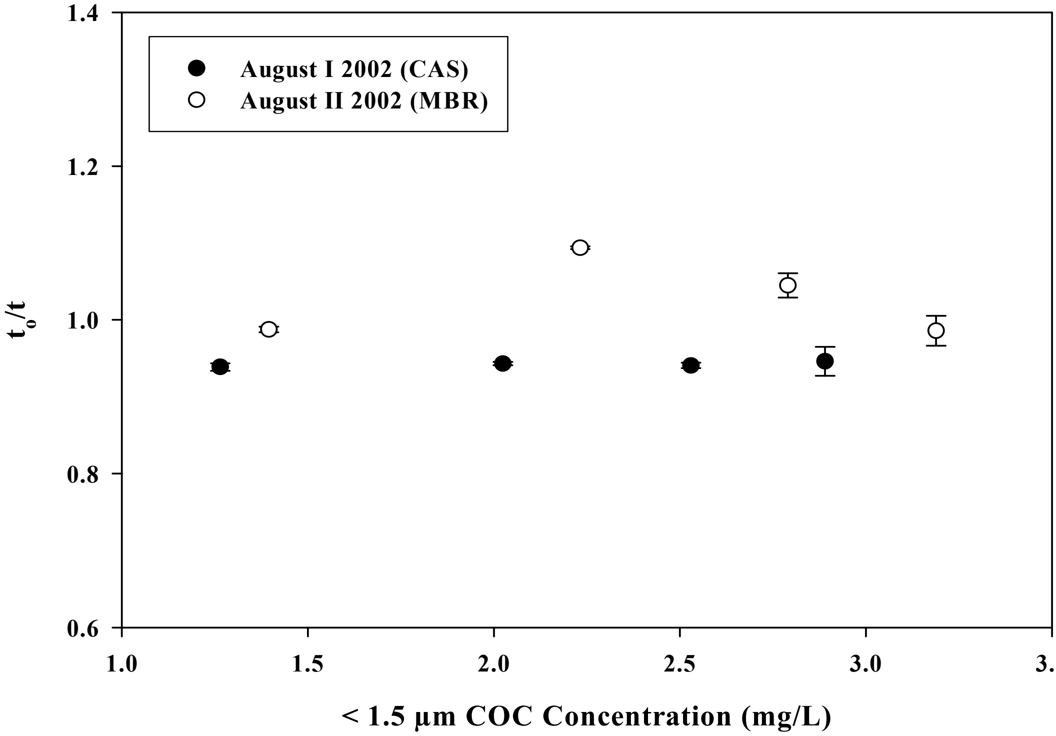

Figure 4-2: Fluorescence lifetime ratios for pyrene. Ratio are measurements made in both colloid free permeate (τ_o) and < 1.5 µm COC solutions (τ). Error bars are ranges of duplicate measurements.

The results of the fluorescence lifetime data stress the need for using the dissolved phase (< 1 kD) as the diluent in all FQ experiments involving colloids from biological wastewater treatment facilities. Failure to include the significant dynamic quenching component of the dissolved phase in all samples may cause significant overestimation of K_{coc} coefficients. Additionally, identification of all potential dynamic quenchers (i.e., oxygen, nitrate, etc.) is not necessary if these compounds are present in equal concentrations in all samples. However, if the dynamic quenchers are not truly dissolved, and therefore do not have equal concentrations in each sample, the τ_o/τ ratio vs. COC concentration would be expected to be statistically greater than zero for some size fractions indicating corrections for dynamic quenching are necessary prior to calculating K_{coc} coefficients.

Size Fractionation. COC concentrations for the concentrated samples (June 2002) ranged from 7.5 – 10.1 mg/L and the DOC concentration had an average concentration of 1.9 mg/L (Table 4-1). The unconcentrated OC concentrations from settled FSAS and settled MBR samples (August I and II 2002) were fairly similar, ranging from 2.4 to 7.7 and 2.2 to 7.8 mg/L, respectively for the different size fractions. The August I and II 2002 samples displayed a similar OC distribution although the average %OC within a specific COC size fraction was typically greater in the MBR system when compared to the FSAS (Figure 4-3). Both samples displayed a similar COC distribution with minimum concentrations between 30 and 100 kD and maximum OC (COC + DOC) concentrations in the < 30 kD fractions. By comparison, the literature contains a broad spectrum of molecular weight (MW) distributions of OC in treated effluents (29,37,38) although bimodal distribution is commonly reported (39-41).

Table 4-1: **Summary of OC, e 280 coefficients, and pyrene K_{coc} for the different size fractions and samples.**

Sample Date	Size Fraction	OC (mg/L)	e280 [a] (L/mole - cm)	K_{coc} [e] (10^3 L/kg_{coc})	Stern-Volmer y-intercept [e]	Stern-Volmer r^2
June 2002	1 kD to 1.5 µm	10.1	220	31.9 (3)	0.96 (0.01)	0.96
(FSAS)	1 kD to 0.22 µm	7.6	243	35.8 (3)	0.91 (0.01)	0.97
	1 to 100 kD	9.4	202	25 (6)	1.1 (0.01)	0.84
	1 to 30 kD	7.5	194	5.3 (3)	1.02 (0.01)	0.99
	< 1 kD [c]	1.9	166 [b]	na	na	na
August I 2002	< 1.5 µm	7.7	250	80.2 (4)	1.0 (0.01)	0.99
(FSAS)	< 0.22 µm	7.5	224	21.6 (3)	0.99 (0.01)	0.94
	< 100 kD	5.9	265	< 1 [d]	na	na
	< 30 kD	5.3	302	< 1 [d]	na	na
	< 1 kD	2.4	156 [b]	na	na	na
August II 2002	< 1.5 µm	7.8	221	29.2 (5)	1.04 (0.01)	0.92
(MBR)	< 0.22 µm	7.1	192	7.4 (1)	1.0 (0.03)	0.93
	< 100 kD	5.4	312	23.1 (5)	0.99 (0.01)	0.88
	< 30 kD	5.1	329	< 1 [d]	na	na
	< 1 kD	2.2	194 [b]	na	na	na

[a] Normalized to COC unless otherwise specified. [b] Normalized to DOC. [c] Average of two samples. [d] Not significantly different than zero ($p > 0.05$). [e] Values in parenthesis represent standard error.

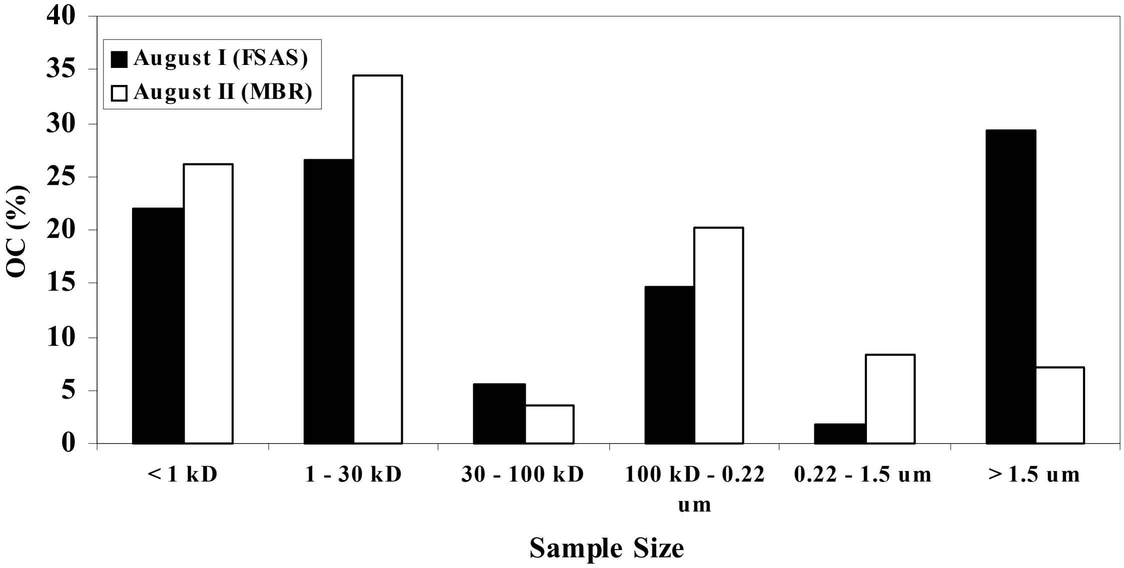

Figure 4-3: OC distribution of August I (FSAS) and II (MBR) samples.

COC-normalized molar extinction coefficients at 280 nm (e_{280}) ranged from 166 to 243 L/mole-cm for the concentrated samples (June 2002) and from 156 to 329 L/mole-cm for the filtrate samples (August I and II 2002) (Table 4-1). Several investigators have determined that e_{280} provides a good estimate of aromatic content (9,18,42) since e_{280} coefficients correspond to more rigorous methods like ^{13}C-NMR. The e_{280} coefficients of this study are within the range reported for freshwater (43) and marine (10) colloids. Interestingly, e_{280} values for the June 2002 colloid suspension samples decreased after becoming concentrated by the < 1 kD ultrafilter suggesting that the DOC in these samples has a relatively high absorption coefficient at 280 nm. Amy *et al.* (40) observed a similar relationship between molecular weight and absorbance at 254 nm for wastewater effluents, demonstrating that greater than 50% of the absorbing material was less than 5 kD.

In the August I and II 2002 samples, colloidal size was inversely related to e_{280} coefficients suggesting that smaller COC fractions (< 30 and <100 kD) possess a higher aromatic content compared to the larger size fractions. For example, the e_{280} coefficients for the < 30 and < 100 kD CAS samples averaged 283 L/mole-cm while the < 0.22 and < 1.5 μm samples averaged 237 L/mole-cm. Similar results were reported for fractionated aerobic sediment pore water (44) and isolated humic acids (9) but are in contrast with results presented by Chin *et al.* (43) who reported a direct correlation between aromaticity and molecular weight. Discrepancies in aromatic content and molecular weight may be caused by variations in microbial degradation intensity and/or the concentration of recalcitrant organic material. Microbial activity promotes the degradation of non-recalcitrant organic material while concomitantly developing structures with aromatic cores (9). Wastewater sludges have a higher active biomass population compared to soil (45), and consequently, the production of low MW, comparatively high aromatic content colloids could be expected.

Partition Coefficient. Partition coefficients between colloidal organic carbon and pyrene (K_{coc}) were calculated using the linear form of the Stern-Volmer equation (21,23):

$$F_o/F = 1 + K_{coc}[COC] \qquad (4)$$

because fluorescence lifetime experiments demonstrated that decreases in fluorescence intensity were mainly static in nature. Following IFE corrections, Stern-Volmer graphs were constructed by plotting the fluorescence intensity ratios (F_o/F) as a function of COC concentration (Figure 4-4). The best-fit slope of the resulting line was used to calculate

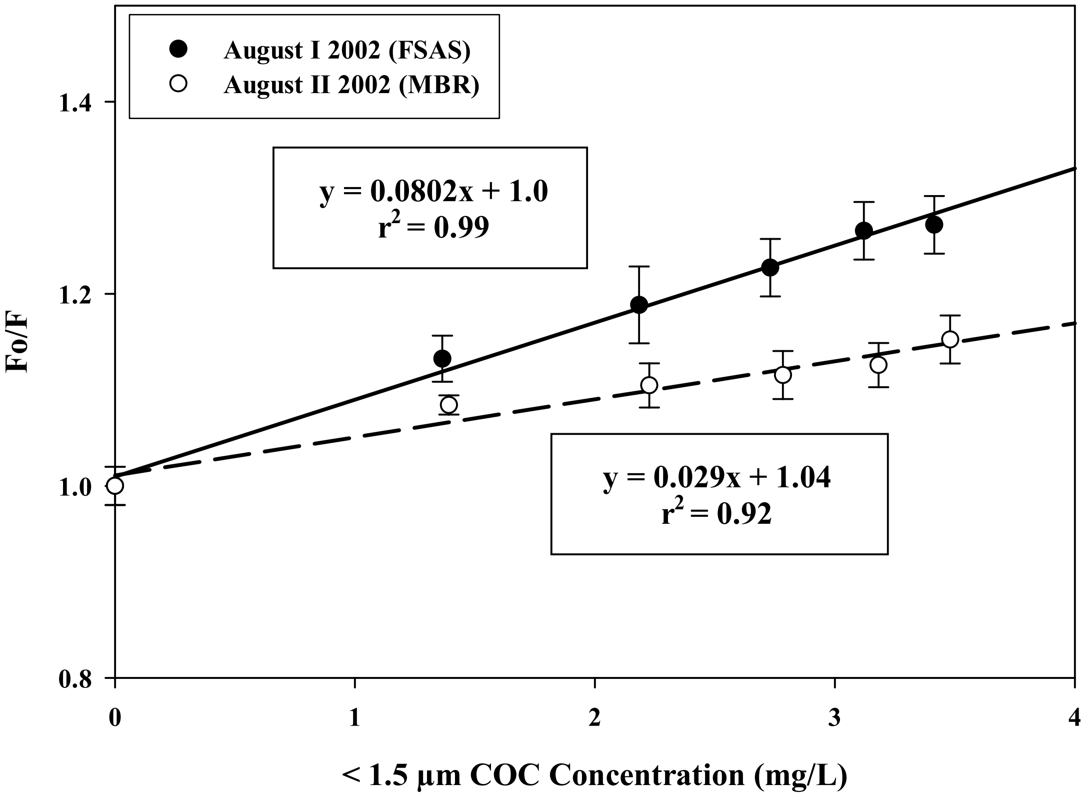

Figure 4-4: IFE-corrected Stern-Volmer plot of FQ experiments of pyrene with < 1.5 µm filtrates from the August I 2002 (FSAS) and August II 2002 (MBR) samples.

K_{coc} for each size fraction (Table 4-1). The results of the linear regressions were good, with the y-intercepts near 1.0 and strong linear correlations ($r^2 > 0.90$), and indicate that the linear Stern-Volmer equation is an appropriate analysis technique for the data collected in this study. Moreover, these results support the fluorescence lifetime analysis, which demonstrated that static quenching between COC and pyrene is the dominant mechanism for the observed decrease in fluorescence intensity.

Pyrene K_{coc} values ranged from $< 1 \times 10^3$ L/kg_{coc} to 80.2×10^3 L/kg_{coc} for the CAS samples (June and August I 2002) and $< 1 \times 10^3$ L/kg_{coc} to 29×10^3 L/kg_{coc} for the MBR samples (August II 2002) (Table 4-1). Statistically significant partitioning in the lower size fractions of the FSAS (< 100 kD) was observed only in concentrated samples (June 2002) suggesting that the smaller, unconcentrated size fractions exhibit a weaker association with pyrene compared to the concentrated samples. This observation corroborates work done by other researchers investigating lake humic substances (26) and groundwater colloids (46). Pyrene partitioning was generally higher in the filtrates of the FSAS samples when compared to the MBR samples. The one notable exception is the < 100 kD filtrate, which exhibited significantly greater pyrene sorption in the MBR system.

The importance of the MW distribution for colloid-facilitated transport of HOCs appears to be one of size vs. quantity. The data from the unconcentrated samples (August I and II 2002) suggests that the larger size fractions of COC have a larger sorption capacity compared to the smaller COC fractions. However, assuming that the concentration method did not significantly influence pyrene partitioning, the results from the concentrated samples (June 2002) imply that pyrene can appreciably partition to smaller COC material provided that the concentration of COC is sufficiently high. Subsequently, the accumulation or degradation of specific size fractions may determine the extent of HOC partitioning to wastewater-derived COC. Effluent MW distribution is related most directly to, among other parameters, substrate composition and strength and sludge age (29,37,47). For example, the degradation of model polysaccarides by both pure and mixed bacterial cultures resulted in accumulation of small MW compounds (< 1 kD) (48), while aerobic degradation of phenol resulted in the majority of effluent OC being greater than 1 kD (49). Therefore, the partitioning behavior of wastewater-derived COC may be expected to fluctuate in response to changes in operational and sewage characteristics, thereby creating a highly variable scenario with regards to HOC sorption. In full-scale systems, such fluctuations in colloidal-facilitated transport of HOCs must be dampened with a specific unit process (e.g., sand or membrane filtration) in order to be an effective barrier to organic pollutants.

The results for K_{coc} obtained in this study are within the same order of magnitude as those reported by other investigators (Figure 4-5). Isolated and purified humic and fulvic acids have the largest partition coefficients, a conclusion that should be expected from the well-documented affinity of humic and fulvic compounds for HOCs (3,50,51). Wastewater effluents have variable amounts of humic and fulvic acids (28,30,52,53). The variation in humic and fulvic acids may play an important role in HOC transport from wastewater systems, although the biotic and abiotic factors that contribute to this variation are still unknown. The K_{coc} coefficients for mixed liquor COC and pyrene measured in this study are in close agreement with the Burkhard (54) relationship between PAH partitioning and naturally-occurring dissolved organic material (DOM), assuming an octanol-water partition coefficient for pyrene of 5.18 (55). Perhaps the most interesting comparison lies between the mixed liquor-derived COC and marine colloid partitioning behavior reported by Gustafson *et al.* (10). These authors observed a decrease in pyrene sorption as the dominant COC source shifted from one of allochtonous (terrestrial run-off) to autochtonous (planktonic) inputs. The implication for biological wastewater facilities is that low-molecular weight effluent SMP produced during the activated sludge process will not have significant affinity for HOCs in aqueous solution. Moreover, facilities with a significant infiltration/inflow (I/I) contribution may inadvertently add allochtonous humic and fulvic acids to the effluent COC pool, thereby facilitating HOC partitioning. The samples obtained in this study were taken during a severe drought and are therefore thought to reflect a greater autochtonous COC contribution.

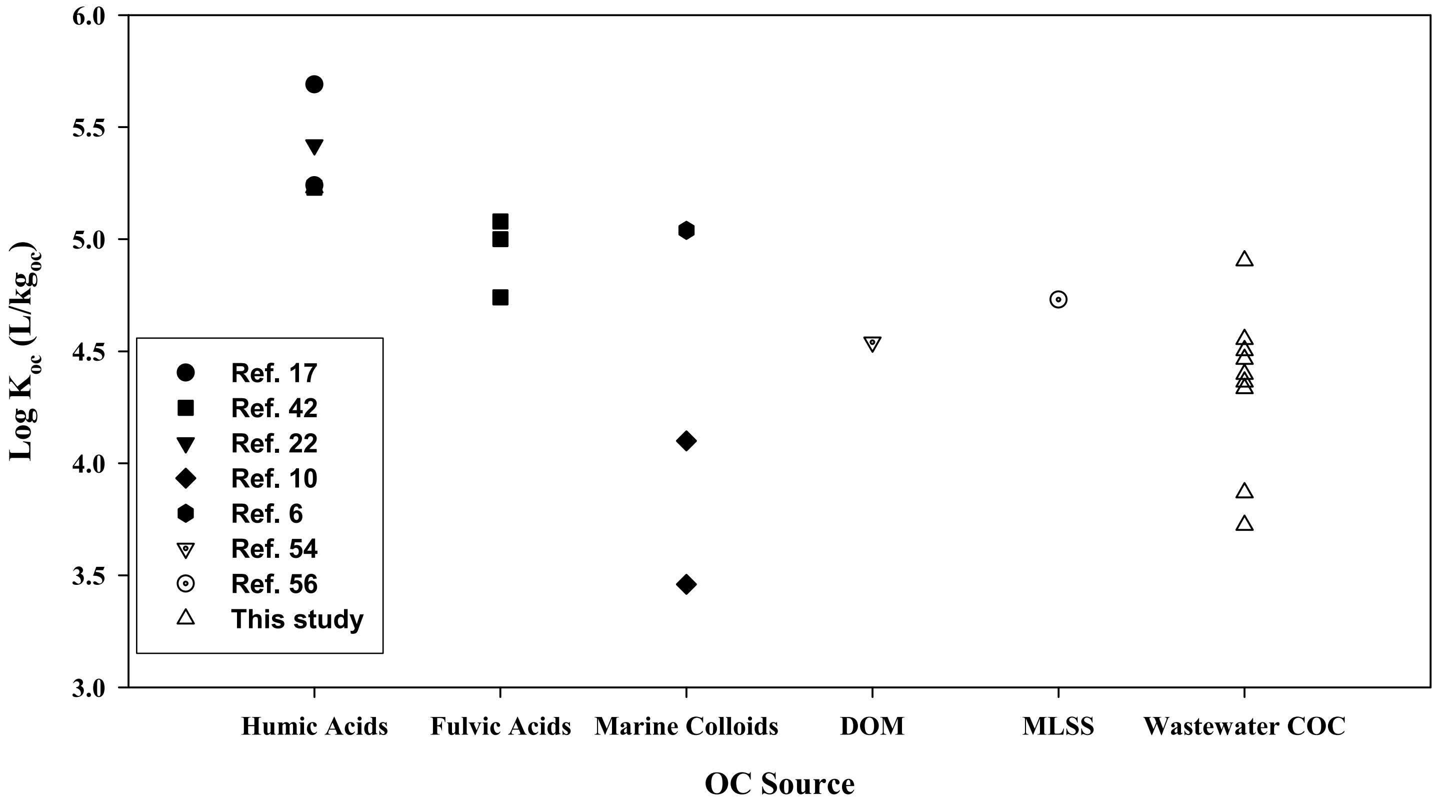

Figure 4-5: Comparison between log K_{oc} between pyrene and different colloids for this study and previous investigations.

Impact of Colloid Structure on Partitioning Behavior. The large variations in OC-normalized partition coefficients (54) have led investigators to consider additional, and possibly more critical, aspects of aquatic sorbents, which provide a better description of HOC sorption behavior. A correlation between aromatic content and partitioning behavior has been frequently reported (9,19,42). Perminova *et al.* (9) analyzed several molecular descriptors of sorption behavior and concluded that ^{13}C NMR data and H/C atomic ratios provided the best quantification for aromatic content. While these techniques may be available to investigators at large research institutions, they will not be accessible to wastewater treatment plant operators. Subsequently, Gustafson *et al*. (10) has suggested that e_{280} coefficients should be used as an alternative descriptor of aromaticity due to minimal sample preparation and easily measurable components.

A strong correlation between pyrene K_{coc} and e_{280} was found for the FSAS samples (June and August 2002) but was less predictive for the MBR samples (Figure 4-6 and Table 4-2). The results of this study have been added to the literature summary provided by Gustafson *et al*. (10) regarding similar correlations between pyrene partition and e_{280} coefficients (Table 4-2). The impact of variable e_{280} coefficients for COC samples from the FSAS system appears to be much greater than any colloidal material that has been investigated (i.e., slope of regressions, Table 4-2). However, Drewes and Croue (28) recently reported that effluent OC from a FSAS system would have between 2.6 – 3.7 times the aromatic content (as determined by ^{13}C NMR) for a given molar extinction

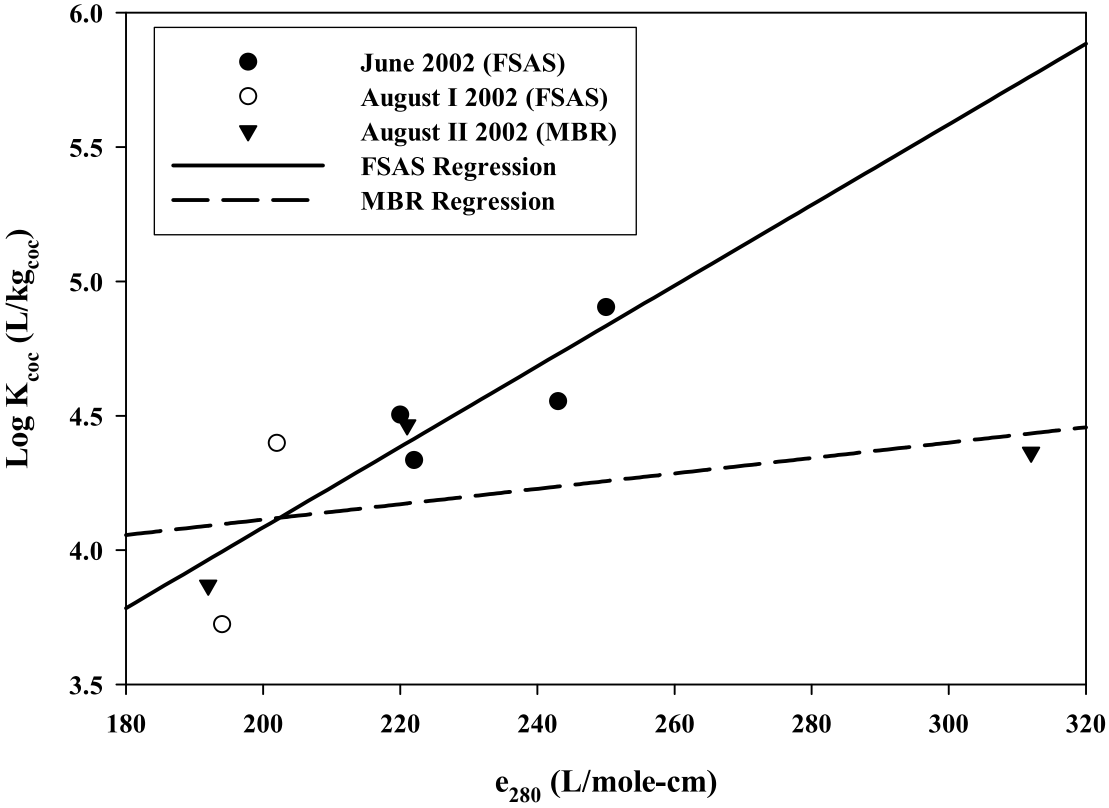

Figure 4-6: Correlation between Log K_{coc} of pyrene and molar extinction coefficient at 280 nm (e_{280}) for the different size fractions of FSAS and MBR samples.

Table 4-2: Results of correlations between Log K_{oc} and molar extinction coefficients at 280 nm (e_{280}) for this study and previous investigations [a].

Colloid Origin	Slope	Intercept	r^2	Reference
FSAS [b]	0.015	1.084	0.72	this study
MBR [b]	0.0029	2.86	0.32	this study
Marine	0.0052	2.58	0.68	(10)
HS mix	0.0018	4.52	0.53	(42)
Aquatic HA + FA	0.0015	3.48	0.79	(18)
Aquatic HA + FA	0.0019	4.44	0.38	(9)
Peat HA + FA	0.0014	4.03	0.64	(9)

[a] Other investigations summarized by (10). [b] Without correction from (28).

coefficient when compared to NOM. Consequently, applying this "correction" factor to the FSAS samples, the slope would decrease to between 0.004 – 0.0057 (L/kg / L/mole-cm), which is more consistent with the autochtonous-dominated marine colloids (10). It is not clear whether the Drewes and Croue investigation (28) can be applied to MBR systems since their dataset was based only on FSAS effluent. In any case, there is a clear difference on the impact of aromatic content and pyrene partitioning behavior between the MBR and FSAS samples. The reason for differences is neither clear nor trivial and requires further investigation. However, our results confirm that COC from the MBR system is able to partition with pyrene, suggesting that these colloids possess some unique properties compared to FSAS or NOM.

The results from this study indicate that wastewater engineers and operators can quantitatively evaluate the impact of operational and process changes for CAS systems on colloidal transport of HOCs by measuring e_{280} coefficients for effluents. Process modifications resulting in comparatively higher e_{280} values may lead to greater colloid partitioning and, subsequently, increased concentrations of potentially toxic compounds in the final effluent. On the other hand, facilities employing final effluent polishing (e.g., granular activated carbon) may observe greater removal of pollutants when they are associated with higher molecular weight colloids (41).

Environmental Significance. Our results demonstrate that a representative HOC (pyrene) is able to partition to COC from settled FSAS and MBR MLSS samples. The magnitude of partitioning appears to be dependent on the nature of the COC, specifically the molecular weight and OC distribution and aromatic content. Subsequently, wastewater-derived COC represents an important transport mechanism for delivering potentially toxic compounds from biological treatment plants to receiving waters. The percentage of bound pyrene can be estimated from the following equation (57):

$$\text{Bound pyrene (\%)} = \frac{[\mathrm{COC}](K_{coc})}{1 + [\mathrm{COC}](K_{coc})} \qquad (5)$$

From this study, approximately 30% of the aqueous pyrene will be associated with the < 1.5 μm size fraction from the FSAS system compared to 14% with the same size fraction from the MBR. In conclusion, the data suggests that colloid capture is more important for FSAS systems compared with MBR facilities due to the nature of the colloidal material. Clearly, more investigation is needed to determine which full-scale operational parameters most critically influence the nature and partitioning behavior of HOC. However, it is clear that colloid-facilitated transport of HOCs and other organic pollutants will be an important issue in the future of wastewater treatment.

We have previously reported on sorption behavior of pyrene for a selected set of activated sludge samples (58). In comparing the results obtained in the two studies, there are some noticeable differences including higher molar extinction coefficients and higher partition coefficients for pyrene for the MBR system. We believe this difference can be attributed to samples taken under different operational conditions (sludge age, food-to-microorganism ratio, etc.) and may reflect the important differences among colloidal properties for pollutant transport. Indeed, biological wastewater facilities have many complicated and uncontrollable factors that contribute to differences in effluent organic material (29).

Acknowledgements

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Chapter 5. Sorption of 17β-Estradiol and 17α-Ethinylestradiol by Colloidal Organic Carbon Derived from Biological Wastewater Treatment Systems

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ABSTRACT

Sorption coefficients (K_{coc}) of 17β-Estradiol (E2) and 17α-Ethinylestradiol (EE2) for size fractionated colloidal organic carbon (COC) derived from two biological wastewater treatment facilities was quantified by fluorescence quenching. The two wastewater treatment systems were a conventional activated sludge system (CAS) and a membrane bioreactor (MBR). The K_{coc} coefficients for E2 and EE2 were highly variable and ranged between (< 1 to 158) × 10^3 L/kg_{coc} for E2 and (< 1 to 228) × 10^3 L/kg_{coc} for EE2. The best descriptor for *a priori* estimates of E2 K_{coc} coefficients was the molar extinction coefficient at 280 nm (e_{280}), suggesting that π-electron interactions are important in determining overall binding behavior. The results of this study did not reveal any strong descriptors of colloidal affinity for EE2. A substantial portion of the aqueous E2 and EE2 concentrations (between 1 and 50%) may be associated with colloidal material indicating that colloidal transport of E2 and EE2 may be a significant route for estrogenic compounds to be discharged to the natural environment from biological wastewater treatment systems.

Introduction

The presence of both natural and synthetic estrogenic substances, or estrogenic endocrine disrupting compounds (EDCs), in treated wastewater effluents has been well documented (1-8). Although effluent concentrations of estrogenic compounds are relatively low, usually on the order of a few ng/L, their potency is sufficient enough to cause alterations in wildlife including feminization and vitellogenin induction in male fish (8-10). Researchers have identified 17β-estradiol (E2), a natural hormone, and 17α-ethinylestradiol (EE2), the synthetic hormone found in oral contraceptives, as the major contributors of effluent estrogenic activity (7,9). Therefore, a better understanding of factors that impact the fate of E2 and EE2 during the wastewater treatment process is critical to the development and implementation of suitable control strategies.

One important factor that determines the fate of organic contaminants in wastewater treatment systems is the contaminant distribution between the dissolved, colloidal, and particulate phases. Sorption of organic compounds to the colloidal phase in sewage, mixed liquor from biological reactors, and effluent is significant interest since colloidal material is often difficult to remove from the aqueous phase without advanced treatment technologies. Colloid-contaminant partitioning has been well-documented between commercially-available and naturally-derived dissolved organic matter (DOM) and nonionic contaminants such as polyaromatic hydrocarbons (PAHs) (11-17). However, effluent colloidal organic carbon (COC) may possess unique properties that either facilitate or impede the sorption of organic contaminants. Indeed, the structure and character of DOM and effluent COC appear to be different (18) probably because they are derived from different sources. Moreover, E2 and EE2 contain one aromatic and three non-aromatic rings, suggesting a unique sorption behavior compared to PAHs.

The hypothesis that E2 and EE2 will readily sorb to colloidal material is supported by the literature. Lai *et al.* (19) observed a decrease in sediment partitioning of selected estrogenic compounds over time, which was attributed to increased concentrations of DOM. Bowman *et al.* (20) determined that the distribution coefficients between estuarine colloids and E2 were two orders of magnitude higher than sediment. Finally, Yamamoto *et al.* (21) concluded that DOM surrogates have significant sorption capacity for a variety of estrogenic compounds.

Quantifying colloidal sorption coefficients (K_{coc}) is inherently difficult since judicious differentiation between truly dissolved and colloidal phases is required. In the past, many researchers simply normalized sorption coefficients to all organic carbon able to pass through a certain filter size (e.g., 0.22 μm). However, since the resulting filtrate will contain both dissolved (DOC) and colloidal organic carbon (COC), the calculated sorption coefficient (K_{doc} or K_{oc}) will be artificially low since "true" DOC will not participate in contaminant sorption (22). In determining partition coefficients between pyrene and isolated marine colloids, Gustafson *et al.* (17) utilized a molecular weight cut-off of 1,000 Da (1 kD) to define the dissolved phase, a definition consistent with thermodynamic considerations (23). In this study, we adopted this definition, and COC is herein defined as carbon between 1 kD and 1.5 μm in size while DOC is any carbon < 1 kD.

The present knowledge regarding the ability of mixed-liquor derived colloidal material to bind contaminants, including estrogenic compounds, is limited by a substantial lack of experimental data. This study attempts to fill that void by investigating the sorption behavior of E2 and EE2 to various size fractions of organic colloidal material derived from the mixed liquor of two wastewater treatment systems – a full-scale activated sludge system (FSAS) and a pilot-scale membrane bioreactor (MBR). Mixed liquor was used since the amount of effluent colloidal material from the MBR system is much lower compared to the FSAS system. The objectives of this study were to: (i) quantify sorption coefficients for E2 and EE2; and (ii) identify the key colloidal structural and/or composition features that facilitate E2 and EE2 sorption.

Materials and Methods

Sorbates. E2 (99% pure, Aldrich), and EE2 (98% pure, Aldrich), were used without further purification. Stock solutions of 1 g/L were prepared in ethanol and stored in amber bottles at 4° C to minimize evaporation.

Colloid Sampling and Characterization. Two activated sludge processes, including a FSAS system that contains anoxic and aerobic zones to achieve biological nitrogen removal, and a fully aerobic pilot-scale MBR, were used in this investigation. Process details are described elsewhere (24). The FSAS and MBR systems treated the same influent wastewater (primary effluent) and operated at similar mean cell retention times (MCRTs) (approximately 10 - 12 days for the May sampling period and 20 – 30 days for the August sampling period). The MBR utilized a ZW-10 ZeeWeed® unit

(Zenon Environmental, Inc., Burlington, Canada) with a nominal pore diameter of 0.04 μm while the FSAS unit used secondary clarifiers for liquid-solid separation. The pH of each system was 7.1 ± 0.2.

Mixed liquor suspended solids (MLSS) samples were collected on two separate (May and August 2002) occasions in glass containers and allowed to settle for 1 hour. The resulting supernatant was filtered through pre-combusted 1.5 μm glass fiber filters (Whatman 934-AH) and collected (< 1.5 μm filtrate). Aliquots of the < 1.5 μm filtrate were processed, in parallel, with 0.22 μm filters (nitrocellulose, Fisherbrand) and YM100, YM30, and YM1 ultrafiltration membranes (Amicon, Inc., Beverly, MA) which have a molecular weight cut-off of 100, 30, and 1 kDa, respectively. The 0.22 μm filters were used without any special preparation. Prior to use, the ultrafiltration membranes were soaked in deionized (DI) water for 1 hour with three water changes, overnight at 4˚C in DI water, and finally flushed with 200 mL of DI water to remove the glycerin preservative added by the manufacturer. Following conditioning, separate ultrafiltration units were operated at 400 rpm and a pressure of 55 psi to produce < 100 kDa, < 30 kDa, and < 1 kDa samples. Sodium azide was added to each sample at a final concentration of 100 mg/L to inhibit microbial activity. All samples were used within 12 hours to avoid polymerization of organic material.

Grab samples of secondary effluent from the FSAS system were also taken upstream and downstream of the chlorine contact chamber to assess the impact of the chlorination/dechlorination process on colloidal aromatic content.

Fluorescence Quenching Measurements. Fluorescent quenching (FQ) experiments were conducted according to Gustafsson *et al.* (17). Borosilicate glass tubes (15 mL) containing a known volume of each COC filtrate were diluted, in parallel, with < 1 kDa permeate to yield a series of five to seven solutions with varying organic carbon concentrations. E2 and EE2 stock solutions were used to spike these individual glass tubes to final concentrations of 1000 μg/L and the fluorescence intensity (FI) was recorded (F_{spike}). The concentrations of E2 and EE2 used in these experiments are well above those reported for raw sewage and treated effluents (e.g., (6,9,25)) to improve fluorescence detection, but are below their maximum aqueous solubility concentrations (26). The final concentration of ethanol was below 0.5% and not expected to interfere with E2 or EE2 sorption behavior (22). Solutions of equal COC concentrations that were not spiked with E2 or EE2 but received equal concentrations of ethanol were used to measure the background FI (F_{back}). The same procedure was followed using only < 1 kDa permeate to determine the FI of E2 and EE2 in both the presence and absence of COC ($F_{o,spike}$ and $F_{o,back}$, respectively). All tubes were capped with aluminum foil, rotated end-over end at 30 rpm for 2 minutes, and allowed to equilibrate in the dark for an additional 20 minutes before measuring the FI. Preliminary results showed that apparent equilibrium was reached within 5 minutes (data not shown). FI measurements were performed on a Perkin-Elmer 650-10S fluorescence spectrophotometer using a 1-cm quartz cuvette. The excitation/emission wavelengths were 285/345 and a 5 nm band-pass filter was used for both excitation/emission beams. Absorbance measurements were made at 285 and 345 nm and used to correct the fluorescence measurements for both primary and secondary inner filter effects (IFE) (27). FI measurements were adjusted for background interference by the following equations:

$$F_o = (F_{o,spike})_{\text{IFE corrected}} - (F_{o,back})_{\text{IFE corrected}} \quad (1)$$

$$F = (F_{spike})_{\text{IFE corrected}} - (F_{back})_{\text{IFE corrected}} \quad (2)$$

Stern-Volmer (S-V) plots were constructed from the F_o/F ratios and the COC concentration of the different solutions. COC sorption coefficients (K_{coc}) were calculated from the slope of the S-V plots, which were fitted using linear regression. Sorption coefficients were considered significant (i.e., statistically different that zero) if p value of the slope was less than 0.05 ($p < 0.05$). Each experiment was conducted in duplicate.

Analytical Methods. Non-volatile organic carbon concentrations were measured using a Sievers 800 TOC Analyzer. Molar extinction coefficients at 280 nm (e_{280}) of each size fraction were calculated by dividing the absorbance measurements at 280 nm by their resepective COC concentrations. Protein was measured using the Frolund *et al.* (28) modification of the Lowry *et al.* (29) method. Polysaccharides were measured using the method of Dubois *et al.* (30). Protein standards were prepared with pre-made, serially diluted bovine serum albumin, and polysaccharide standards were prepared with glucose.

For total iron, samples were acidified with nitric acid and measured by a flame atomic adsorption.

Results and Discussion

Characteristics of Effluent Organic Carbon. Organic carbon (OC), protein, polysaccharide, and iron concentrations in the 1.5 µm samples from the FSAS and MBR systems decreased with smaller molecular size fractions (Table 5-1). The majority of dissolved OC in wastewater effluents is of microbial origin (31-35), and this pool of organic matter is collectively called soluble microbial products (SMPs). SMPs have been identified as proteins, polysaccharides, humic and fulvic acids, and structural components of cells (36) and include organic material that passes through a 1.5 µm filter. A greater fraction, on average, of the organic carbon in the MBR system is attributed to protein and polysaccharide concentrations compared with the FSAS system. For example, assuming a carbon content for protein and polysaccaride of 55 and 43% by mass, respectively (37), these SMP components comprised between 26 to 62% (FSAS) and 49 to 76% (MBR) of the measured OC concentration (Figure 5-1). The data also suggest that polysaccharides are preferentially accumulated in the MBR system, averaging 30 (± 8)% of the measured OC concentrations compared with 19 (± 3)% for the FSAS system. It should be noted that characterization of effluent organic matter is a challenging task; Dignac *et al*. (38) was able to characterize only 20% of the organic carbon in treated effluent using various chromatographic techniques.

Table 5-1: Summary of Effluent Organic Carbon Characteristics.

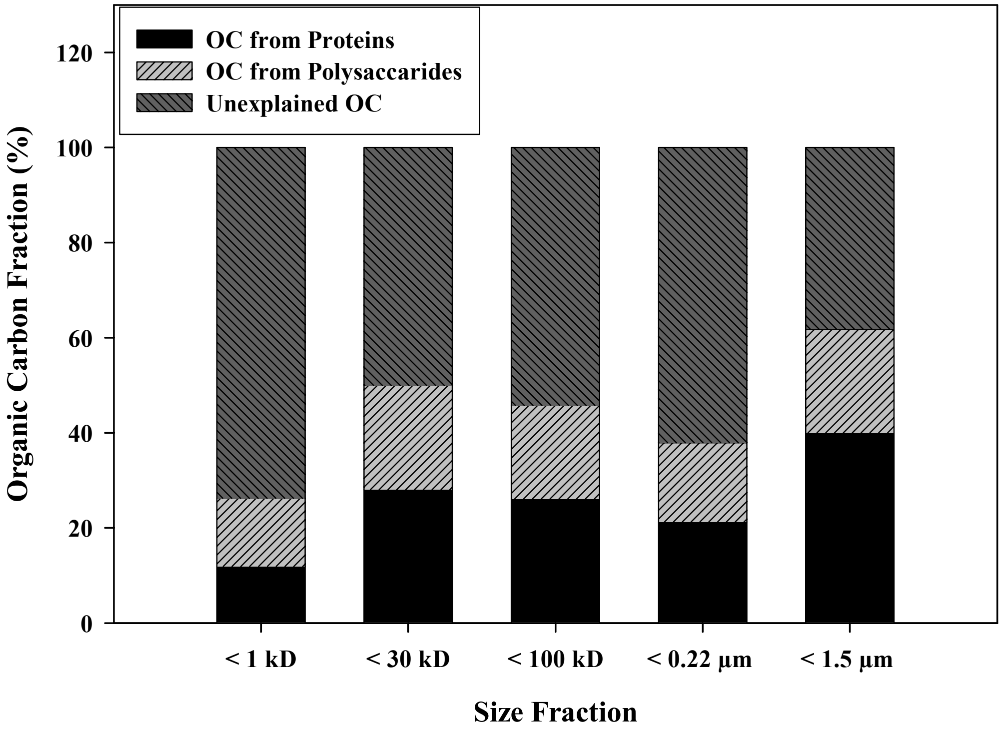
FSAS

Sample Date	Size Fraction	OC [a] (mg/L)	Protein (mg/L)	Polysaccaride (mg/L)	Iron (mg/L)	e_{280} [b] (L/mol - cm)
May 2002	< 1.5 µm	8.9	nm	nm	nm	229
	< 0.22 µm	7.7	nm	nm	nm	261
	< 30 kD	6.0	nm	nm	nm	311
	< 1 kD	2.5	nm	nm	nm	178 [c]
August 2002	< 1.5 µm	7.7	5.6	3.9	0.23	250
	< 0.22 µm	7.5	2.9	2.9	0.23	224
	< 100 kD	5.9	2.8	2.7	0.22	255
	< 30 kD	5.3	2.7	2.7	0.21	276
	< 1 kD	2.4	0.52	0.80	0.19	154 [c]

MBR

Sample Date	Size Fraction	OC[a] (mg/L)	Protein (mg BSA/L)	Polysaccaride (mg glucose/L)	Iron (mg/L)	e_{280} [b] (L/mol - cm)
May 2002	< 1.5 µm	6.20	nm	nm	nm	230
	< 0.22 µm	5.50	nm	nm	nm	334
	< 30 kD	4.90	nm	nm	nm	337
	< 1 kD	2.10	nm	nm	nm	211 [c]
August 2002	< 1.5 µm	7.8	4.9	7.8	0.14	221
	< 0.22 µm	7.1	3.4	4.6	0.13	192
	< 100 kD	5.4	2.9	3.5	0.12	312
	< 30 kD	5.1	2.7	2.4	0.12	329
	< 1 kD	2.2	1.5	1.6	0.1	194 [c]

[a] organic carbon. [b] normalized to COC. [c] normalized to DOC. nm = not measured.



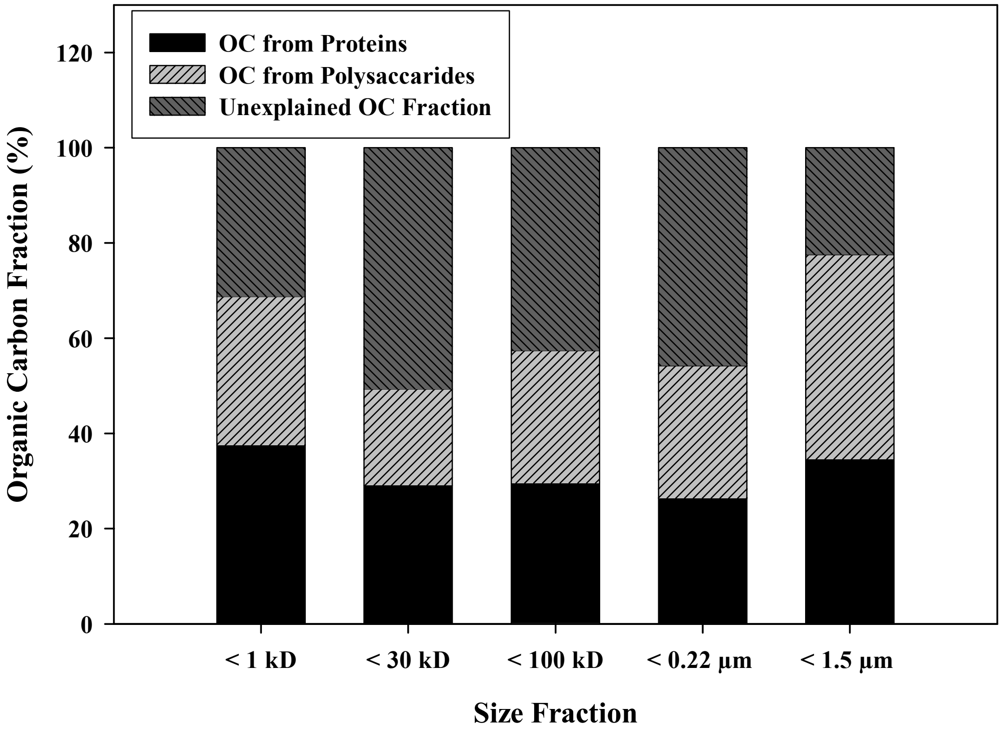


Figure 5-1: Fraction of DOC attributed to protein and polysaccaride content for the FSAS system (top) and MBR system (bottom).

The colloidal and dissolved iron concentrations in the FSAS samples were almost twice as high as the iron concentrations in the MBR samples (Table 5-1). However, the majority (approximately 72%) of iron for both systems was found in the dissolved phase. The physical location of the inorganic phase within the colloidal structure is an important factor in FQ experiments since sorption of any organic contaminant by an inorganic surface will not quench the compound's fluorescence intensity (39,40). This may result in a reduced sorption coefficient for colloidal material integrated with iron compared to colloidal material that is comprised solely of organic matter. It is currently accepted that iron precipitates are efficient scavengers of organic material in aquatic (41,42) and wastewater (43) environments and therefore are predominantly coated with organic material. However, Schlautman and Morgan (40) demonstrated that the adsorption of commercially available humic and fulvic acids onto aluminum decreased the capacity of the humic and fulvic acids to sorb perylene. Consequently, the inorganic colloidal material should be of considerable interest in any FQ investigation, and should be quantified when possible to account for variations in the affinity of aquatic colloids for organic contaminants.

The molar extinction coefficient at 280 nm (e_{280}) for the FSAS and MBR samples with colloidal material increased with decreasing size fraction, but was lowest for the samples without colloidal material (Table 5-1). Additionally, the e_{280} coefficients for the MBR samples were generally higher than the FSAS samples. Investigators have observed a correlation between molar extinction coefficients measured at 254 to 280 nm and the aromatic content of colloidal material (11,44). For example, Gauthier *et al.* (11) observed that higher e_{272} coefficients correlated strongly with aromatic content quantified by ^{13}C NMR for a group of isolated humic acids. In this study, the higher e_{280} coefficients indicate that the smaller size colloids are enriched in aromatic moieties. Similarly, Amy *et al.* (32) demonstrated maximum molar extinction coefficients (at 254 nm) for size fractionated FSAS effluent between 30 and 100 kD. Accordingly, the data presented here suggest that the colloidal material in settled mixed liquor samples has more aromatic content than the dissolved phase, and that the MBR system had slightly higher aromatic content than the FSAS.

Fluorescence Quenching and Sorption Coefficients. FQ is a useful method for determining sorption coefficients between fluorescent compounds and colloidal material (45,46). However, FQ may overestimate sorption coefficients by a factor of 2 to 3 compared to other methods, such as solubility enhancement (12). Possible artifacts causing this overestimation include sorption to glassware (22), loss from volatilization or degradation, dynamic quenching by oxygen (13,47), and static quenching by truly dissolved material. Accordingly, several preliminary experiments were conducted to insure that the measured FQ values reflected true sorption.

Sorption onto the glass tubes for E2 and EE2 resulted in a minimal fluorescence intensity loss (< 4%) after 3 hours, which was approximately six times longer than the time taken to conduct a FQ experiment. Similarly, photodegradation of E2 and EE2 in both COC solutions and nanopure at the fluorescence wavelengths used during the FQ experiments was also minimal (< 2%) after three minutes of exposure, which was approximately three times longer than the time required for routine FI measurements; therefore, sufficient analysis time was available to obtain a stable FI reading. Furthermore, biodegradation was inhibited by the addition of sodium azide. Recently, Yamamoto *et al*. (21) demonstrated that the FI of E2 was unaffected by the presence or absence of oxygen. Therefore, a decrease in FI due to sorption, photodegradation, biodegradation and oxygen were considered minimal. Furthermore, the presence of any static quenchers other than COC will be accounted for by the < 1 kD permeate (17,22,48).

The FI of E2 and EE2 decreased in direct proportion to the amount of COC present in solution and this decrease was attributed to sorption of E2 or EE2 by the COC. Additionally, the IFE correction ratios were very low for these samples, ranging between 1.05 – 1.3, indicating that uncertainties introduced by IFE calculations were minimal (17,49). Consequently, the linear form of the Stern-Volmer equation:

$$Fo/F = 1 + [COC]K_{coc} \qquad (3)$$

was used to calculate the COC sorption coefficient (K_{coc}) by calculating the slope of the best-fit line to IFE-corrected Fo/F data plotted against COC concentrations for each size fraction.

The results of the IFE-corrected Stern-Volmer plots for the FSAS and MBR samples are summarized in Table 5-2. In general, there was a fairly significant difference in K_{coc} coefficients between sampling periods (May and August 2002), and the K_{coc} coefficients for the May 2002 FSAS and MBR samples were greater than the August 2002 samples. However, there were three COC samples that did not demonstrate significant E2 or EE2 sorption. A direct comparison of the FSAS and MBR K_{coc} coefficients for the different size fractions did not reveal any obvious relationships, suggesting that the affinity of E2 and EE2 for COC derived from the two biological wastewater treatment systems is not exclusively dependent on process configuration or colloidal size.

Table 5-2: Summary of Stern-Volmer plots used to calculate K_{coc} coefficients for E2 and EE2.

Estradiol (E2)

		FSAS			MBR		
Sample Date	**Size Fraction**	**K_{coc} [a] (10^3 L/kg_{coc})**	**Stern-Volmer [a] y-intercept**	**Stern-Volmer r^2**	**K_{coc} [a] (10^3 L/kg_{coc})**	**Stern-Volmer [a] y-intercept**	**Stern-Volmer r^2**
May, 2002	< 1.5 µm	38 (13)	0.95 (0.1)	0.95	62 (7)	0.94 (0.1)	0.85
	< 0.22 µm	160 (58)	1.03 (0.1)	0.71	160 (71)	1.0 (0.1)	0.55
	< 30 kD	110 (39)	1.1 (0.1)	0.83	160 (66)	0.99 (0.1)	0.91
August, 2002	< 1.5 µm	76 (12)	0.96 (0.1)	0.93	69 (15)	0.99 (0.1)	0.84
	< 0.22 µm	87 (21)	0.83 (0.1)	0.82	25 (5)	0.94 (0.1)	0.95
	< 100 kD	33 (10)	0.99 (0.1)	0.78	< 1[b]	na	na
	< 30 kD	52 (12)	1.02 (0.1)	0.83	< 1[b]	na	na

Ethinylestradiol (EE2)

		FSAS			MBR		
Sample Date	**Size Fraction**	**K_{coc} [a] (10^3 L/kg_{coc})**	**Stern-Volmer [a] y-intercept**	**Stern-Volmer r^2**	**K_{coc} [a] (10^3 L/kg_{coc})**	**Stern-Volmer [a] y-intercept**	**Stern-Volmer r^2**
May, 2002	< 1.5 µm	68 (10)	1.01 (0.1)	0.83	89 (7)	0.88 (0.1)	0.66
	< 0.22 µm	230 (59)	1.0 (0.1)	0.66	110 (27)	1.1 (0.1)	0.66
	< 30 kD	170 (30)	0.98 (0.1)	0.84	170 (45)	1.1 (0.1)	0.63
August, 2002	< 1.5 µm	95 (28)	0.97 (0.1)	0.74	169 (25)	0.94 (0.1)	0.91
	< 0.22 µm	89 (26)	0.97 (0.1)	0.75	179 (32)	0.93(0.1)	0.89
	< 100 kD	46 (10)	0.91 (0.1)	0.72	31 (8)	1.08 (0.1)	0.83
	< 30 kD	< 1[b]	na	na	32 (9)	1.07 (0.1)	0.82

[a] values listed are averages ± standard error (in parenthesis) of duplicate experiments. [b] Fluorescence quenching not significant ($p > 0.05$).
na = not applicable.

Sorption Coefficients – Comparison of Potential Descriptors. The use of descriptors to make *a priori* estimates of the sorption behavior of organic contaminants is of considerable use in identifying and implementing proper management techniques to mitigate their impact on the natural environment. Several descriptors have been successfully used for this purpose, including octanol-water partition coefficient (K_{ow}), aromatic content of organic material, a single surrogate compound, and organic carbon composition.

K_{ow} coefficients. Several researchers have observed strong correlations between K_{oc} and K_{ow} coefficients for nonionic compounds (e.g., PAHs) (15,50-52) since partitioning is the dominant sorption mechanism. A universal K_{oc}-K_{ow} relationship for all organic material is impractical since the origin of colloidal material is an important factor in organic contaminant sorption behavior (e.g., (44,53)). However, the K_{oc} coefficient predicted by the K_{oc}-K_{ow} relationship is typically less than the K_{ow} coefficient. In contrast, the log K_{coc} coefficients determined by FQ for both E2 and EE2 were substantially greater than their respective K_{ow} coefficients (Figure 5-2) (19,54). Additionally, there is a fairly wide range in log K_{coc} coefficients for each compound. For example, the log K_{coc} coefficients for E2 range between 4.3 and 5.2 while the log K_{coc} coefficients for EE2 range between 4.5 and 5.4. Since the K_{coc} coefficients for the different samples span a relatively wide range and are greater than their respective log K_{ow} coefficients, the use of log K_{coc}-K_{ow} relationships to accurately predict the binding behavior of E2 and EE2 to MLSSderived COC is not suggested. These results are in agreement with Yamamoto *et al.* (21), who observed no relationship between K_{coc} and K_{ow} coefficients for a selected set of EDCs and NOM surrogates. However, our results appear to contradict those of Lai *et al.* (19), who demonstrated the applicability of a fugacity model in predicting E2 and EE2 sorption to riverbed sediment.

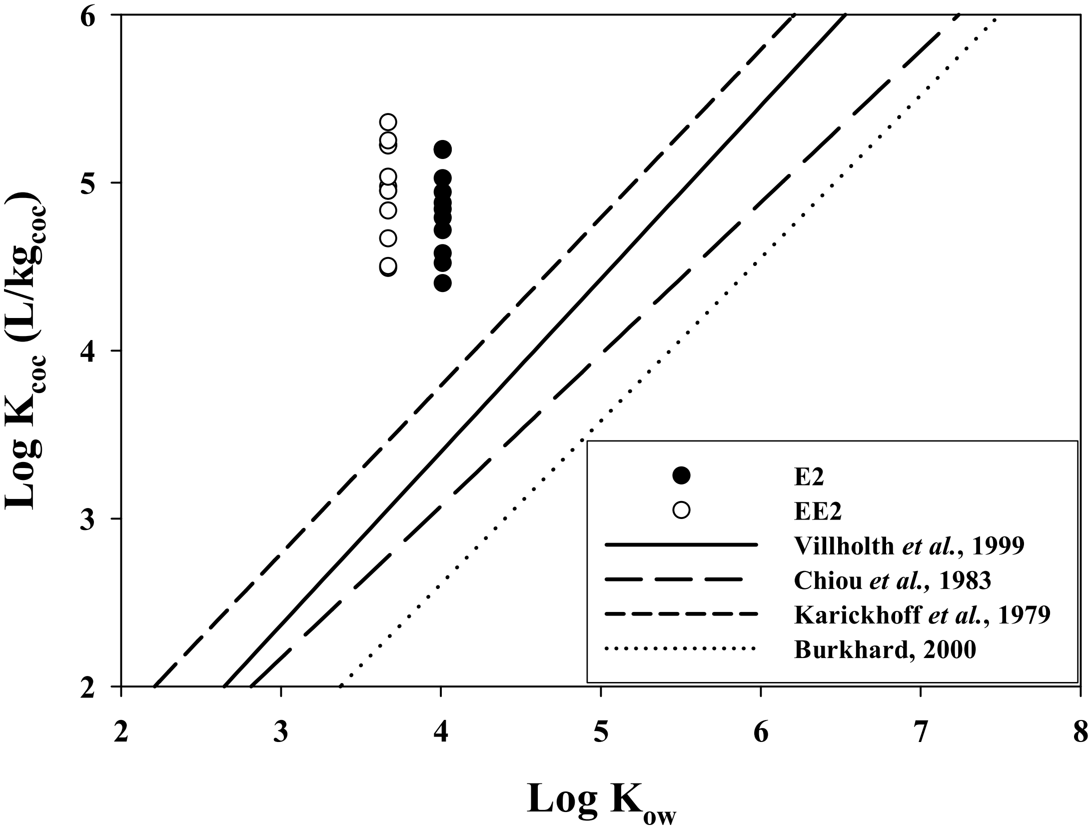


Figure 5-2: Comparison between log K_{coc} coefficients calculated from this study and log K_{ow} coefficients from the literature for E2 and EE2.

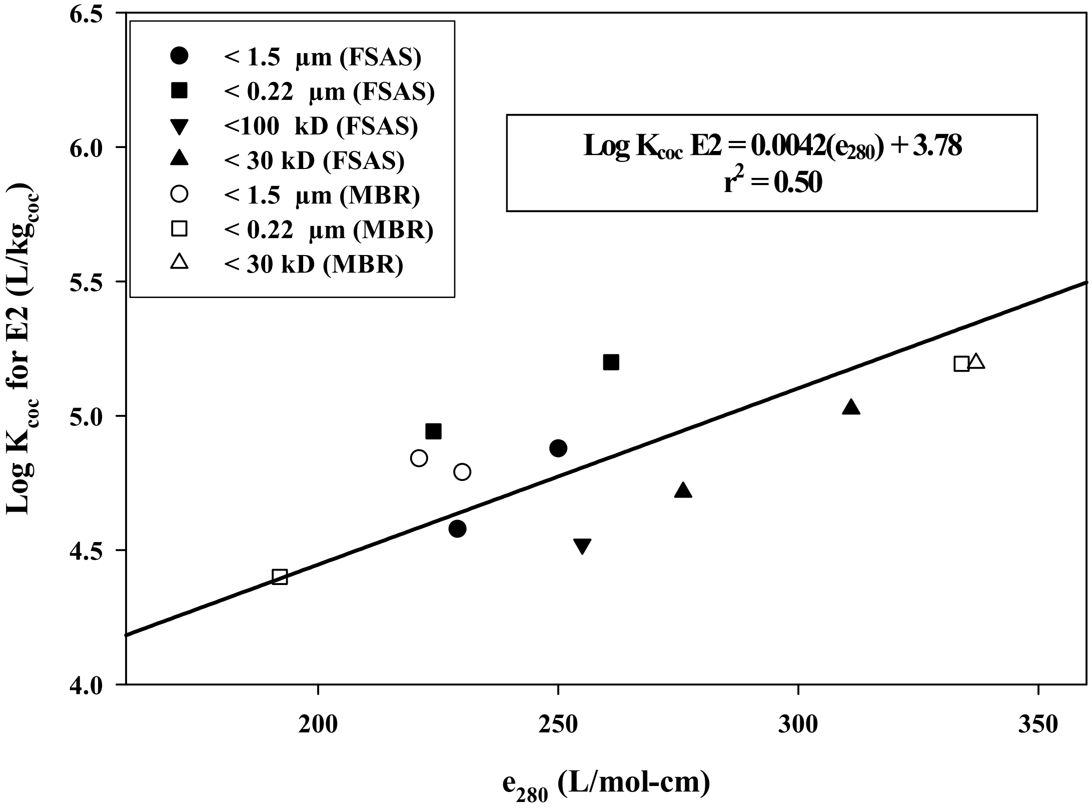


Figure 5-3: Correlation between Log K_{coc} E2 and e_{280} for the FSAS and MBR samples. Regression line is for both FSAS and MBR samples.

Aromatic Content. The correlation between the E2 log K_{coc} and e_{280} coefficients were relatively strong ($r^2 = 0.50$) for both the FSAS and MBR samples (Figure 5-3). The use of a single line to describe the log K_{coc}-e_{280} correlation is justified by the statistically equivalent E2 sorption by the FSAS and MBR samples (student's t-test, $p < 0.05$). The correlation illustrated in Figure 5-3 demonstrates that the COC aromatic content plays an integral role in the colloidal affinity for E2. Moreover, the correlation also suggests that π-electron interactions between E2 and MLSS-derived COC are an important sorption mechanism (11,16).

There was no significant correlation between the log K_{coc} for EE2 and e_{280} coefficients for the FSAS and MBR samples (Figure 5-4). The p value of the regression line was equal to 0.23, indicating that the slope was not statistically different than zero.

Therefore, the aromatic content of the colloidal material was not an important factor in describing the affinity of EE2 for MLSS-derived COC.

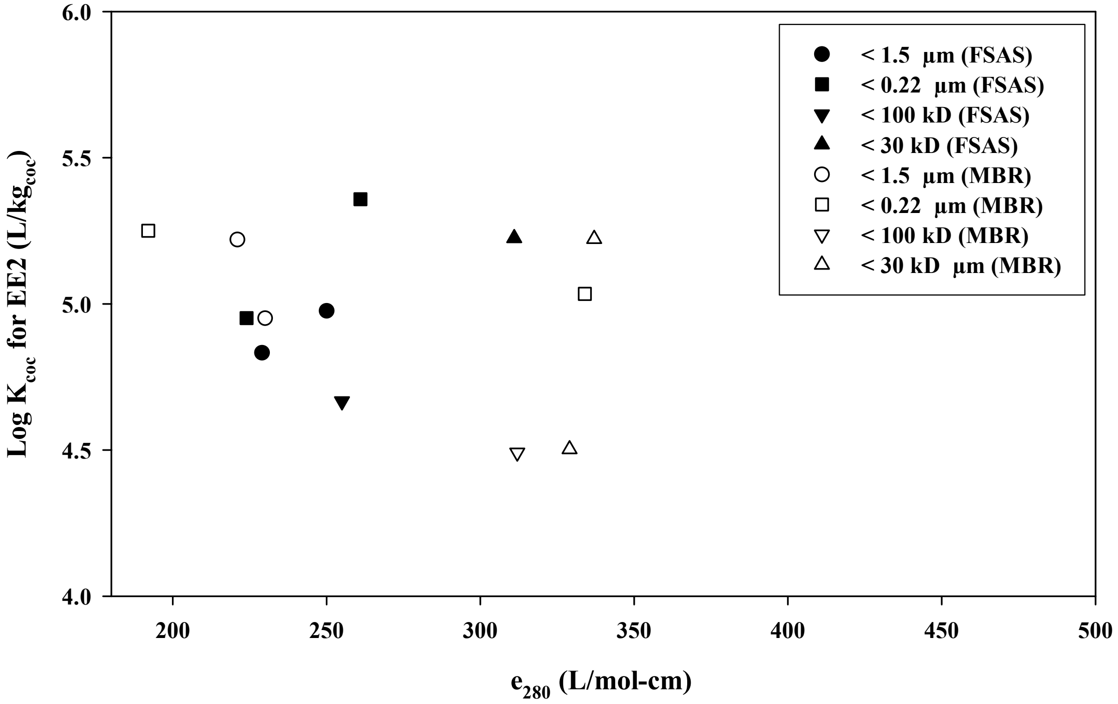


Figure 5-4: Correlation between log K_{coc} EE2 and e_{280} for the FSAS and MBR samples. Separate regression lines are shown for both FSAS and MBR samples.

The different relationships between the COC aromatic content and K_{coc} coefficients for E2 and EE2 is somewhat surprising due to the structural similarity of the two compounds. However, several investigators have observed that aliphatic compounds are the dominant structural component of wastewater organic matter (55). The interaction between the wastewater organic aliphatic groups and the ethyl group of EE2 may be important to overall EE2 sorption behavior during the biological wastewater treatment process, but will require further investigations to fully quantify its role in organic contaminant sorption (38,56,57).

The correlations between log K_{coc} and e_{280} coefficients are in partial agreement with Yamamoto *et al.* (21). These investigators demonstrated a positive correlation between sorption and e_{280} coefficients for both E2 and EE2. Again, comparison of the results between this study and (21) suggest that the colloidal structure and/or composition from the MBR systems possess unique qualities compared to other COC materials.

Surrogate Compounds. A correlation analysis was conducted to see if the log K_{coc} for E2 could predict log K_{coc} for EE2 and vice versa. The correlation as very strong for the FSAS samples ($r^2 = 0.89$) but was very weak and statistically insignificant ($r^2 = 0.07$, $p = 0.54$) for the MBR samples (Figure 5-5). These results indicate that either E2 or EE2 could be used to accurately estimate the sorption behavior of the other compound for the different size fractions of FSAS-derived COC material, but not for COC originating from the MBR samples.

Organic Carbon Composition. Correlations between individual concentrations and concentration ratios of organic components (i.e. protein, polysaccharide) for different colloid size fractions and sorption coefficients were weak ($r^2 < 0.2$) suggesting that neither protein nor polysaccharide concentrations are a dominant factor in determining sorption behavior for these samples (data not shown).

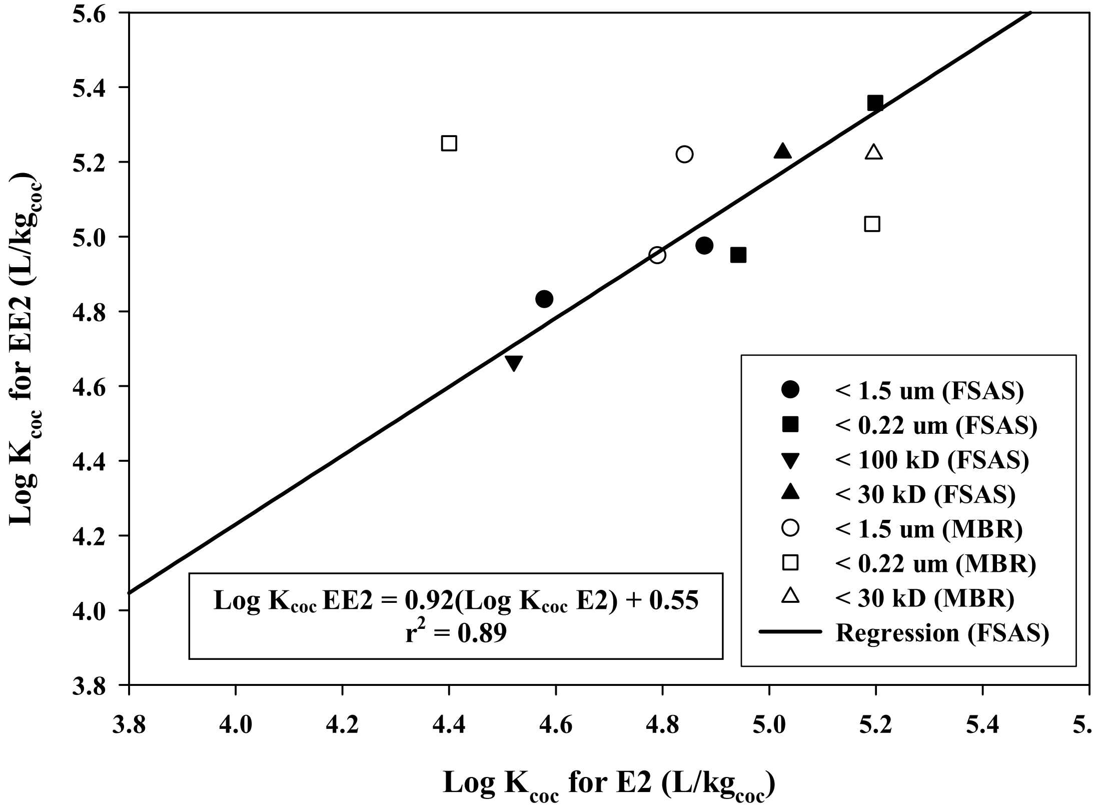


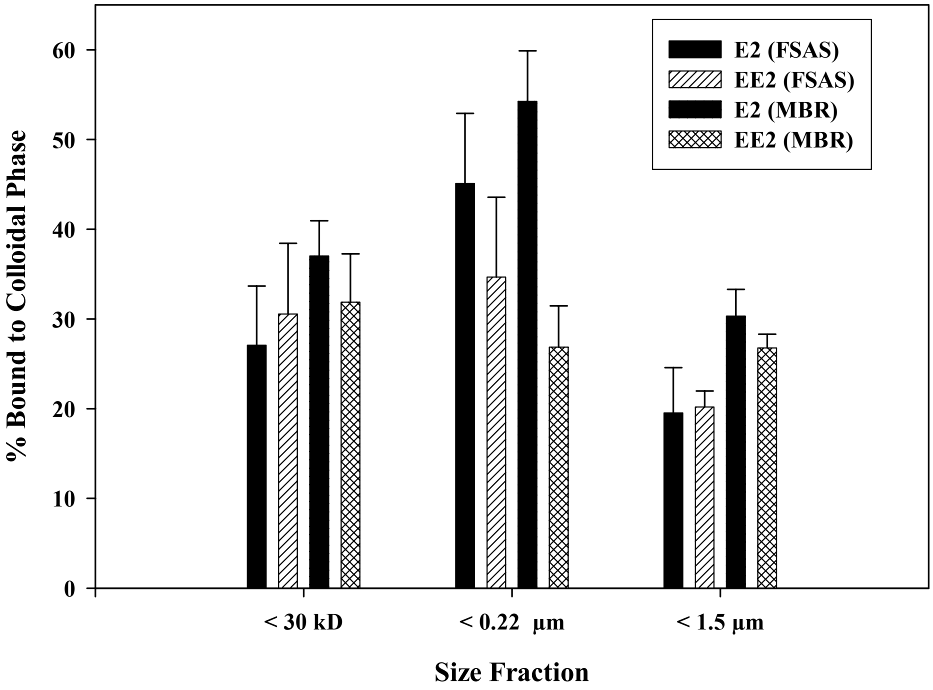
Figure 5-5: Correlation between log K_{coc} EE2 and log K_{coc} E2 for the FSAS and MBR samples. Regression line and equation is for the FSAS samples only.

An organic matter component that was not measured in this study due to sample volume restrictions is the humic and fulvic acid concentrations. Effluent humic and fulvic acid concentrations in wastewater effluent appear to be variable (55,57) but may represent an important sorption source for wastewater organic contaminants (58) due to their polyphenolic and aromatic structure (42,59,60). Yamamoto *et al.* (21) demonstrated that NOM with higher phenolic group concentrations had higher sorption capacities for both E2 and EE2. In comparing natural and anthropogenic fulvic acids extracted from waste activated sludge (WAS), Esteves da Silva *et al.* (61) observed that fulvic acids extracted from WAS had higher amounts of phenolic groups compared to natural fulvic acids, suggesting that wastewater derived fulvic acids would have a higher sorption capacity for E2 and EE2 compared to the NOM used by Yamamoto *et al.* (21). For our samples, if humic and fulvic acids do contribute to the uncharacterized OC, then the FSAS samples have higher concentrations of humic and fulvic acids compared to the MBR samples (Figure 5-1).

Implications for Full-Scale Wastewater Treatment Plants. The variation in K_{coc} coefficients between the different systems (FSAS vs. MBR) and between the different sample periods (May vs. August 2002) indicate that temporal and spatial variations in the colloidal-aqueous phase distribution of E2 and EE2 can be expected from biological wastewater treatment plants. The percentage of colloid-bound E2 and EE2 can be estimated from the following equation (62):

$$\text{Bound E2 and EE2 (\%)} = \frac{[\mathrm{COC}](\mathrm{K_{COC}})}{1+[\mathrm{COC}](\mathrm{K_{COC}})} \qquad \text{(Equation 4)}$$

Using the data summarized in Tables 5-1 and 5-2, we estimate that between 1 and 54% of E2 andEE2 may be associated with colloidal material for a given size fraction (Figure 5-6). In Figure 5-6 (top), it is not immediately clear why the < 1.5 µm size fraction should have a lower fraction of E2 and EE2 associated with the COC compared to the other size fractions, but this result suggests that a significant amount of carbon in the < 1.5 µm sample was unable to quench the fluorescence intensity of E2 and EE2. Nonetheless, these results demonstrate that the extent to which E2 and EE2 associate with colloidal material will be both significant and highly variable.



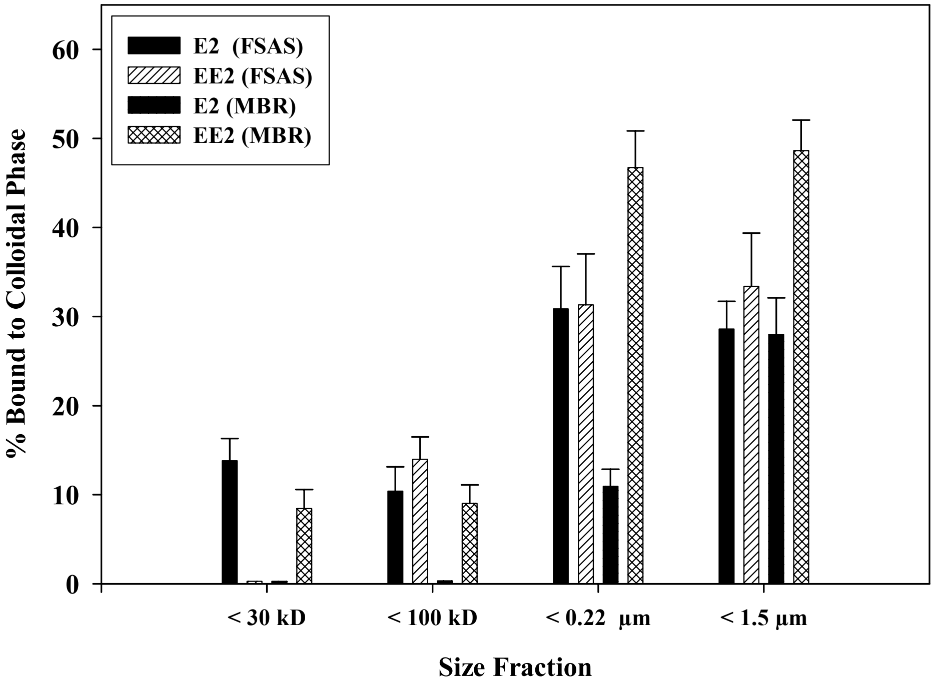


Figure 5-6: Fraction of E2 and EE2 bound to colloidal material for May 2002 (top) and August 2002 (bottom) samples for the different size fractions. Error bars represent standard error.

Activated sludge treatment processes that enhance colloidal material retention, such as MBR systems, have lower effluent concentrations of E2 and EE2 (6) as well as lower effluent estrogenic activity levels (63). The reduction in effluent E2 and EE2 concentrations is likely caused by the capture of larger colloidal material (e.g., > 0.22 μm), which can sorb the estrogens. However, the ability of low molecular weight colloidal material (< 100 kD, which is equivalent to a molecular size of approximately 10 nm) to bind E2 and EE2 indicates that MBR permeate will contain colloid-associates contaminants. Consequently, reducing the colloid-associated estrogenic activity fraction of treated effluent by physical processes (i.e., sand or membrane filtration) will require pore sizes less than 1 kD (equivalent to approximately 0.94 nm (64)). Complete removal of the colloidal-bound fraction can be accomplished with reverse osmosis that, if operating with a molecular weight cut-off of 200 g/mol, will remove even dissolved E2 and EE2. However, this type of advanced treatment is typically cost prohibitive for rural and urban areas, indicating that new wastewater management and/or treatment options must be developed in order to mitigate the release of estrogenic compounds to the natural environment. Based on the results of this study, new treatment options could include a colloid destabilization, flocculation, and particulate capture processes to maximize removal efficiency

It should also be noted that these experiments were designed to investigate the abiotic behavior of E2 and EE2 and eliminate microbial activity. Full-scale wastewater treatment facilities rely on microbial degradation for sewage stabilization and E2 has been shown to be readily degraded while EE2 appears to be more recalcitrant (65,66). Therefore, microbial degradation may influence the distribution of E2 and EE2 between the colloidal and dissolved phases.

Tanghe and Verstraete (67) reported that nonylphenol, a detergent metabolite EDC that can contribute to effluent estrogenic activity at high concentrations (68), was not as efficiently removed by granular activated carbon (GAC) when mixed with humic acids. These authors suggested that the removal efficiency reduction was caused by the competition for nonylphenol between the humic acid and GAC, suggesting that colloidal-associated contaminants in treated effluents may be more difficult to remove with conventional polishing processes compared to contaminants in the dissolved phase.

The results of the current study indicate that the aromatic content of the COC size fractions plays a large role in the sorption behavior of E2 for the FSAS system, and therefore unit processes which reduce the COC aromatic content will reduce the colloidal sorption of E2 and improve the performance of effluent polishing processes such as GAC. For example, the process of effluent chlorination appears to destroy a portion of the COC aromatic content (Figure 5-7), which is illustrated by the reduction in e_{280} coefficients of the different size fractions downstream of this unit process. Therefore, chlorination can also be used to increase the dissolved concentrations of E2 and increase the removal efficiency of downstream polishing processes such as GAC. However, the influence of chlorination on releasing sorbed EDCs has not been studied, but deserves further assessment.

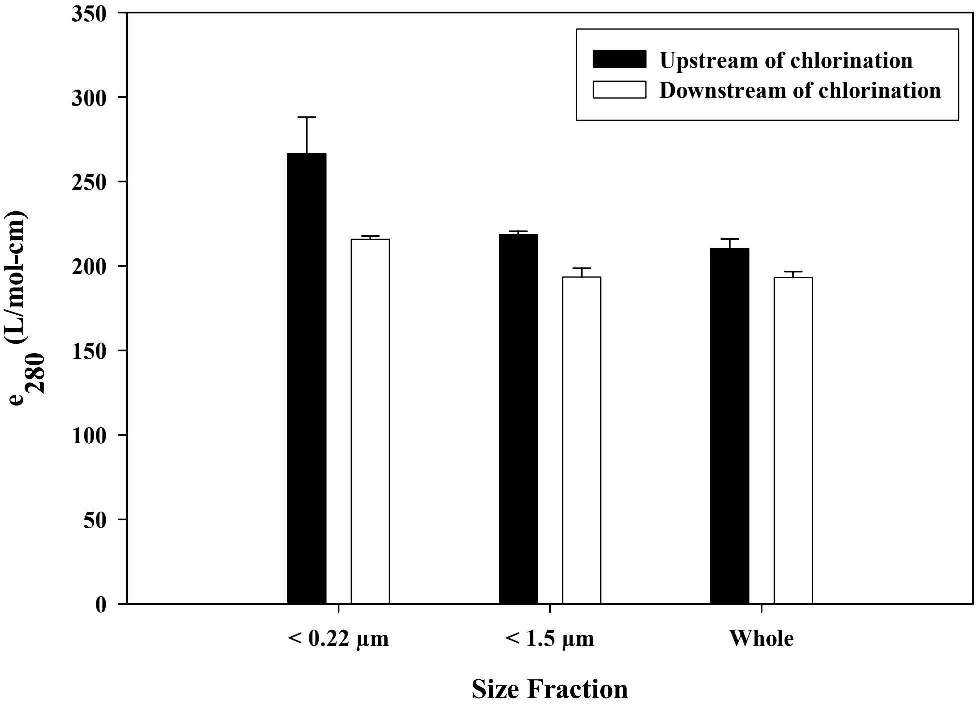


Figure 5-7: Comparison of molar extinction coefficients of FSAS-treated effluent upstream and downstream of the chlorination process. Error bars represent 95% confidence interval (n = 6).

Comparison with Other Investigations. Table 5-3 provides a comparison of sorption coefficients for E2 and EE2 from this study and other published reports. The range of organic carbon-normalized sorption coefficients is fairly wide, once again demonstrating that organic carbon quantification is not sufficient for accurate assessment of sorption behavior. However, the results of Table 5-3 imply that smaller material (i.e., colloids) will possess a greater sorption affinity for E2 and EE2 compared to either sediment or suspended particles. For example, Holthaus *et al.* (69) observed that smaller bed sediment particles possessed greater affinity for E2 and EE2 compared to larger particles. Indeed, Bowman *et al.* (20) observed that colloid particles had sorption coefficients two orders of magnitude greater than sediment particles on a dry weight basis.

Table 5-3: Sorption Coefficients Obtained for E2 and EE2 for Different Samples.

Sample	E2 (x 10^3 L/kg)	EE2 (x 10^3 L/kg)	Comments	Reference
River Bed Sediments	0.64 - 2.9	nm	1997	(69)
River Bed Sediments	0.72 - 5.6	1.4 - 10	1999/2000	(69)
River Suspended Solids	0.43 - 1.6	0.19 - 2.9		(69)
Estuarine Suspended Solids	3.03	nm	Average value	(20)
Estuarine Colloids	23	nm	Assuming $f_{oc} = 0.6$	(20)
DOM Surrogates	0.58 - 190	1.1 - 166		(21)
Wastewater COC	< 1 - 158	< 1 - 228		This study

nm = not measured.

Recently, Yamamoto *et al.* (21) published the results of an investigation that utilized FQ to quantify the affinity of DOM surrogates for E2 and EE2. Interestingly, the relative impact of the aromatic content appears to be much greater in our study compared to that of Yamamoto *et al.* (21). For example, the log K_{oc}/molar extinction coefficient ratio for the wastewater COC is higher (ratio = 0.0042) compared to the DOM surrogates used by Yamamoto *et al.* (21) (ratio = 0.0015) (Figure 5-8). This comparison suggests that there are some important differences in the composition between the DOM surrogates used by Yamamoto *et al.* (21) and the COC used in this study. Indeed, Drewes and Croute (18) determined that the aromatic content of COC was 2.6 – 3.7 times higher for a given molar extinction coefficient when compared to DOM samples. By comparison, the log K_{oc}/molar extinction coefficient ratios between this study and that of Yamamoto et al. (21) were 2.8 (0.0042 divided by 0.0015), which is in excellent agreement with Drewes and Croute (18). Given that the dominant sorption mechanism between DOM and E2 are π-electron interactions, DOM should have significant affinity for these compounds due to their comparatively higher molar extinction coefficients (Figure 5-9). Logically, the receiving water quality (i.e., colloidal quality) will play a large role in the cycling and transport of estrogenic compounds in the natural environment.

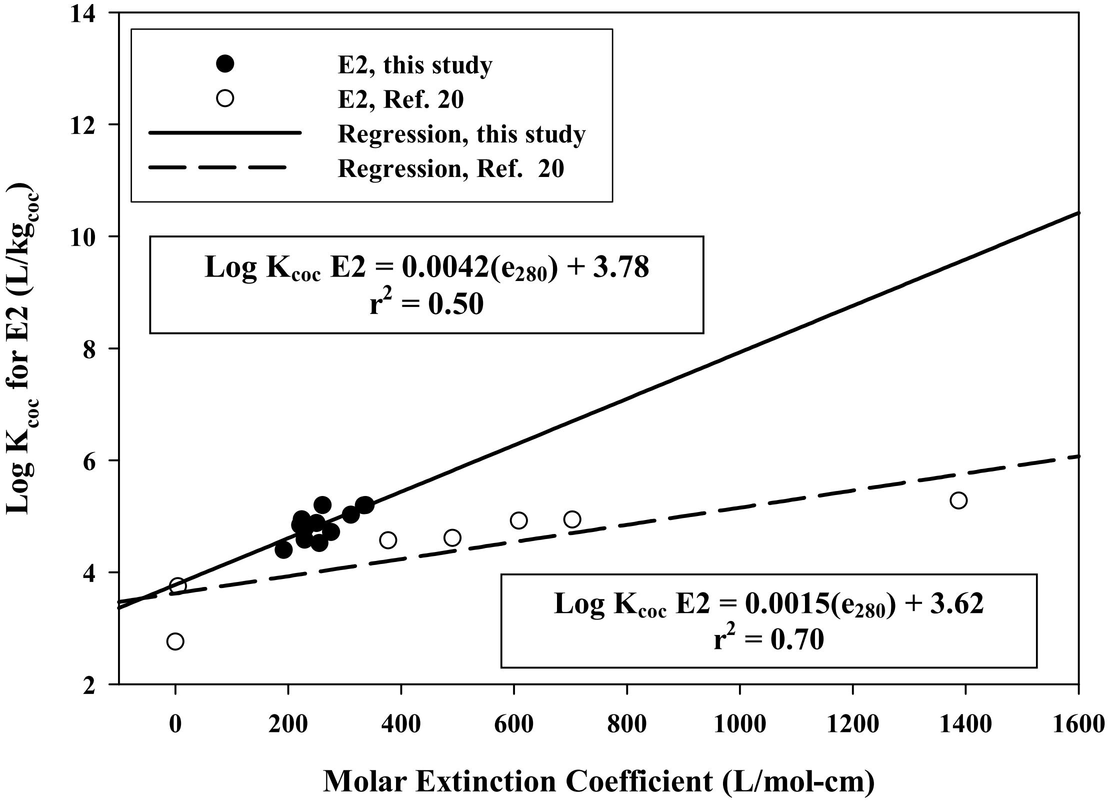


Figure 5-8: Comparison of correlations between log K_{coc} and molar extinction coefficients for this study and reference 20.

Conclusions

Fluorescence quenching experiments revealed that COC from FSAS and MBR systems has a strong capacity for sorbing E2 and EE2. Of all the investigated descriptors, the molar extinction coefficient at 280 nm appears to provide the best *a priori* estimates of colloidal sorption coefficient (K_{coc}) for E2 suggesting that interaction between the π-electrons of E2 and COC is an important sorption mechanism. Consequently, wastewater processes that facilitate the destruction of aromatic moieties, such as chlorination for disinfection, may decrease sorption of E2 to colloids in the discharged effluent. The results of this study did not reveal any strong descriptors of colloidal sorption of EE2. A substantial portion of the aqueous E2 and EE2 concentrations (between 1 and 50%) may be associated with colloidal material, indicating that colloidal transport of E2 and EE2 may be a significant route for estrogenic compounds to be discharged to the natural environment from biological wastewater treatment systems.

Acknowledgements

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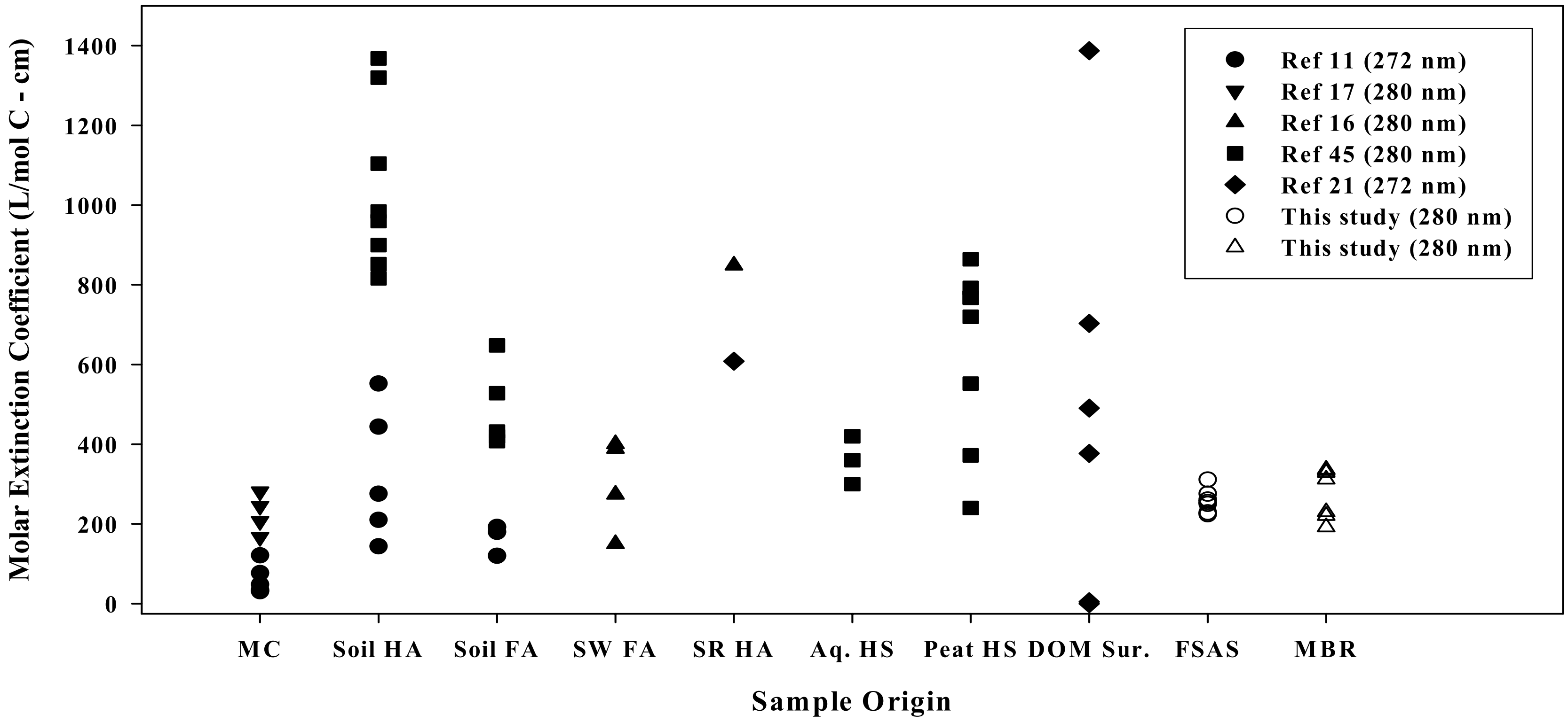

Figure 5-9: Comparison of molar extinction coefficients between NOM from different sources and COC from this study. Numbers in parenthesis are the measurement wavelength for the different investigations. Sample origins are marine colloids (MC), soil humic acid (Soil HA), soil fulvic acid (Soil FA), surface water fulvic acid (SW FA), Suwannee River humic acid (SR HA), aquatic humic substances (Aq. HS), peat humic substances (Peat HS), dissolved organic matter surrogates (DOM), final settling activated sludge colloidal organic carbon (FSAS), and membrane bioreactor colloidal organic carbon (MBR).

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Chapter 6. Evaluation of a Recombinant Yeast Bioassay to Quantify the Bioavailability of Colloid-Associated 17β-Estradiol

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ABSTRACT

Sorption by colloidal material can dramatically reduce the impact of toxic contaminants in the natural environment by reducing the toxicant's bioavailability. A recombinant yeast bioassay (YES bioassay) containing the human estrogen receptor (hER) gene was evaluated as a potential tool for measuring the bioavailability of 17β-estradiol (E2) sorbed to size fractionated colloidal organic carbon (COC) originating from a biological wastewater treatment facility. The EC_{50} values of the E2 dose-response curves were used to quantitatively measure the estrogenic activity of the COC-E2 mixture. EC_{50} values significantly increased as a function of COC concentration. However, there was a high degree of variability in the EC_{50} values that may be attributed to the complexity and unpredictability of wastewater samples. There was no strong correlation between the normalized EC_{50} values and colloidal protein, polysaccharide, humic and fulvic acid concentrations. Moreover, estimates of the colloid-associated E2 fraction were unsuccessful in accurately predicting increases in EC_{50} values. Small colloidal material (< 10 kD) may act as partial hER antagonists and cause increase in EC_{50} values. Nevertheless, the increased EC_{50} values for the larger size fractions (> 100 kD) were attributed to a reduction in bioavailability. However, matrix effects of the specific environment being tested need to be closely evaluated due to the sensitivity of the hER and reporter plasmid.

Introduction

The presence of hermaphrodite fish in wastewater treatment lagoons led Purdom *et al.* (1) to hypothesize in 1994 that estrogenic compounds were both present and biologically active in treated wastewater. These authors demonstrated that male fish were producing significant concentrations of vitellogenin, a female protein used for egg yolk development, when exposed to wastewater effluents. This ground-breaking study generated world-wide interest in the presence and effect of natural and anthropogenic chemicals that can modulate and/or disrupt the endocrine system of wildlife and humans (2-6). Identifying and quantifying estrogenic endocrine disrupting compounds (EDCs) in treated effluents has remained a research priority, an effort warranted by reports of widespread sexual disruption of fish in receiving waters (5,7-11) and potential negative impact on human health (2,12-14). The major EDCs found in wastewater effluents include natural and synthetic hormones (15-21) such as 17β-estradiol (E2), 17α-ethinylestradiol (EE2), and estrone, and industrial surfactant metabolites (22-25) including nonylphenol (NP) and octylphenol. The majority of estrogenic activity in treated effluents is caused by E2 and EE2 (18,19) although high effluent concentrations of NP have also resulted in sexual disruption of juvenile bass (8).

While identification and quantification of EDCs is an important element of environmental risk management, the true assessment of EDC potency is the bioavailability of these compounds once they are discharged to the natural environment. For example, carbonaceous resins reduce the bioavailability of polyaromatic hydrocarbons (PAHs) without complete removal of the compound from the environment and therefore can be considered a remediation technique (26). Currently, the most accurate assessment of EDC bioavailability is the use of *in vivo* bioassays, such as the widespread use of fish (e.g., (11,27)). Logically, *in vivo* bioassays are the best approach for determining the overall health impact of a specific chemical since they incorporate the entire organism into the experiment. However, the disadvantages of *in vivo* bioassays include relatively high cost, the need for large sample volumes, and the questionable practice of using fish for gauging the impact of EDCs on human health. Indeed, the estrogen receptors in humans and trout are structurally distinct and, therefore, may have no relationship to each other and varying sensitivities to EDCs (28).

Since *in vivo* studies involving EDCs and human subjects are impossible, two *in vitro* assays that simulate aspects of the human endocrine system have been developed. These bioassays include proliferation of the human breast cancer cell line (MCF-7)

(12,29) and the yeast estrogenic screen (YES) using a recombinant yeast strain (*Saccharomyces cerevisiae*) containing the human estrogen receptor (hER). The YES bioassay has been widely studied (30,31) and used in determining the estrogenic activity of specific compounds (32,33) and environmental samples (21,34-36). However, these studies have relied on extraction and concentration techniques designed to provide maximum bioassay response for a given EDC concentration. To our knowledge, the YES bioassay has never been used to measure the bioavailability of EDCs from wastewater samples.

Although use of the YES bioassay for measuring EDC bioavailability will not provide the robust, complex biological system information obtained by an *in vivo* experiment, it may provide a less-expensive and rapid means for obtaining bioavailability data specific to the hER. Subsequently, screening for potential EDC remediation techniques based on reduction of EDC bioavailability could be done more efficiently.

In this study, we investigated the YES bioassay as a method for determining the bioavailability of E2 in the presence of colloidal organic carbon (COC) derived settled mixed liquor suspended solids (MLSS) from a biological wastewater treatment facility. The effects of various concentrations and size fractions of COC on E2 bioavailability were evaluated by comparing dose-response curves generated by the YES bioassay. Correlations between E2 bioavailability and components of the COC are presented, and factors that may influence the dose-response curves are discussed.

Materials and Methods

Experimental Approach. In earlier investigations, we demonstrated that MLSS-derived COC has a significant sorption capacity for E2 (37,38). In this study, we exposed aliquots of MLSS-derived COC and E2 solutions to the YES bioassay and compared dose-response curves to quantify E2 bioavailability. There are four parameters that describe a typical dose-response curve, including minimum absorbance (min), maximum absorbance (max), slope, and the EC_{50} value (Figure 6-1). In evaluating toxicity data, it is customary to compare EC_{50} values (the concentration of E2 is half way between the minimum and maximum absorbance readings) to determine the relative potency of contaminants (39,40). For the situation studied here, the EC_{50} concentrations will be compared for the same contaminant (E2) in the presence of various COC size fractions and COC concentrations. An increase in EC_{50} indicates that more E2 is needed to effect the same degree of YES expression, and suggests that some E2 is being bound by COC in the sample, making it less bioavailable. For example, in the hypothetical dose-response curves depicted in Figure 6-1, a single toxicant chemical is added to samples with different COC concentrations. Sample 2 appears to bind the toxicant to a greater degree, making the toxicant less bioavailable, since EC_{50}- 2 is greater than EC_{50}-1.

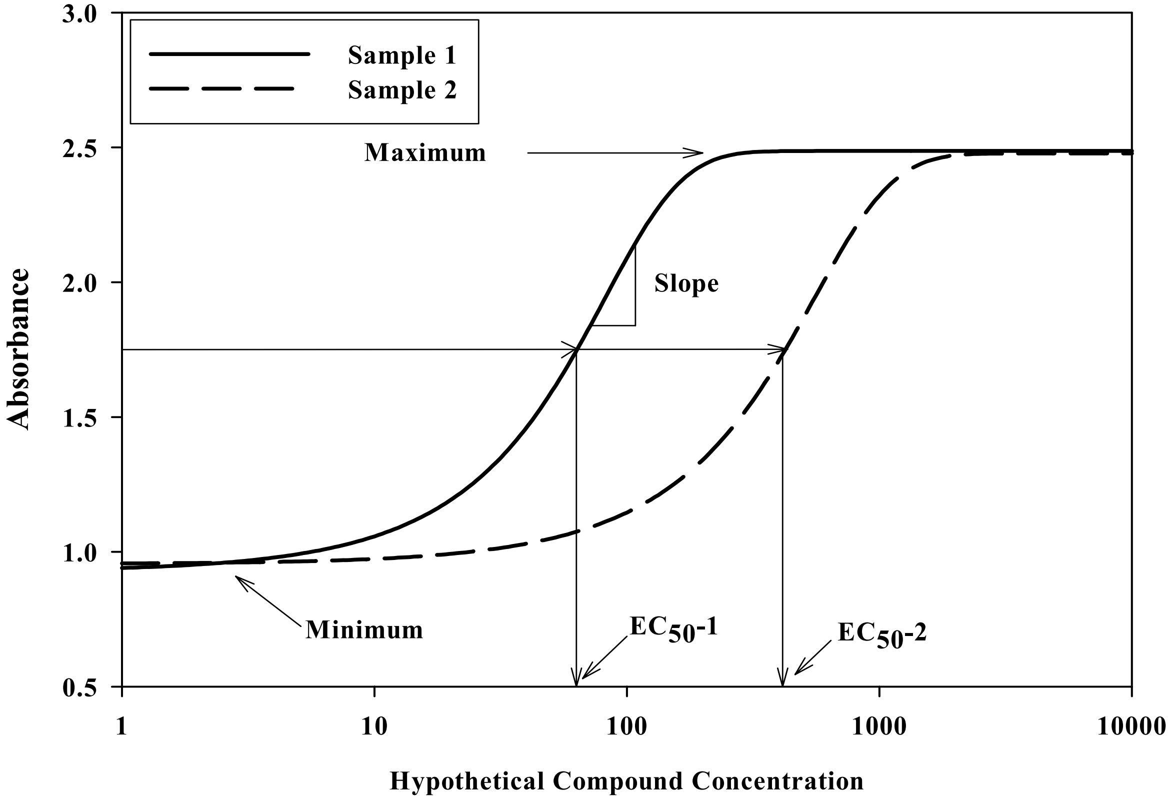

Figure 6-1: Hypothetic dose-response curve illustrating the locations of the four parameters.

Facility Overview. Samples were collected from the west aeration basin of the Blacksburg-Virginia Polytechnic Institute and State University (VPI & SU) Sanitation Authority Stroubles Creek Wastewater Treatment Plant (WWTP). This WWTP receives primarily domestic sewage, though the strength depends heavily on the presence or absence of the transient student population. The bulk of the student population was present during the January, March, and April 2003 sampling period but absent for the May 2003 sample. The process flow scheme included primary clarifiers, anoxic and aerobic bioreactors, and secondary clarifiers for liquid-solid separation. The mean cell residence time (MCRT) during the sampling periods was approximately 12 days and mixed liquor suspended solids (MLSS) concentrations averaged approximately 1,600 mg/L.

Sample Collection, Preparation, and Size Fractionation. Four separate grab samples were collected from the west basin of the aerobic bioreactor between January and May 2003. During each sampling period, approximately 5 L of MLSS were collected in glass bottles, which had been cleaned by rinsed in hot tap water, soaked in 1 M NaOH (minimum 1 hour), rinsed in Milli-Q water, rinsed in ethanol, and thorough rinsed again in Milli-Q water. Samples were transported from the WWTP to the laboratory within 15 minutes of collection, and the MLSS was allowed to settle for another 45 minutes for a total settling time of 1 hour. The MLSS supernatant was decanted into a similarly cleaned glass container and aliquoted for protein, polysaccharide, humic and fulvic acids, and organic carbon concentrations. Approximately 2 L of supernatant was transferred to a glass filtration apparatus and filtered through pre-combusted 1.5 μm glass fiber filters (Whatman 934-AH). Aliquots of the < 1.5 μm filtrate were used for further size fractionation by using 0.22 μm cellulose nitrate disc filters (Fisher) and ultrafilter membranes, in parallel, to yield a total of 6 COC solutions (unfiltered supernatant and 5 size fractions defined as < 1.5 μm, < 0.22 μm, < 100 kD, < 30 kD, and < 10 kD. These COC solutions were used in the bioavailability experiments. In addition, a < 1 kD solution was prepared and is defined as colloid-free, or truly dissolved.

The 0.22 μm disc filters were conditioned with approximately 10 mL of < 1.5 μm filtrate prior to producing the < 0.22 μm filtrate. Amicon (Billercia, MA) YM100, YM30, and YM10 and YM1 ultrafilters with nominal molecular weight unit cut-offs (MWCO) of 100, 30, 10, and 1kD, respectively, were used in these experiments. Prior to using the ultrafilters, each membrane was soaked for 1 hour in distilled water that was changed three times, soaked overnight at 4˚C, and finally flushed with 200 mL of Milli-Q water to remove the glycerine preservative added by the manufacturer. All ultrafiltration experiments were conducted in magnetically stirred batch cells (200 mL volume), pressurized with nitrogen gas (350 kPa), stirred at approximately 300 rpm, and operated at room temperature (23 ± 1˚ C). For the ultrafilter cells, collecting only 70% of the applied volume was collected (140 mL) to reduce colloid breakthrough (41). Two individual ultrafilter cells were used to produce approximately 280 mL of the dissolved fraction (< 1 kD), which was subsequently used as the dilution solution for all experiments. The 1.5 µm and 0.22 µm filters were used only once and discarded. The ultrafilter membranes, which were used a maximum of three times before being replaced, were cleaned between filtering events by flushing with 50 mL of 0.1 N NaOH followed by 100 mL of Milli-Q water and stored in Milli-Q water at 4˚ C.

Sorption Procedure. 8 mL of < 1 kD permeate and 4 mL of a specific COC solution were combined in borosilicate glass tubes. A total of three glass tubes were used for each COC solution. 17β-estradiol (E2) was spiked to a final concentration of 10^{-7} M in two of the tubes while the third tube was used as a negative control. Each tube was covered with aluminum foil, capped, tumbled at 30 rpm for 2 minutes, and allowed to equilibrate for 20 minutes. This procedure allowed sorption between E2 and COC to reach apparent equilibrium as previously determined in our lab (37). Fifty µL aliquots from each tube were transferred to 96 –well microtiter plates (Linbro/Titertek, ICN Biomedicals, Aurora, OH) for use in the YES bioassay. The entire procedure was repeated two times by adding four mL aliquots of the specific COC solution to the previously used glass tubes to increase the COC concentration, mixed, allowed to equilibrate, and aliquoted in the 96-well microtiter plate. The same protocol was used for the < 1 kD permeate, which served as the dissolved phase control.

All glassware and equipment used in the sorption procedure was autoclaved (121˚ C for 30 minutes) to eliminate interferences resulting from microbial contamination.

Recombinant Yeast Estrogen Screen (YES) Bioassay. Bioavailability of 17β-estradiol was determined using the YES bioassay as previously described by Routledge and Sumpter (31), except for modifications as described below. A recombinant yeast strain containing the hER gene was obtained from Dr. John Sumpter of Brunel University

(Middlesex, UK). The strain also contains an expression plasmid carrying the *lac-Z* reporter gene. When the cells are incubated in the presence of estrogenic compounds, the *lac-Z* product, ß-galactosidase (ß-gal), is secreted into the medium and causes the chromogenic substrate, chlorophenol red-ß-D-galactopyranoside (CPRG) (Roche Diagnostics, Indianapolis, IN), to turn red. This color change can be quantified by measuring absorbance (Spectracount Microplate Photometer BS-10,000, Packard, Meriden, CT). In the 96-well microtiter plates, 50 µL aliquots from each glass tube was serially diluted in twelve wells using < 1 kD permeate. Ten µL aliquots from each of the serially-diluted wells were transferred, in duplicate, to new wells to which 200 µL of growth medium containing yeast cells (grown to an absorbance of 1 at 600 nm) was added. The plates were incubated at 32° C for 3 days, then moved to room temperature for 2 days, as suggested by Beresford *et al*. (33).

After the total incubation period, dose-response curves were created from absorbance readings taken at 540 nm and 620 nm for each size fraction and at each carbon concentration. Since β-gal may be present in the COC solutions, the results from any E2-spiked sample were discarded if the accompanying negative control had any measurable color change. As a result, unfiltered supernatant and < 1.5 µm COC solutions had to be autoclaved prior to use while the other size fractions (1 kD to 0.22 µm) could be used without any treatment.

A plate containing standards and control samples was included for every YES bioassay performed and included: 3 rows of serially diluted 17 ß-estradiol (> 98% purity, Sigma Chemical Company, St. Louis, MO) using both Milli-Q water as the diluent (positive control); a row containing negative control samples (Milli-Q only); and a row containing blank samples (growth media only). The concentrations for the E2 serially diluted wells ranged from 12 to 25,000 pM. The standard curve used to determine estradiol-equivalents (E2-Eq) in molar units was determined from the arithmetic mean of the 3 wells containing the same 17 ß-E2 concentrations.

Analytical and Calculation Methods. Non-volatile organic carbon concentrations were measured using a Sievers 800 TOC Analyzer. Protein was measured using the Frolund *et al.* (42) modification of the Lowry *et al.* (43) method. Polysaccharides were measured using the method of Dubois *et al.* (44). Protein standards were prepared with bovine serum albumin, and polysaccharide standards were prepared with glucose. Humic (HA) and fulvic acids (FA) were quantified by a Beckman DU-640 spectrophotometer and Perkin-Elmer 650-10S fluorescence spectrophotometer, respectively, using the method described by Hautala *et al.* (45). Absorbance measurements were taken of the COC solution at 280 nm on the Beckman spectrophotometer. Each organic carbon component was measured in triplicate, and the arithmetic average was used in the analysis.

COC solution concentrations were calculated by subtracting the dissolved phase (< 1 kD permeate) concentration of a specific analyte (e.g., total organic carbon) from the concentration of a specific size fraction (e.g., < 1.5 μm filtrate). Molar extinction coefficients at 280 nm (e_{280}) were calculated by mathematically dividing the absorbance measurements by the colloidal organic carbon concentration (COC) for a given solution. Colloidal size was considered to be the largest expected particle in a specific size fraction, and operationally defined as the pore size of the filter. Ultrafiltration pore sizes were obtained from Schäfer *et al.* (46) and are 1500, 220, 9.1, 4.8, 2.6, and 0.94 nm for the < 1.5 μm, < 0.22 μm, < 100 kD, < 30 kD, < 10 kD, and < 1 kD. Additionally, four thousand nm was used as the maximum colloid size in the unfractionated (unfiltered) samples.

EC_{50} values were normalized to overcome the large variability experienced with wastewater samples. Normalization was achieved by dividing the EC_{50} values by the < 1 kD EC_{50} value (Equation 6-1) since the dissolved phase is unable to bind to organic compounds (48-51) and E2 should be completely bioavailable to the yeast cells. Furthermore, the < 1 kD permeate will account for any matrix-specific effect on the yeast cells, such as chemical inhibition.

$$\text{Normalized } EC_{50} \text{ value} = (EC_{50})_{i,j}/(EC_{50})_{1,j} \qquad (1)$$

where i = specific size fraction (unfiltered, < 1.5 μm, etc.) and j = COC dilution series (one, two, or three).

Regression and Statistical Analysis. Best-fit dose-response curves and the E2 concentration giving 50% of the average between the minimum and maximum response (EC_{50}) were calculated with non-linear, logistic regression using Origin 6.1

(Northampton, MA). Multivariate outlier and multiple regression analyses were performed with NCSS 2001 (Kaysville, UT) using an alpha value of 0.05 to determine significance. SigmaPlot 6.0 (Chicago, IL) was used for all linear regressions. Surfer 6.0 (Golden, CO) was used to generate 3D surface graphs.

Results

Size Fractionation. The results of the size fractionation by filtration and ultrafiltration are summarized in Table 6-1. As expected, the organic carbon, protein, and polysaccharide concentrations increased with increasing size fraction. The results for the humic acid (HA) and fulvic acid (FA) generally followed this same trend although some of the smaller size fractions had higher measurable concentrations of these components compared to the larger size fractions (e.g., < 30 kD sample in May 2003). The advantage of the method we used for HA and FA quantification (45) is that little or no sample preparation is necessary prior to analysis, making this method suitable for experiments where sample volumes are limited. However, the HA and FA quantification method (42) was calibrated with natural organic matter (NOM), and interference caused by the unique characteristics of wastewater samples (52) or by protein-like fluorophores (53) could provide overestimates of HA and FA concentrations for the COC solutions.

The e_{280} coefficients for the January, March, and April 2003 samples generally increased with decreasing colloidal size fraction while the dissolved phase had the lowest values. However, the e_{280} coefficients for the May 2003 sample had a maximum value for the < 100 kD permeate. The reduced sewage load to the wastewater treatment facility caused by the sudden decline of the student population may have influenced the e_{280} coefficients for the May 2003 sample. The reduced sewage organic load would cause a substantial reduction in the food-to-microorganism ratio (F/M), resulting in a larger proportion of cell lysis products being released to the MLSS supernatant. In contrast, the other sampling periods were operated at comparatively higher F/M ratios, which suggests a larger contribution of growth-associated soluble microbial products (SMPs) in the MLSS supernatant (54-56).

Table 6-1: Colloidal Characteristics of Size Fractionated Samples

Sample Date	Size Fraction	OC[a] (mg/L)	Protein (mg/L)	Polysaccarides (mg/L)	HA[b] (mg/L)	FA[c] (mg/L)	e_{280}[d] (L/mol-cm)
January 2003	< 1 kD	3.0	3.0	0.1	0.3	3.9	162[e]
	< 10 kD	5.7	4.8	0.6	0.9	6.6	375
	< 30 kD	6.4	5.4	0.7	0.6	6.7	307
	< 100 kD	6.9	5.7	0.8	0.6	6.6	265
	< 0.22 µm	7.5	6.0	0.9	0.5	6.4	232
	< 1.5 µm	8.4	7.0	1.0	0.6	8.7	206
	Whole	9.3	8.2	1.1	1.1	8.8	192
March 2003	< 1 kD	1.9	0.9	0.7	0.1	3.5	189[e]
	< 10 kD	3.6	2.1	0.7	0.4	4.9	396
	< 30 kD	3.7	2.4	0.8	0.5	5.1	417
	< 100 kD	5.7	5.7	1.0	1.5	5.8	329
	< 0.22 µm	5.1	4.0	1.0	0.6	5.1	263
	< 1.5 µm	8.7	6.6	1.3	3.4	8.7	302
	Whole	15.4	19.4	1.7	8.8	8.6	250
April 2003	< 1 kD	1.8	3.0	0.7	0.0	0.3	211[e]
	< 10 kD	4.0	4.7	0.7	0.3	0.5	364
	< 30 kD	4.2	5.0	0.8	4.8	0.5	361
	< 100 kD	4.3	5.0	0.7	0.5	0.5	341
	< 0.22 µm	4.4	4.9	0.8	0.6	0.5	333
	< 1.5 µm	5.2	5.3	1.0	1.2	0.5	348
	Whole	14.8	30.2	1.5	5.2	3.0	275
May 2003	< 1 kD	1.8	2.1	0.1	0.5	5.1	134[e]
	< 10 kD	4.0	3.2	0.5	0.8	7.8	369
	< 30 kD	4.2	3.3	0.6	0.8	9.6	363
	< 100 kD	4.1	3.3	0.6	0.8	7.7	441
	< 0.22 µm	5.3	3.4	0.6	0.9	7.6	180
	< 1.5 µm	4.7	3.9	0.6	1.2	9.3	411
	Whole	7.0	5.7	0.8	3.5	9.0	254

[a] organic carbon. [b] humic acid. [c] fulvic acid. [d] normalized to COC. [e] normalized to dissolved OC.

Dose-Response Curves. ***Positive control.*** The EC_{50} values of the positive control dose-response curves varied over the course of this investigation (Table 6-2). During the January and March 2003 experiments, the EC_{50} values were 72 and 39 pM, respectively. However, the EC_{50} values for the April and May 2003 samples were 116 and 118 pM. The differences in EC_{50} values may be caused by variations in the yeast culture density used to inoculate the microtiter plates (22).

Table 6-2: Summary of Parameters from E2 Dose-Response Curves in Milli-Q Water Control and COC mixtures

Sample Date	EC_{50} (pM E2)	Min (Abs)	Max (Abs)	Slope (Abs/pM E2)
Control - January 2003	72 (4)	1.1 (0.03)	2.9 (0.05)	3.3 (0.5)
Control - March 2003	39 (1)	1.0 (0.01)	2.4 (0.1)	2.2 (0.1)
Control - April 2003	118 (9)	0.65 (0.04)	3.6 (0.2)	2.0 (0.3)
Control - May 2003	116 (10)	1.1 (0.03)	3.4 (0.1)	3.6 (0.7)
Control - Averages	86 (19)	0.96 (0.1)	3.0 (0.3)	2.8 (0.5)
Experimental	See Table 6-3	0.99 (0.1)	3.0 (0.3)	3.3 (0.9)

Values are average ± (standard deviation).

Experimental. The correlations of the non-linear regressions for the experimental dose-response curve were excellent ($r^2 > 0.98$) for most samples and never below 0.94. Multiple regression analysis demonstrated that the min, max, and slope parameters have a significant effect ($p < 0.05$) on the EC_{50} values, and consequently, the dose-response parameters were screened by multivariate outlier analysis to remove any aberrant data. The multivariate outlier analysis revealed that 11% of the total dataset (19 out of 160 samples) had min, max or slope parameters that were considered outliers and therefore were not considered during the ensuing analysis. The dose-response parameters were not statistically different ($p > 0.05$) between the average Milli-Q control and experimental datasets from each COC solution (Table 6-2), suggesting that any differences in the dose-response curves could be attributed to E2 bioavailability.

A loss of sensitivity to E2 by the yeast culture was observed when the yeast incubation was carried out in the presence of colloidal organic material. Figure 6-2 illustrates that the dose-response curve of the experimental reactor containing < 0.22 µm colloidal material is shifted to the right compared to the control, resulting in an increase in the EC_{50} value. This increase in EC_{50} values for the experimental dose-response curves relative to the Milli-Q control was universal for all size fractions, including the < 1 kD permeate.

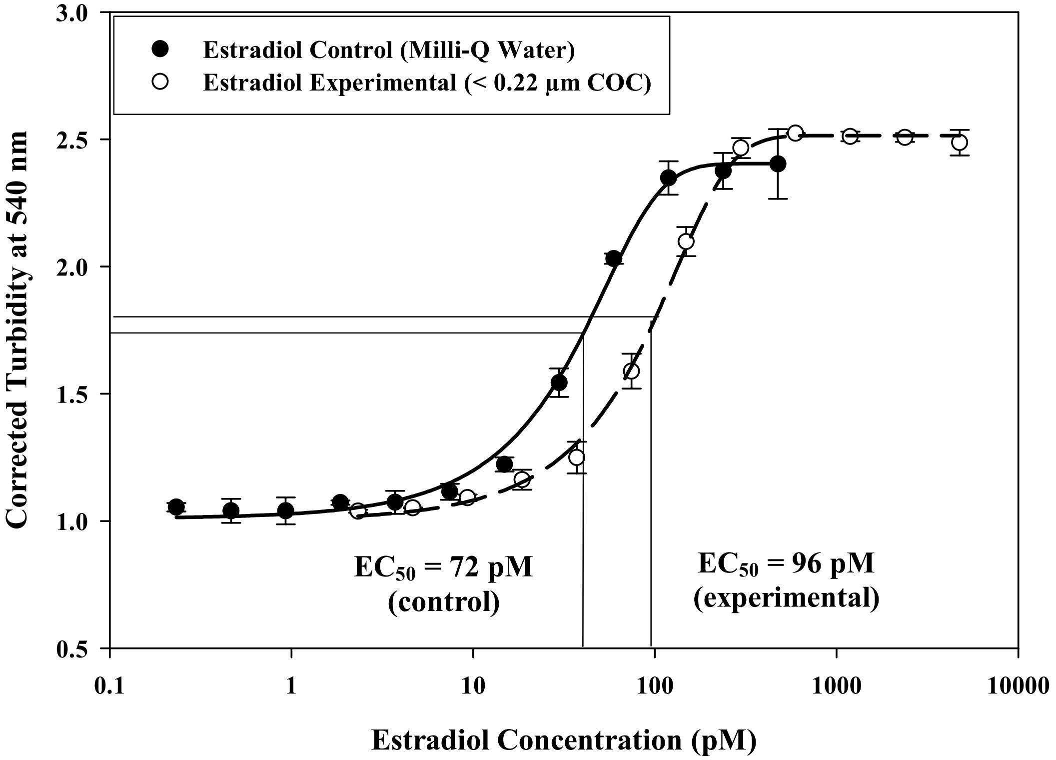

Figure 6-2: E2 Dose-Response Curves from the YES Bioassay. Closed symbols indicate the response of the YES bioassay to E2 that is diluted in Milli-Q water. The open symbols represent the response of the YES bioassay to E2 diluted in a < 0.22 µm colloidal organic carbon suspension.

The EC_{50} results (average and standard error, n = 2) of the different dose-response curves are summarized in Table 6-3. The greatest increase in EC_{50} values was observed for increasing COC concentrations of the larger size fractions whereas the EC_{50} values for the smaller size fractions remained relatively constant as a function of COC concentration (e.g., Figure 6-3). This observation suggests that the bioavailability of E2 was reduced to a greater degree by the larger size fractions of colloidal material. However, the EC_{50} values

Table 6-3: EC_{50} values for the dose-response curves for the different size fractions and COC concentrations

Sample Date	Size Fraction	COC 1 EC_{50} (pM E2)	COC 2 EC_{50} (pM E2)	COC 3 EC_{50} (pM E2)
January 2003	< 1 kD	93 (3)	83 (4)	101 (6)
	< 10 kD	85 (3)	81 (4)	82 (2)
	< 30 kD	75 (3)	78 (3)	85 (3)
	< 100 kD	83 (4)	98 (5)	110 (9)
	< 0.22 um	96 (4)	85 (4)	118 (10)
	< 1.5 um	nm	nm	nm
	Whole	nm	nm	nm
March 2003	< 1 kD	42 (3)	150 (4)	156 (6)
	< 10 kD	41 (3)	130 (4)	161 (2)
	< 30 kD	35 (3)	139 (3)	107 (3)
	< 100 kD	45 (4)	167 (5)	125 (9)
	< 0.22 um	45 (4)	166 (4)	161 (10)
	< 1.5 um	43 (8)	170 (18)	233 (18)
	Whole	nm	nm	nm
April 2003	< 1 kD	168 (8)	195 (9)	167 (9)
	< 10 kD	179 (8)	200 (10)	167 (10)
	< 30 kD	173 (7)	188 (27)	186 (8)
	< 100 kD	155 (8)	187 (7)	169 (5)
	< 0.22 um	140 (8)	160 (6)	163 (7)
	< 1.5 um	149 (8)	182 (8)	166 (8)
	Whole	nm	nm	nm
May 2003	< 1 kD	103 (5)	102 (4)	116 (2)
	< 10 kD	140 (5)	115 (6)	173 (38)
	< 30 kD	97 (5)	105 (4)	125 (2)
	< 100 kD	105 (9)	103 (12)	105 (7)
	< 0.22 um	108 (4)	123 (5)	155 (7)
	< 1.5 um	104 (7)	119 (6)	145 (7)
	Whole	125 (10)	154 (4)	169 (10)

nm = not measured.

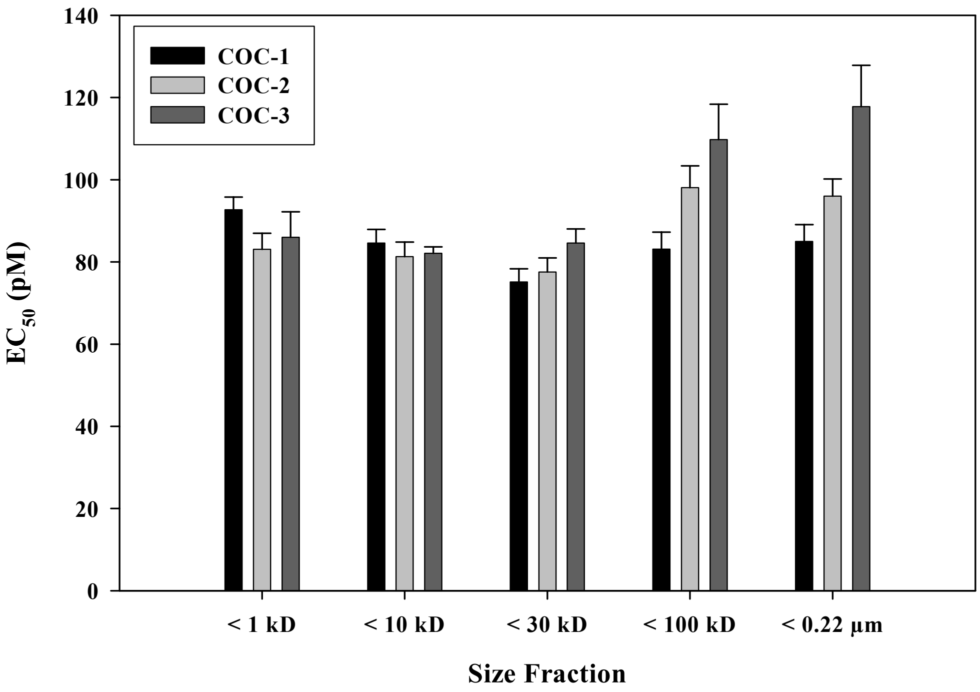

Figure 6-3: EC$_{50}$ values for January 2003 samples determined from the YES dose-response curves in the presence of different COC concentrations and size fractions.

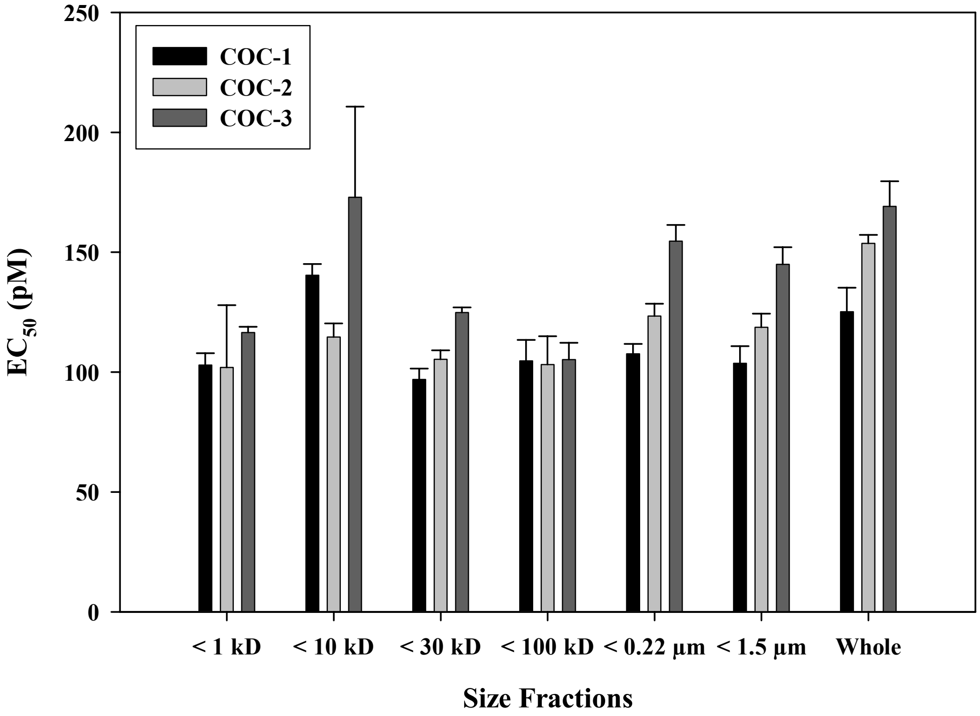

Figure 6-4: **EC$_{50}$ values of the May 2003 samples determined from the YES dose-response curves in the presence of different COC concentrations and size fractions.**

of the smaller size fractions for some samples was greater than the larger size fractions (Figure 6-4). The variability in the EC_{50} values demonstrates the complex nature of the MLSS-derived colloidal material.

Influence of Colloidal Material on Normalized EC_{50} values. Both multiple and linear regression were used to determine what organic carbon components could be used to make *a priori* estimates of the reduction in E2 sensitivity by the yeast culture. The relationship between the average normalized EC_{50} values and average COC concentrations for a specific size fraction had the greatest correlation ($r^2 = 0.79$, $p = 0.0072$) indicating that the concentration of colloids available for E2 sequestering is an important factor in reducing E2 bioavailability (Figure 6-5). However, the standard deviations around the average of the normalized EC_{50} values and COC concentrations suggest there is a significant amount of variability in these parameters.

Linear correlations between the normalized EC_{50} values and the colloidal organic components of the different size fractions were relatively weak ($r^2 < 0.22$) (Figures 6-6 through 6-10). Statistically significant slopes were observed for the colloidal organic carbon ($p = 0.004$), polysaccaride ($p < 0.0001$), and FA ($p < 0.0001$) concentrations while both protein ($p = 0.08$) and HA ($p = 0.06$) were marginal. However, multiple regression analysis revealed that the interaction between COC and HA ($p = 0.0044$) and protein and HA ($p = 0.018$) had a significant impact on the normalized EC_{50} values. Figures 6-11 and 6-12 and illustrate the relationship between COC and HA concentration and protein and HA concentration and normalized EC_{50} values, respectively. Both Figures 6-11 and 6-12 demonstrate that increases in normalized EC_{50} values can be observed at both low and high concentrations of each compound, and that relatively no change in EC_{50} is observed for concentrations in between these two extremes. Similar to Figure 6-5, the maximum normalized EC_{50} values are found in areas of high COC concentrations.

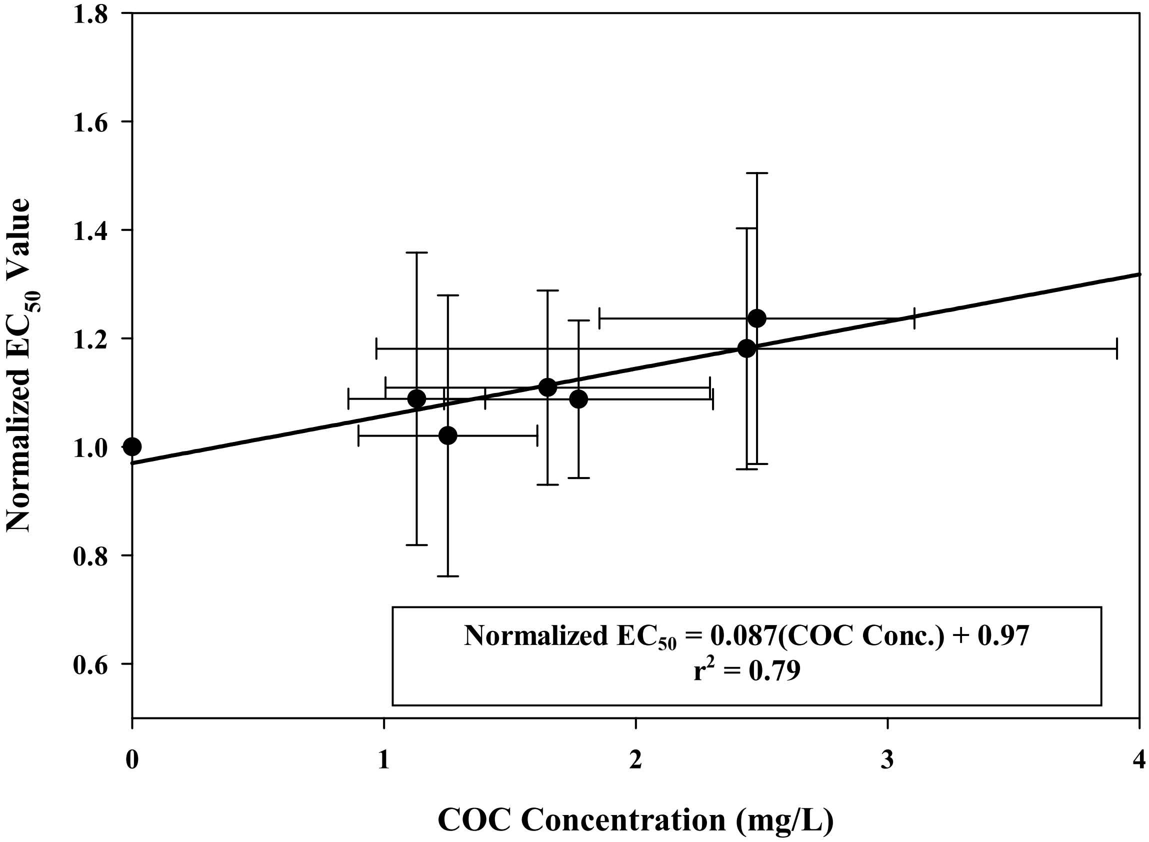

Figure 6-5: Correlation between the average normalized EC_{50} values and average COC concentrations for a specific size fraction. Error bars represent the standard deviations.

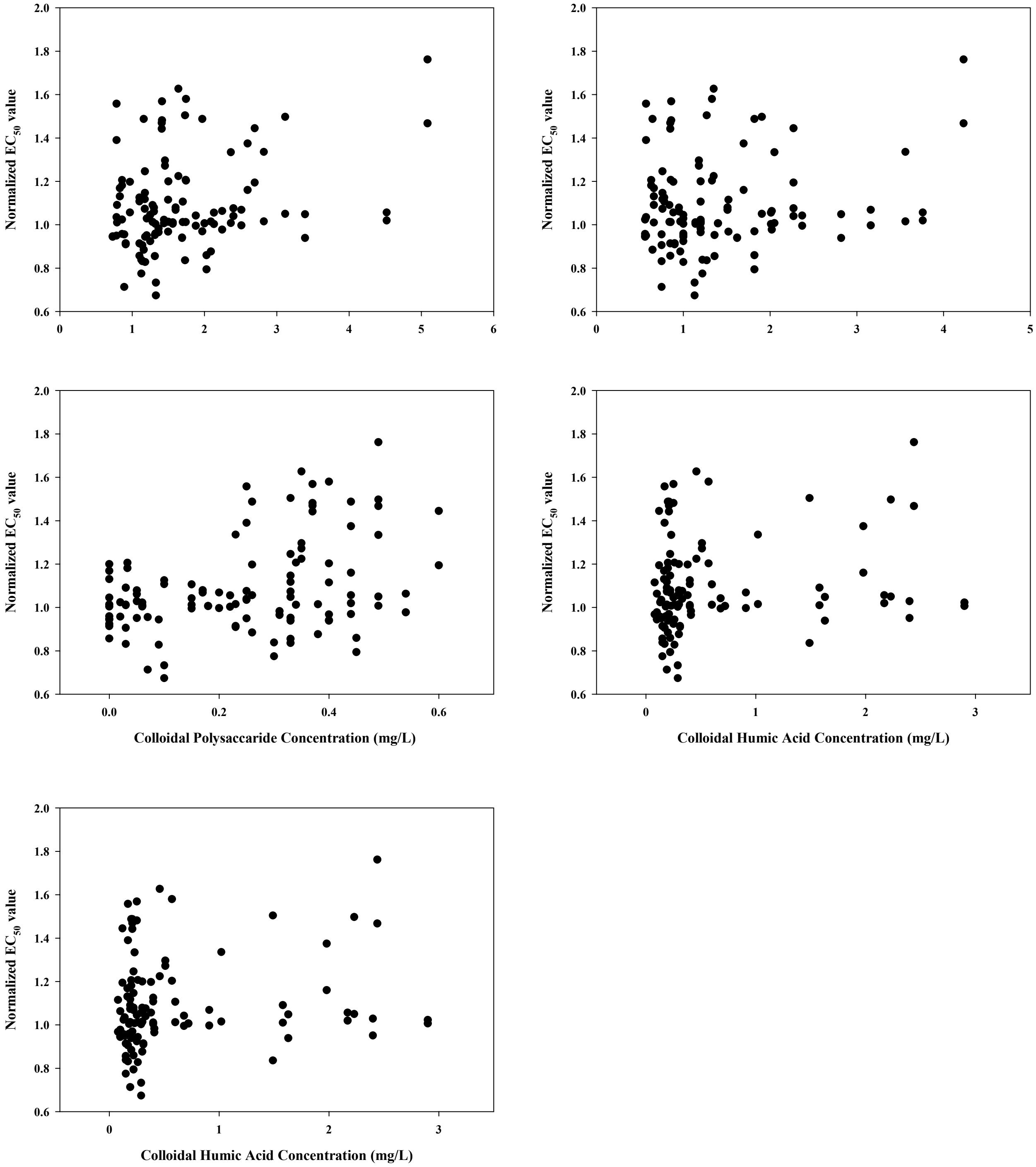

Figure 6-6 through 6-10:Relationship between the normalized EC_{50} value and colloidal organic carbon concentration (Figure 6-6, top left), protein (Figure 6-7, top right), polysaccharide (Figure 6-8, middle left), humic acid (Figure 6-9, middle right), and fulvic acid concentrations (Figure 6-10, bottom left).

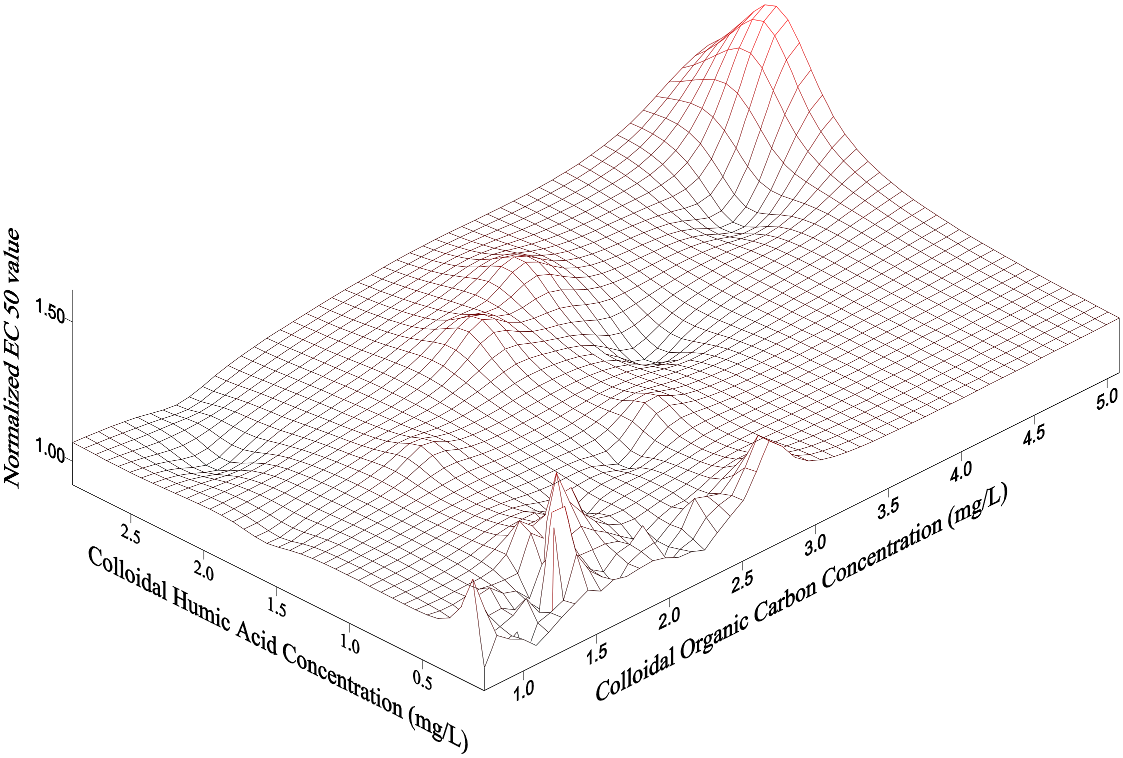

Figure 6-11: Relationship between the normalized EC_{50} value, colloidal humic acid concentration, and colloidal organic carbon concentration.

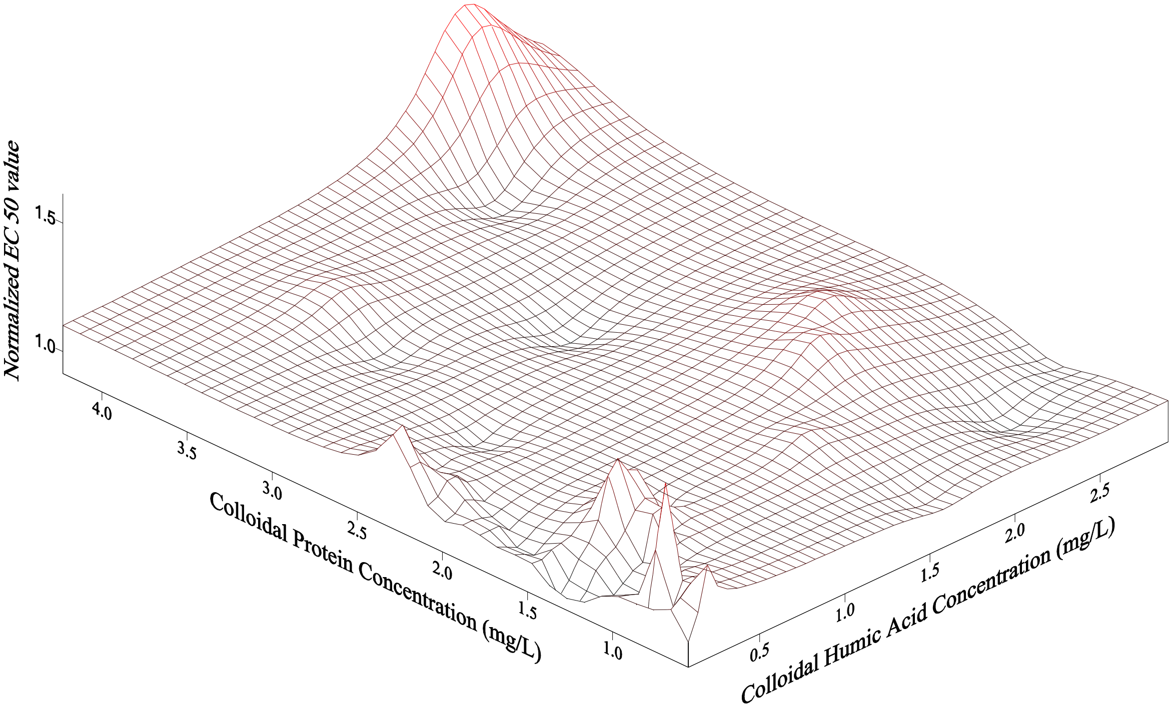

Figure 6-12: Relationship between the normalized EC_{50} value, colloidal humic acid concentration, and colloidal protein concentration.

Comparison Between Normalized EC_{50} Values and Colloidal Sorption Model. We recently demonstrated that the binding coefficient between E2 and COC in a size fractionated MLSS-derived colloidal matrix could be estimated by the following equation (38):

$$\text{Log } K_{coc} = 0.0042(e_{280}) + 3.78 \qquad (2)$$

Furthermore, the distribution of E2 between the dissolved and colloidal phase can be estimated by (57):

$$\text{Colloid-Associated E2} = \frac{[COC](K_{coc})}{1 + [COC](K_{coc})} \qquad (3)$$

Using the e_{280} coefficients and COC concentrations from Table 6-1, the amount of colloid-associated E2 can be calculated from equations 2 and 3. For our samples, we estimated that between 10 and 40% of the E2 would be associated with COC. However, this estimate is highly dependent upon the size fraction (i.e., e_{280} coefficient) and COC concentration and therefore was variable among the different sampling periods. A linear regression between the normalized EC_{50} values and the calculated colloid-associated E2 fraction yielded a significant slope ($p = 0.0034$) but a very weak correlation ($r^2 = 0.07$) (Figure 6-13).

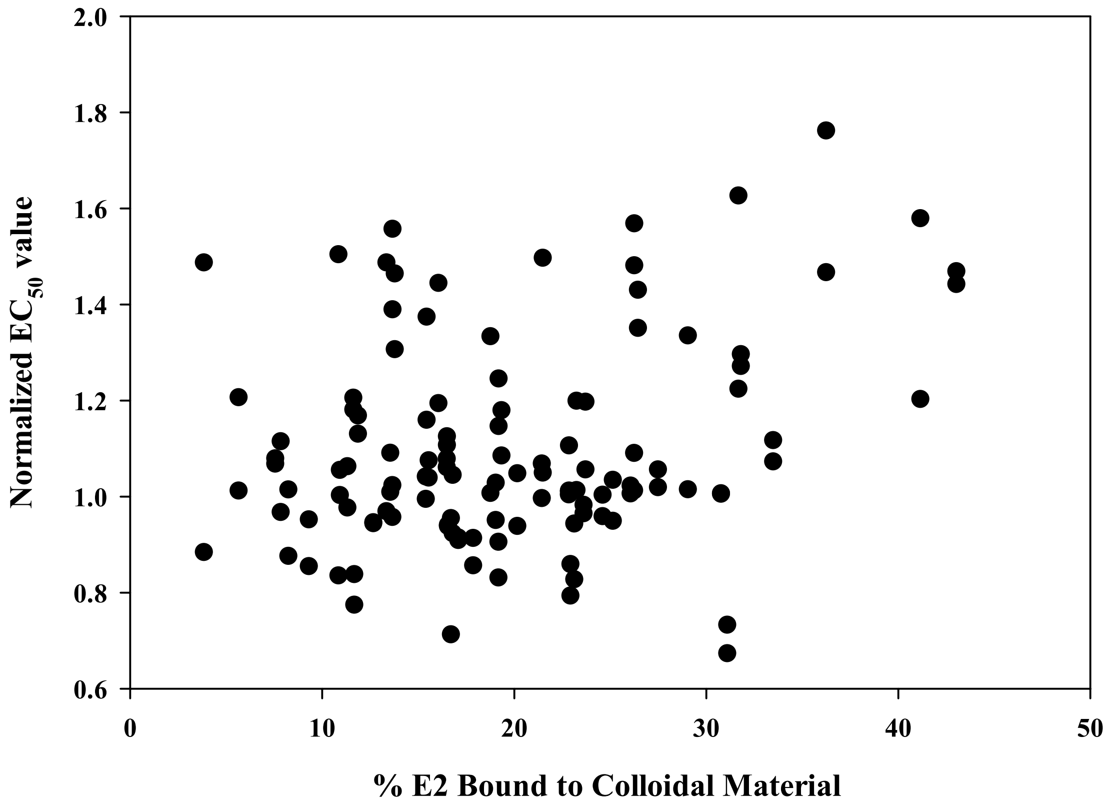

Figure 6-13: Relationship between the normalized EC_{50} value and estimated fraction of colloidal bound E2.

Discussion

The increase in EC_{50} values for a given E2 concentration in the presence of COC may be caused to one or more of the following factors including: (i) increased toxicity to the yeast culture caused by unknown organic pollutants in the wastewater samples; (ii) anti-estrogenic (antagonist) activity or inhibition of YES bioassay caused by unknown organic pollutants present in the wastewater samples; (iii) interference of the hER binding sites by colloidal material in the wastewater samples, or; (iv) reduced bioavailability of E2 caused by sorption to colloidal material. Each of these is considered in the following discussion.

Increased Toxicity. Yeast toxicity can be identified by the presence of clear yellow (or white) microtiter plate wells (33), and should be most expected at the higher

COC concentrations (22). In our experiments, we observed a red color in wells containing E2-spiked samples indicating production of ß-gal, and an opaque yellow color in the wells containing the negative control samples. The red color was reduced in a dose-dependent manner consistent with serial dilutions of an uninhibited bioassay (31,34). Furthermore, the absorbance (measured at 620 nm) of the positive control was not significantly different than the absorbance of the experimental systems ($p < 0.05$). Subsequently, the shift in the dose-response curves is not believed to be caused by toxicity to the yeast cells. Furthermore, the presence of a dissolved toxin would have been detected by the < 1 kD fraction, and corrected for in the normalization procedure.

Anti-estrogenic (antagonist) or inhibitory compounds. The comparatively large binding cavity of the hER makes it susceptible to both activation and inhibition by a wide and structurally diverse set of organic compounds (58,59). Anti-estrogenic compounds are able to mask the estrogenic potency of other compounds by interfering with the hER while inhibitory compounds can reduce the sensitivity of the YES bioassay either directly (binding to or modifying the structure of the hER) or indirectly (inhibition of β-gal production). Some individual anti-estrogenic compounds have been identified and include polyaromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs) and various phytochemicals (60-62). Both anti-estrogenic and inhibitory compounds may be present in wastewater effluents (34,63) and therefore could potentially contribute to increased EC_{50} values of the dose-response curves. However, anti-estrogenic and/or inhibitory compounds must first be bioavailable to the yeast culture suggesting their presence in the dissolved phase (< 1 kD permeate) is a prerequisite for toxic action. Subsequently, any anti-estrogenic or inhibitory compound would have been present in equal concentrations in all size fractions and therefore no change in EC_{50} values would have been observed. In comparison, the present experiments revealed a statistically significant impact of COC concentration on the normalized EC_{50} values. Moreover, ultrafilter membranes sorb substantial concentrations (> 90%) of nonpolar compounds (41) and, consequently, a significant reduction in yeast sensitivity would not be expected for the colloid size fractions below 100 kD. In contrast, the results of the present study indicate an increase in normalized EC_{50} values with some of the ultrafiltered samples (Figure 6-6). Furthermore, all EC_{50} values were normalized by the < 1 kD EC_{50} value which would, in effect, normalize for the presence of dissolved toxic compound. Therefore, the increase in EC_{50} values is unlikely to be caused by anti-estrogenic or inhibitory compounds.

Direct interference of hER by colloidal material. It is possible for colloidal material to bind with the hER and, in effect, become a YES bioassay antagonist. Since the hER are located intracellularly, colloidal material would first need to be transported across the cell membrane prior to binding with the hER. Macromolecules with molecular weights less than 1000 kD are able to directly pass through the cell membrane via diffusion or passive diffusion (64) while larger molecules first require hydrolysis prior to cell membrane transport. Subsequently, changes in the dose-response curve due to the antagonistic action of the colloidal material should increase with smaller size fractions. Although multiple regression analysis indicated no significant impact of colloidal size on the normalized EC_{50} values, all 3D surface graphs (Figures 6-12 and 6-13) indicate an increase in EC_{50} values at small size fractions and low concentrations of COC, protein and HA. The data suggests that antagonistic activity to the YES bioassay is minimal for the larger colloidal material, most likely caused by the inability of transporting large molecules across the cell membrane. However, direct interference of the hER with smaller colloidal material cannot be completely ruled out.

Reduction in bioavailability of E2 caused by sorption to colloidal material. The bioavailability and toxic action of organic compounds is intimately related to the presence of and affinity for colloidal organic material. Many investigators have shown reduced toxicity of organic pollutants in the presence of dissolved organic matter from naturally derived systems (65,66). The presence of colloidal material, whether natural or wastewater-derived, would appear to be most critical for environmental endocrine disruptors since the mechanism of disruption is based on binding (or lack thereof) to molecular receptors. Logically, organic compounds such as E2 that are bound to colloidal material will not be able to interact with the hER due to lack of recognition by the hER for the COC-E2 ligand (22).

The results of this study indicate that the single most important factor in increasing normalized EC_{50} values is COC concentration. Our results are in agreement with other investigators using conventional *in vivo* assays (i.e., *Daphnia magna*) (66,67), who reported that increasing dissolved organic matter (DOM) concentrations resulted in a concomitant reduction in the bioavailability of toxic organic compounds. Similarly, Tanghe *et al.* (22) observed a substantial increase in the EC_{50} value of a YES bioassay dose-response curve when samples contained both E2 and 150 mg/L of HA. Although our samples contained much lower concentrations of COC (1 – 5 mg/L) compared to the Tanghe *et al.* (22) study, we did observed statistically significant increases in EC_{50} values. Therefore, we attribute the increase in EC_{50} values of the dose-response curves to a reduction in bioavailability of E2 caused by sorption to colloidal organic material.

This experiment demonstrated that normalized EC_{50} values using settled MLSS-derived COC material as a sorbent for reducing E2 bioavailability are highly variable (Figure 6-5). Such variability may be caused by differences in the composition and character of the colloidal material or from the sample matrix itself. We were not able to accurately correlate the reduction in E2 bioavailability to wastewater relevant concentrations of COC, protein, polysaccaride, HA or FA (Figures 6-7 through 6-11) or to the E2 distribution between the colloidal and dissolved phases (Figure 6-13). The lack of correlation may be caused by dilution of the E2-COC sample by the high ionic strength yeast growth media. In high ionic strength solutions, COC will fold upon themselves to protect their hydrophobic domains (68). Although binding of E2 to COC occurs by π-electrons interactions (37,69) and hydrogen bonding between phenolic groups (69) instead of hydrophobic partioning, the structural modifications in COC caused by changes in ionic strength could reduce the accessibility of E2 to preferred bonding sites. Subsequently, there is a higher fraction of dissolved, and therefore bioavailable, E2 than estimated from the calculations. However, it is also possible that colloid-associated E2 elicits a YES response.

The statistical insignificance of size fraction on E2 bioavailability is caused by an increase in normalized EC_{50} values for the < 10kD size fractions. There are two possible caused for the observed behavior. First, COC material that is readily transported across the cell membrane can act as a hER antagonist by direct interference. This phenomenon would appear as a decrease in E2 bioavailability, but is caused by the colloids themselves itself rather than the association between E2 and COC. Secondly, free macromolecules and small colloidal material will aggregate more quickly than larger colloids (70) resulting in a colloidal suspension that contains material larger than the ultrafilter pore size. Additionally, the impact of the yeast growth media may also promote aggregation of the smaller size colloids. A multiple regression analysis of the dataset without the < 10 kD size fraction indicates that COC concentration ($p = 0.052$), colloidal size ($p = 0.0035$) and the interaction between COC and colloidal size ($p = 0.036$) are significant factors in determining the reduction in E2 bioavailability. Therefore, results from YES bioavailability experiments conducted with samples containing macromolecules or small colloids transported must be treated carefully.

The results of this study suggest that the YES bioassay can be used to quantitatively evaluate the bioavailability of colloid-associated E2 at high COC concentrations and large size fractions (greater than 10 kD). However, the potential for dose-response interferences must be evaluated closely for each matrix.

Conclusions

To our knowledge, this is the first attempt at using the YES bioassay to evaluate the role of wastewater-derived colloidal organic material in reducing the ability of E2 to bind and activated the hER. In hindsight, a more reductionist approach using surrogate NOM compounds such as commercially available HA and FA, although likely to be structurally and compositionally unrelated to wastewater derived COC, may have yielded more definitive correlations regarding the impact of colloidal composition and size on E2 bioavailability. Moreover, the use of wastewater samples severely limited the COC concentrations tested to 1 – 5 mg/L. While 1 – 5 mg/L of COC may be environmentally relevant for some natural systems, eutrophic freshwater systems and highly productive marine systems may posses 2 to 10 times this amount of colloidal material (71) suggesting an even greater reduction in E2 bioavailability caused by colloidal sorption. Based on the results of this study, we can concluded the following:

- After multivariate outlier analysis and normalization, the EC_{50} values generated by the YES bioassay had significant variability in the presence of wastewater derived COC. The complex wastewater matrix may cause a portion of this variability.
- Increases in normalized EC_{50} values were attributed to reduced E2 bioavailability due to E2 association with colloidal material.

- Relatively weak correlations ($r^2 < 0.22$) between the normalized EC_{50} values and COC, protein, polysaccaride, HA and FA concentrations and estimated E2-colloid association may be caused by the high ionic strength yeast growth media. However, it is also possible that colloid-associated E2 elicits a YES response.
- When using the YES bioassay to determine bioavailability, samples should not contain appreciable amounts of small molecular weight (< 10 kD) colloidal material. Such material may be directly transported across the cell membrane and act as a hER antagonist.
- The YES bioassay can be used as a quantitative tool to assess the bioavailability of colloid-associated contaminants. However, matrix effects of the specific environment being tested need to be closely evaluated due to the sensitivity of the hER and reporter plasmid.

Acknowledgements

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Chapter 7. Engineering Significance

There are several results of this research that should be of interest to the engineering community. First, the majority of estrogenic activity discharged from biological wastewater treatment facilities is through the treated effluent and not through the waste solids (WAS). Although estrogenic endocrine disrupting compounds (EDCs) are expected to sorb onto the particulate phase and therefore be removed via waste activated sludge (WAS), the WAS flowrate is relatively small compared to the effluent flowrate (between 5 – 10%) and therefore does not represent a significant source of EDC removal from the aqueous phase. Consequently, treatment strategies designed to optimize EDC removal should concentrate on the liquid streams. During preliminary studies with digested sludges, overall estrogenic activity in anaerobic digestors increased at detention times less than 40 days throughout the process while it increased, then decreased during aerobic digestion at detention times greater than 40 days, suggesting that the digestion and solids handling processes can be optimized for EDC degradation.

Second, the colloidal phase comprises a significant fraction of < 1.5 µm organic carbon from settled mixed liquor suspended solids, averaging 69% (± 5%) for the samples analyzed in these experiments. Although the settling method used in this research was not designed to mimic conditions within a secondary clarifier, full-scale treatment plants using secondary clarifiers for liquid-solid separation can be expected to have a similar dominance of colloidal organic carbon (COC) in their treated effluent compared to the dissolved phase. Consequently, the majority of effluent organic carbon may be susceptible to removal by wastewater processes such as coagulation, flocculation, and filtration. The organic colloidal material may also be removed from the water column in the receiving streams through natural flocculation and sedimentation. The impact of water chemistry and inorganic species, such as divalent cation and metal concentrations, is expected to impact the rate of colloidal flocculation (e.g., 1-3) and may dictate effluent quality.

Third, COC derived from biological wastewater treatment facilities has the capacity to bind trace organic contaminants contained in the dissolved phase. Therefore, treatment facilities that promote the capture of COC will have lower effluent concentrations of organic contaminants and, consequently, a lower detrimental impact on the water quality of the receiving stream. Wastewater treatment facilities that are engaged in direct water reuse practices should include a colloidal capture process to reduce human exposure and health risk. The capacity of COC to sorb to organic contaminants is dependent on the nature of the colloidal material (e.g., aromaticity), suggesting that the activated sludge process or processes downstream (such as chlorination) may be optimized to increase or decrease the colloidal affinity for organic contaminants. This area requires more research for implementation in full-scale facilities, but is critical to developing cost effective removal alternatives for organic contaminants that are either recalcitrant to microbial degradation or have concentrations significantly lower than their half-saturation coefficient.

Lastly, the presence or absence of COC will dictate the bioavailability of EDCs, suggesting that water quality of receiving streams may dictate whether or not both *in vitro* and *in vivo* bioassays can be used to accurately predict environmental exposure to and associated risk of organic contaminants in the environment. This fact appears most critical for the popular *in vitro* assessment of estrogenic activity by measuring vitellogenin production in male fish (e.g., 4-6). Clearly, water quality parameters such as turbidity, total and dissolved organic carbon concentrations, and aromaticity of the colloidal phase should be monitored during these experiments to quantify the bioavailability of the organic contaminants. Significantly, a sample or ecosystem that produces a reduced or non-significant response in the bioassay should not be regarded as "contaminant-free" until representative samples are confirmed by traditional analytical chemistry methods such as gas or liquid chromatography/mass spectroscopy.

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Appendix A (Chapter 3).
Estrogen Receptor Agonist Fate during Wastewater and Biosolids Treatment Processes: A Mass Balance Analysis

Mass Balances for Facility A, B, and C - Averages			
Facility A	**Average E2 Conc. (M)**	**Stand. Dev.**	**Average E2 Conc. (ng/L)**
Raw Influent	6.56167E-11	1.09825E-10	17.84774472
Raw Influent (sonicated)	1.94447E-10	2.48484E-10	52.88951067
Primary Effluent	6.80221E-11	5.11886E-11	18.50200791
Primary Effluent (sonicated)	5.25248E-11	8.11292E-11	14.28675527
Pilot - Permeate	1.82E-11	2.21878E-11	4.956499016
PAC effluent (sonicated)	1.63E-11	2.70482E-11	4.434082062
GAC effluent (sonicated)	2.14E-11	2.79581E-11	5.809681214
MLSS - 6th stage pilot	2.98E-10	2.4038E-10	81.02949233
Mass Balance Calculations			
Influent Flow (gpd)	20,162		
Effluent Flow (gpd)	19,842		
WAS (gpd)	320		
MLSS Concentration (mg/L)	9,600		
Solid-based estrogenic activity (M)	2.80E-10		
Facility B			
Raw Influent	6.56167E-11	1.09825E-10	17.84774472
Raw Influent (sonicated)	1.94447E-10	2.48484E-10	52.88951067
Primary Effluent	6.80221E-11	5.11886E-11	18.50200791
Primary Effluent (sonicated)	5.25E-11	8.11292E-11	14.28675527
Secondary Effluent	2.63328E-11	4.83677E-11	7.162518683
Secondary Effluent (sonicated)	2.49592E-11	2.40479E-11	6.788902974
MLSS - conventional	5.89705E-10	8.91714E-11	160.3996361
Digester Feed - Sludge	9.67E-10	8.89831E-11	263.0363609
Primary Digester - Sludge	1.56E-09	5.69328E-10	423.4591312
Secondary Digester - Sludge	1.91458E-09	2.69715E-10	520.7652745
Digester Feed centrate - Sludge	1.90E-09	2.69715E-10	518.0452745
Primary Digester centrate - sludge	6.62838E-11	6.41541E-11	18.02919619
Secondary Digester centrate - sludge	5.05459E-11	6.70918E-11	13.74849743

Mass Balance Calculations			
Influent Flow (gpd)	3,235,428		
Effluent Flow (gpd)	2,855,288		
WAS (gpd)	51,205		
MLSS Concentration (mg/L)	4,360		
Digester Feed Solids Concentration (mg/L)	13,100		
Primary Digester Solids Concentration (mg/L)	16,840		
Secondary Digester Solids Concentration (mg/L)	8,900		
Digester Volume (L) - each	425,480		
Digester Volume (gallons) - each	112,412		
Volume Transferred (gallons/day)	5,621		
Solid-Based E2 Activity (M)			
Digester Feed	9.40713E-10		
Primary Digester Solids	1.49055E-09		
Secondary Digester Solids	1.86403E-09		
Facility C			
Raw Influent	8.78669E-11	8.09379E-11	23.89980422
Secondary Effluent	3.89381E-11	5.92252E-11	10.5911564
MLSS	7.82833E-11	1.87256E-11	21.29306097
Digester Feed	7.11686E-10	1.4337E-10	193.5784909
Digester 1	7.21386E-10	1.24687E-10	196.2169113
Digester 2	1.16219E-09	1.09264E-10	316.1160771
Digester 3	1.54127E-09	1.81892E-10	419.2263388
DSHT 1	4.56416E-09	1.62128E-09	1241.450208
DSHT 2	3.01476E-09	8.00067E-10	820.0153213
ATAD Feed - Centrate	2.22778E-10	4.03379E-10	60.59559974
ATAD Digester 1 Centrate	1.05806E-10	4.63756E-11	28.77916798
ATAD Digester 2 Centrate	1.12073E-10	8.47938E-11	30.48392583
ATAD Digester 3 Centrate	9.09167E-11	6.40534E-11	24.72934296
DSHT 1 Centrate	5.64068E-10	1.37326E-09	153.4263649
DSHT 2 Centrate	6.46634E-10	1.59407E-09	175.8844155
Mass Balance Calculations			
Influent Flow (gpd)	5,878,093		

Effluent Flow (gpd)	5,731,945		
Total Activated Sludge Volume (gallons)	3,946,000		
SRT (days)	9		
WAS (gpd)	146,148		
Feed Sludge Transferred (gpd)	17,760		
MLSS (mg/L)	3,360		
WAS (mg/L)	10,080		
Digester Feed (mg/L)	43,660		
Digester 1 (mg/L)	39,540		
Digester 2 (mg/L)	37,140		
Digester 3 (mg/L)	36,020		
DSHT 1 (mg/L)	28,580		
DSHT 2 (mg/L)	27,180		
Solid Based E2 Activity			
MLSS	3.93452E-11		
Digester Feed	4.88908E-10		
Digester 1	6.1558E-10		
Digester 2	1.05012E-09		
Digester 3	1.45036E-09		
DSHT 1	4.00009E-09		
DSHT 2	2.36813E-09		

Facility A and B – March, 2001
Plate 1 (top) – Absorbance at 540 nm
Plate 2 (bottom) – Absorbance at 690 nm
Columns 1 and 2 – Whole Sample
Columns 11 and 12 – Sonicated Sample

Raw Influent	1	2	3	4	5	6	7	8	9	10	11	12
A	1.371	1.485	0.041	0.041	0.04	0.04	0.041	0.042	0.041	0.039	1.366	1.527
B	1.4	1.403	0.04	0.04	0.04	0.041	0.041	0.04	0.042	0.039	1.496	1.557
C	1.397	1.466	0.039	0.047	0.039	0.038	0.039	0.047	0.04	0.039	1.438	1.547
D	1.442	1.505	0.039	0.039	0.04	0.044	0.039	0.053	0.04	0.038	1.554	1.529
E	1.512	1.543	0.041	0.042	0.04	0.041	0.041	0.041	0.041	0.039	1.557	1.654
F	1.56	1.635	0.044	0.045	0.039	0.039	0.04	0.04	0.041	0.039	1.639	1.614
G	1.569	1.631	0.039	0.04	0.04	0.039	0.041	0.041	0.044	0.039	1.602	1.569
H	1.465	1.58	0.042	0.04	0.039	0.039	0.04	0.041	0.041	0.045	1.561	1.568
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.709	0.679	0.041	0.041	0.04	0.04	0.041	0.041	0.04	0.039	0.601	0.695
B	0.677	0.651	0.04	0.04	0.04	0.04	0.04	0.04	0.041	0.039	0.692	0.761
C	0.673	0.722	0.039	0.046	0.039	0.038	0.039	0.045	0.04	0.039	0.661	0.778
D	0.666	0.716	0.039	0.039	0.04	0.044	0.039	0.05	0.04	0.038	0.718	0.786
E	0.731	0.727	0.04	0.041	0.04	0.04	0.041	0.04	0.04	0.039	0.726	0.801
F	0.748	0.754	0.043	0.044	0.039	0.039	0.04	0.04	0.041	0.039	0.722	0.76
G	0.776	0.772	0.039	0.04	0.04	0.039	0.04	0.041	0.043	0.038	0.721	0.774
H	0.688	0.755	0.041	0.04	0.039	0.04	0.04	0.04	0.041	0.045	0.707	0.794

Primary Effluent	1	2	3	4	5	6	7	8	9	10	11	12
A	1.326	1.506	0.04	0.041	0.039	0.039	0.042	0.043	0.04	0.039	1.422	1.466
B	1.414	1.57	0.045	0.041	0.039	0.039	0.04	0.04	0.04	0.039	1.492	1.524
C	1.485	1.53	0.039	0.039	0.038	0.038	0.039	0.039	0.039	0.038	1.565	1.598
D	1.524	1.624	0.039	0.039	0.038	0.039	0.039	0.039	0.04	0.038	1.595	1.579
E	1.519	1.618	0.04	0.04	0.039	0.041	0.042	0.041	0.04	0.039	1.601	1.51
F	1.505	1.623	0.04	0.04	0.039	0.039	0.041	0.04	0.041	0.039	1.637	1.568
G	1.552	1.547	0.039	0.04	0.039	0.038	0.04	0.04	0.039	0.039	1.56	1.56
H	1.504	1.564	0.04	0.04	0.038	0.038	0.039	0.041	0.041	0.038	1.467	1.706
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.573	0.695	0.04	0.04	0.039	0.039	0.042	0.043	0.04	0.039	0.71	0.713
B	0.624	0.722	0.043	0.04	0.04	0.039	0.04	0.04	0.04	0.039	0.718	0.757
C	0.674	0.711	0.039	0.039	0.038	0.038	0.039	0.039	0.039	0.038	0.74	0.803
D	0.68	0.774	0.039	0.039	0.038	0.039	0.039	0.039	0.04	0.038	0.797	0.791
E	0.675	0.739	0.04	0.04	0.039	0.041	0.041	0.04	0.04	0.039	0.757	0.809
F	0.668	0.753	0.04	0.04	0.039	0.039	0.041	0.04	0.04	0.039	0.79	0.772
G	0.708	0.732	0.039	0.04	0.039	0.038	0.039	0.04	0.039	0.039	0.735	0.805
H	0.65	0.685	0.04	0.04	0.039	0.039	0.039	0.041	0.04	0.039	0.671	0.819

Secondary Effluent Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.238	1.257	0.04	0.043	0.04	0.042	0.042	0.042	0.042	0.041	1.237	1.417
B	1.289	1.43	0.041	0.04	0.04	0.04	0.043	0.041	0.042	0.039	1.425	1.282
C	1.388	1.496	0.04	0.04	0.039	0.039	0.039	0.04	0.04	0.039	1.267	1.41
D	1.378	1.402	0.039	0.039	0.038	0.039	0.04	0.04	0.04	0.039	1.496	1.468
E	1.43	1.471	0.041	0.042	0.04	0.041	0.041	0.041	0.045	0.04	1.509	1.483
F	1.436	1.479	0.04	0.041	0.041	0.04	0.041	0.041	0.041	0.04	1.501	1.474
G	1.477	1.514	0.04	0.04	0.039	0.039	0.04	0.041	0.041	0.039	1.317	1.43
H	1.462	1.422	0.041	0.04	0.039	0.039	0.044	0.042	0.041	0.039	1.478	1.495
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.564	0.556	0.04	0.043	0.04	0.041	0.042	0.041	0.042	0.04	0.581	0.65
B	0.578	0.636	0.041	0.04	0.04	0.04	0.042	0.041	0.042	0.039	0.638	0.624
C	0.618	0.664	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.567	0.644
D	0.588	0.603	0.04	0.039	0.039	0.039	0.04	0.04	0.041	0.039	0.684	0.645
E	0.629	0.688	0.04	0.041	0.04	0.041	0.041	0.041	0.044	0.04	0.726	0.678
F	0.614	0.658	0.04	0.041	0.041	0.04	0.041	0.041	0.041	0.04	0.704	0.666
G	0.659	0.712	0.04	0.04	0.039	0.039	0.04	0.041	0.041	0.039	0.527	0.665
H	0.66	0.604	0.041	0.04	0.039	0.039	0.043	0.042	0.041	0.039	0.655	0.663

Permeate - Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.332	1.306	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.039	1.221	1.326
B	1.383	1.403	0.042	0.041	0.04	0.039	0.04	0.04	0.041	0.039	1.384	1.465
C	1.439	1.441	0.039	0.039	0.038	0.038	0.043	0.039	0.039	0.038	1.419	1.486
D	1.496	1.469	0.042	0.042	0.039	0.038	0.04	0.04	0.043	0.038	1.527	1.545
E	1.486	1.498	0.043	0.041	0.04	0.041	0.041	0.041	0.04	0.039	1.603	1.59
F	1.48	1.553	0.04	0.041	0.04	0.04	0.041	0.043	0.043	0.04	1.591	1.623
G	1.414	1.467	0.039	0.04	0.039	0.039	0.042	0.041	0.04	0.038	1.647	1.595
H	1.518	1.543	0.04	0.041	0.038	0.039	0.042	0.041	0.041	0.039	1.682	1.641
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.565	0.539	0.04	0.04	0.039	0.039	0.04	0.041	0.04	0.039	0.48	0.525
B	0.549	0.576	0.041	0.04	0.04	0.039	0.04	0.04	0.04	0.039	0.518	0.61
C	0.561	0.591	0.039	0.039	0.038	0.038	0.043	0.039	0.039	0.038	0.561	0.615
D	0.595	0.598	0.041	0.041	0.039	0.038	0.039	0.039	0.042	0.039	0.59	0.64
E	0.576	0.606	0.042	0.04	0.039	0.04	0.04	0.04	0.04	0.039	0.664	0.635
F	0.575	0.657	0.04	0.04	0.04	0.039	0.04	0.043	0.042	0.04	0.626	0.674
G	0.626	0.588	0.039	0.04	0.039	0.039	0.042	0.04	0.04	0.038	0.648	0.641
H	0.638	0.649	0.039	0.04	0.039	0.039	0.041	0.04	0.04	0.039	0.682	0.698

GAC effluent Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.419	1.39	0.041	0.041	0.04	0.04	0.041	0.042	0.045	0.04	1.269	1.478
B	1.362	1.388	0.041	0.041	0.041	0.042	0.041	0.041	0.041	0.04	1.346	1.418
C	1.37	1.451	0.04	0.041	0.04	0.038	0.04	0.04	0.039	0.038	1.537	1.461
D	1.448	1.428	0.04	0.04	0.039	0.039	0.04	0.04	0.041	0.039	1.564	1.432
E	1.519	1.517	0.04	0.041	0.04	0.041	0.041	0.041	0.041	0.039	1.485	1.517
F	1.401	1.462	0.04	0.041	0.04	0.04	0.041	0.041	0.041	0.039	1.366	1.553
G	1.522	1.448	0.04	0.04	0.039	0.039	0.04	0.041	0.04	0.039	1.281	1.502
H	1.48	1.473	0.04	0.04	0.039	0.039	0.04	0.041	0.041	0.039	1.526	1.544
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.663	0.574	0.041	0.041	0.04	0.04	0.041	0.042	0.045	0.04	0.524	0.645
B	0.574	0.583	0.04	0.041	0.041	0.042	0.041	0.041	0.041	0.04	0.574	0.649
C	0.564	0.616	0.04	0.042	0.04	0.039	0.04	0.04	0.04	0.039	0.719	0.643
D	0.607	0.599	0.04	0.04	0.04	0.039	0.04	0.04	0.04	0.039	0.697	0.646
E	0.637	0.663	0.04	0.041	0.04	0.041	0.041	0.041	0.04	0.04	0.687	0.692
F	0.606	0.638	0.04	0.041	0.04	0.04	0.041	0.041	0.041	0.04	0.578	0.713
G	0.639	0.63	0.04	0.04	0.039	0.039	0.04	0.041	0.04	0.039	0.493	0.688
H	0.566	0.611	0.04	0.04	0.04	0.039	0.04	0.041	0.041	0.04	0.707	0.694

PAC effluent Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.327	1.48	0.04	0.042	0.04	0.04	0.041	0.041	0.04	0.04	1.276	1.399
B	1.382	1.393	0.04	0.04	0.04	0.04	0.04	0.04	0.041	0.039	1.523	1.533
C	1.411	1.445	0.039	0.039	0.038	0.038	0.039	0.039	0.039	0.038	1.515	1.53
D	1.383	1.512	0.039	0.039	0.038	0.038	0.039	0.039	0.04	0.038	1.565	1.555
E	1.46	1.528	0.04	0.045	0.04	0.041	0.04	0.04	0.047	0.039	1.632	1.563
F	1.598	1.575	0.04	0.04	0.039	0.039	0.041	0.041	0.041	0.045	1.557	1.622
G	1.606	1.579	0.039	0.039	0.038	0.039	0.04	0.04	0.04	0.038	1.632	1.618
H	1.464	1.577	0.04	0.039	0.038	0.038	0.044	0.041	0.041	0.038	1.558	1.661
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.581	0.621	0.04	0.042	0.04	0.041	0.041	0.041	0.041	0.04	0.552	0.629
B	0.564	0.585	0.04	0.04	0.04	0.041	0.04	0.041	0.041	0.04	0.619	0.644
C	0.585	0.633	0.04	0.04	0.039	0.039	0.04	0.04	0.039	0.039	0.64	0.64
D	0.513	0.633	0.04	0.039	0.039	0.039	0.04	0.04	0.04	0.039	0.669	0.655
E	0.607	0.647	0.04	0.044	0.04	0.041	0.04	0.04	0.046	0.04	0.706	0.706
F	0.684	0.671	0.04	0.04	0.04	0.039	0.041	0.041	0.041	0.044	0.696	0.744
G	0.687	0.688	0.04	0.04	0.039	0.04	0.04	0.04	0.04	0.039	0.755	0.725
H	0.619	0.717	0.04	0.04	0.039	0.039	0.045	0.041	0.041	0.039	0.655	0.718

MLSS – Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.006	1.122	0.045	0.041	0.042	0.04	0.041	0.041	0.04	0.039	1.246	1.128
B	1.18	1.236	0.04	0.04	0.04	0.039	0.042	0.04	0.041	0.039	0.993	1.098
C	1.146	1.164	0.039	0.039	0.038	0.039	0.043	0.039	0.04	0.038	1.175	1.082
D	1.12	1.187	0.043	0.039	0.038	0.038	0.039	0.039	0.04	0.038	1.039	1.157
E	1.194	1.236	0.041	0.04	0.039	0.041	0.04	0.04	0.04	0.039	1.25	1.174
F	1.15	1.236	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	1.105	1.284
G	1.222	1.135	0.04	0.039	0.038	0.038	0.039	0.04	0.039	0.039	1.26	1.236
H	1.112	1.308	0.04	0.044	0.038	0.038	0.041	0.04	0.041	0.038	1.375	1.383
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.577	0.547	0.043	0.04	0.041	0.04	0.04	0.041	0.04	0.039	0.636	0.621
B	0.619	0.592	0.04	0.04	0.039	0.039	0.042	0.04	0.04	0.039	0.521	0.622
C	0.622	0.558	0.039	0.039	0.038	0.039	0.042	0.039	0.04	0.038	0.636	0.626
D	0.59	0.584	0.042	0.039	0.038	0.038	0.039	0.039	0.04	0.038	0.513	0.619
E	0.641	0.604	0.041	0.04	0.039	0.04	0.04	0.04	0.039	0.039	0.686	0.65
F	0.651	0.638	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.581	0.697
G	0.623	0.645	0.04	0.039	0.038	0.038	0.039	0.04	0.039	0.039	0.685	0.683
H	0.632	0.618	0.039	0.044	0.039	0.039	0.041	0.04	0.04	0.038	0.644	0.64

MLSS - 2nd stage Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.069	1.094	0.04	0.041	0.039	0.04	0.041	0.041	0.043	0.04	1.16	1.137
B	1.028	1.12	0.04	0.042	0.04	0.039	0.041	0.041	0.041	0.039	1.162	1.117
C	1.039	1.116	0.039	0.039	0.038	0.038	0.039	0.04	0.039	0.038	1.174	1.209
D	1.069	0.973	0.04	0.047	0.042	0.038	0.04	0.04	0.041	0.038	1.212	1.189
E	1.065	1.061	0.04	0.041	0.04	0.041	0.041	0.041	0.04	0.039	1.109	1.293
F	1.088	0.931	0.04	0.04	0.039	0.039	0.04	0.041	0.041	0.039	1.383	1.446
G	1.155	1.161	0.039	0.04	0.038	0.038	0.04	0.04	0.04	0.038	1.325	1.364
H	1.101	1.239	0.04	0.04	0.038	0.038	0.039	0.04	0.041	0.038	1.369	1.368
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.642	0.625	0.04	0.041	0.039	0.039	0.04	0.04	0.043	0.039	0.74	0.688
B	0.599	0.663	0.04	0.041	0.04	0.039	0.04	0.04	0.04	0.039	0.727	0.675
C	0.617	0.648	0.039	0.039	0.038	0.038	0.039	0.04	0.039	0.038	0.704	0.682
D	0.647	0.53	0.04	0.045	0.041	0.038	0.04	0.04	0.04	0.038	0.739	0.692
E	0.651	0.598	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.039	0.75	0.669
F	0.667	0.509	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.785	0.728
G	0.712	0.694	0.039	0.039	0.038	0.038	0.039	0.04	0.039	0.038	0.807	0.727
H	0.657	0.708	0.039	0.039	0.038	0.039	0.039	0.04	0.04	0.038	0.705	0.711

MLSS - 6th stage Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.306	1.392	0.041	0.041	0.04	0.04	0.041	0.042	0.042	0.04	1.149	1.284
B	1.161	1.312	0.041	0.041	0.041	0.041	0.042	0.041	0.041	0.04	1.169	1.252
C	1.286	1.381	0.041	0.04	0.039	0.039	0.04	0.04	0.04	0.039		1.276
D	1.237	1.31	0.04	0.04	0.039	0.039	0.04	0.04	0.041	0.038	1.29	1.291
E		1.37	0.042	0.041	0.04	0.041	0.041	0.041	0.041	0.04	1.287	1.31
F	1.347	1.412	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.04	1.152	1.42
G	1.187	1.288	0.04	0.04	0.039	0.039	0.04	0.041	0.04	0.039	1.337	1.394
H	1.379	1.388	0.04	0.04	0.039	0.039	0.04	0.041	0.041	0.04	1.312	1.345
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.642	0.649	0.04	0.041	0.04	0.04	0.041	0.041	0.042	0.04	0.695	0.593
B	0.539	0.592	0.04	0.04	0.04	0.04	0.041	0.04	0.041	0.04	0.538	0.653
C	0.62	0.677	0.041	0.04	0.039	0.039	0.039	0.04	0.039	0.038	0.492	0.697
D	0.59	0.609	0.04	0.04	0.039	0.039	0.04	0.039	0.04	0.038	0.727	0.703
E	0.661	0.642	0.042	0.04	0.04	0.04	0.041	0.04	0.04	0.04	0.705	0.725
F	0.744	0.652	0.04	0.04	0.04	0.039	0.04	0.04	0.04	0.039	0.58	0.776
G	0.721	0.712	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.739	0.762
H	0.637	0.633	0.04	0.04	0.039	0.039	0.04	0.04	0.041	0.04	0.667	0.731

Digester B – supernatant Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	0.907	0.946	0.041	0.042	0.04	0.04	0.042	0.042	0.041	0.039	1.186	0.955
B	1.227	1.346	0.042	0.041	0.04	0.04	0.041	0.041	0.042	0.04	0.948	1.235
C	0.91	1.3	0.042	0.04	0.039	0.039	0.039	0.04	0.04	0.038	0.919	0.921
D	1.323	1.383	0.04	0.04	0.039	0.04	0.041	0.041	0.041	0.04	1.352	1.167
E	1.274	1.326	0.041	0.041	0.041	0.042	0.043	0.041	0.041	0.041	1.257	0.95
F	1.343	1.246	0.041	0.041	0.042	0.04	0.041	0.041	0.041	0.04	0.975	1.018
G	1.353	1.232	0.04	0.041	0.039	0.039	0.04	0.041	0.041	0.04	1.033	0.909
H	1.38	1.085	0.041	0.041	0.039	0.04	0.04	0.047	0.044	0.041	0.966	0.917
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.677	0.658	0.04	0.041	0.04	0.04	0.041	0.041	0.04	0.039	0.639	0.671
B	0.572	0.642	0.041	0.04	0.04	0.039	0.041	0.04	0.041	0.04	0.713	0.657
C	0.674	0.638	0.041	0.04	0.038	0.039	0.039	0.039	0.04	0.038	0.664	0.67
D	0.633	0.654	0.039	0.039	0.039	0.039	0.04	0.04	0.041	0.04	0.717	0.647
E	0.631	0.635	0.04	0.04	0.04	0.041	0.042	0.04	0.041	0.04	0.676	0.688
F	0.641	0.547	0.04	0.04	0.041	0.039	0.04	0.041	0.04	0.04	0.755	0.775
G	0.629	0.576	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.777	0.7
H	0.643	0.65	0.04	0.04	0.039	0.04	0.04	0.046	0.043	0.041	0.734	0.657

Digester B – sludge Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	0.949	1.282	0.042	0.041	0.04	0.04	0.041	0.041	0.041	0.04	1.267	1.146
B	1.276	0.895	0.041	0.045	0.04	0.04	0.041	0.04	0.041	0.039	1.34	1.316
C	0.953	1.271	0.04	0.04	0.039	0.04	0.039	0.04	0.039	0.038	1.364	1.284
D	1.391	1.013	0.039	0.041	0.039	0.039	0.04	0.04	0.039	0.038	1.316	1.307
E	0.99	1.273	0.042	0.042	0.042	0.042	0.042	0.041	0.04	0.039	1.285	1.311
F	1.363	1.354	0.04	0.042	0.04	0.04	0.041	0.042	0.04	0.039	1.054	1.27
G	1.436	1.37	0.04	0.041	0.04	0.038	0.04	0.04	0.04	0.038	1.307	1.435
H	1.187	1.415	0.042	0.041	0.04	0.043	0.04	0.04	0.04	0.039	1.46	1.496
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.71	0.722	0.041	0.041	0.04	0.04	0.041	0.04	0.04	0.04	0.702	0.684
B	0.658	0.695	0.041	0.045	0.04	0.039	0.04	0.04	0.04	0.039	0.72	0.675
C	0.73	0.595	0.04	0.04	0.039	0.04	0.039	0.039	0.039	0.038	0.709	0.629
D	0.642	0.676	0.039	0.04	0.039	0.039	0.04	0.039	0.039	0.038	0.715	0.616
E	0.765	0.636	0.042	0.041	0.041	0.041	0.041	0.04	0.04	0.039	0.623	0.668
F	0.658	0.654	0.04	0.041	0.04	0.04	0.04	0.041	0.04	0.039	0.808	0.623
G	0.711	0.683	0.04	0.04	0.04	0.038	0.039	0.04	0.039	0.038	0.694	0.798
H	0.787	0.706	0.041	0.041	0.04	0.042	0.039	0.039	0.04	0.039	0.812	0.793

Digester D – supernatant Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.328	1.248	0.041	0.04	0.039	0.04	0.041	0.048	0.04	0.039	1.221	1.206
B	1.284	1.251	0.042	0.04	0.039	0.039	0.04	0.04	0.041	0.039	1.213	1.258
C	0.936	1.29	0.04	0.039	0.038	0.038	0.039	0.04	0.039	0.038	1.093	1.135
D	0.946	1.349	0.041	0.039	0.038	0.039	0.039	0.039	0.04	0.038	1.302	1.332
E	1.415	1.428	0.04	0.04	0.039	0.041	0.041	0.04	0.04	0.039	1.1	1.282
F	1.47	1.468	0.042	0.041	0.041	0.039	0.04	0.04	0.041	0.039	1.338	1.338
G	1.494	1.398	0.04	0.04	0.038	0.038	0.04	0.04	0.039	0.038	1.406	1.369
H	1.462	1.269	0.039	0.039	0.038	0.039	0.039	0.041	0.041	0.038	1.162	1.375
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.586	0.565	0.041	0.041	0.04	0.04	0.041	0.049	0.041	0.04	0.637	0.878
B	0.578	0.585	0.042	0.041	0.04	0.04	0.041	0.041	0.041	0.04	0.63	0.637
C	0.685	0.586	0.04	0.04	0.039	0.039	0.04	0.041	0.04	0.039	0.52	0.522
D	0.693	0.595	0.042	0.04	0.039	0.039	0.04	0.04	0.041	0.039	0.658	0.648
E	0.648	0.63	0.041	0.041	0.04	0.041	0.041	0.041	0.041	0.04	0.828	0.677
F	0.665	0.688	0.042	0.041	0.041	0.039	0.041	0.041	0.041	0.04	0.699	0.703
G	0.699	0.663	0.04	0.041	0.039	0.039	0.04	0.04	0.04	0.039	0.788	0.743
H	0.633	0.51	0.04	0.04	0.039	0.04	0.04	0.042	0.041	0.04	0.8	0.721

Digester D – sludge Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.344	1.326	0.04	0.041	0.042	0.04	0.044	0.041	0.041	0.039	0.89	1.226
B	1.398	1.422	0.04	0.041	0.04	0.039	0.041	0.04	0.041	0.039	1.259	1.263
C	1.354	1.326	0.04	0.039	0.038	0.039	0.039	0.04	0.04	0.039	0.877	1.207
D	1.448	1.285	0.039	0.039	0.038	0.039	0.04	0.039	0.04	0.038	1.383	1.289
E	1.51	1.41	0.04	0.041	0.04	0.041	0.041	0.041	0.041	0.04	1.323	1.319
F	1.508	1.464	0.04	0.04	0.04	0.039	0.041	0.04	0.041	0.039	1.426	1.412
G	1.478	1.422	0.039	0.042	0.039	0.038	0.04	0.042	0.04	0.038	1.066	1.382
H	1.482	1.461	0.04	0.041	0.039	0.038	0.04	0.041	0.041	0.039	1.503	1.486
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.593	0.557	0.04	0.04	0.04	0.04	0.043	0.04	0.04	0.039	0.651	0.616
B	0.596	0.595	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.601	0.601
C	0.587	0.533	0.039	0.039	0.038	0.038	0.039	0.039	0.039	0.038	0.677	0.675
D	0.614	0.571	0.039	0.039	0.038	0.038	0.039	0.039	0.04	0.038	0.685	0.638
E	0.671	0.581	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.039	0.668	0.655
F	0.643	0.642	0.039	0.04	0.04	0.039	0.04	0.04	0.04	0.039	0.706	0.669
G	0.669	0.611	0.039	0.041	0.039	0.038	0.039	0.041	0.04	0.038	0.457	0.687
H	0.674	0.623	0.039	0.04	0.039	0.038	0.039	0.04	0.04	0.039	0.715	0.733

Estradiol Control	1	2	3	4	5	6	7	8	9	10	11	12
A	2.063	2.111	2.08	1.918	1.429	1.051	0.934	0.885	0.884	0.849	0.876	0.841
B	2.139	2.142	2.116	1.874	1.49	1.069	0.895	0.826	0.821	0.824	0.86	0.861
C	2.112	2.164	2.109	1.902	1.516	0.994	0.88	0.868	0.838	0.824	0.809	0.853
Sonic Blank	1.371	1.887	1.481	1.544	1.448	1.451	1.431	1.392	1.38	1.459	1.38	1.478
E	0.86	0.829	0.832	0.827	0.826	0.83	0.825	0.834	0.858	0.848	0.852	0.827
Milli Q Blank	0.86	0.829	0.832	0.827	0.826	0.83	0.825	0.834	0.858	0.848	0.852	0.827
G	0.038	0.038	0.04	0.04	0.04	0.038	0.04	0.042	0.041	0.039	0.039	0.04
Solvent Blank	0.974	0.984	0.932	0.902	0.908	0.971	0.869	0.895	0.916	0.901	0.897	0.838

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.565	0.566	0.563	0.576	0.593	0.579	0.579	0.591	0.594	0.579	0.599	0.589
B	0.581	0.514	0.517	0.534	0.53	0.541	0.54	0.536	0.534	0.542	0.571	0.589
C	0.548	0.542	0.497	0.531	0.539	0.543	0.54	0.54	0.532	0.541	0.538	0.58
Sonic Blank	0.767	0.794	0.811	0.766	0.785	0.728	0.758	0.738	0.705	0.759	0.714	0.693
E	0.593	0.551	0.545	0.548	0.545	0.554	0.553	0.546	0.564	0.561	0.568	0.582
Milli Q Blank	0.593	0.551	0.545	0.548	0.545	0.554	0.553	0.546	0.564	0.561	0.568	0.582
G	0.039	0.039	0.04	0.04	0.041	0.039	0.04	0.042	0.041	0.04	0.04	0.04
Solvent Blank	0.661	0.642	0.603	0.594	0.572	0.616	0.563	0.544	0.575	0.571	0.6	0.552

Calculations with Standard Curve Plate

Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
Estradiol Concentration (M)	2.50E-08	1.25E-08	6.25E-09	3.12408E-09	1.56388E-09	7.81938E-10	3.89134E-10	1.94567E-10	9.91189E-11	4.77239E-11	2.56975E-11	1.10132E-11
Average Absorbance	2.10E+00	2.16E+00	2.14E+00	1.91E+00	1.48E+00	1.04E+00	9.09E-01	8.63E-01	8.54E-01	8.38E-01	8.38E-01	8.25E-01
Milli-Q water Blank			0.559166667									
Solvent Blank			0.591083333									

Mass Balance for Facility A and B – March 2001		
Facility A	Average E2 Conc. (M)	Average E2 Conc.(ng/L)
Raw Influent	3.28084E-11	8.923872361
Raw Influent (sonicated)	9.72234E-11	26.44475533
Primary Effluent	3.4011E-11	9.251003956
Primary Effluent (sonicated)	2.62624E-11	7.143377633
Pilot - Permeate	9.11121E-12	2.478249508
PAC effluent (sonicated)	8.15089E-12	2.217041031
GAC effluent (sonicated)	1.06796E-11	2.904840607
MLSS - 6th stage pilot	1.48951E-10	40.51474617
Mass Balance Calculations		
Influent Flow (gpd)	20,162	
Effluent Flow (gpd)	19,842	
WAS (gpd)	320	
Solid-based estrogenic activity (M)	1.40E-10	
Facility B		
Raw Influent	3.28084E-11	8.923872361
Raw Influent (sonicated)	9.72234E-11	26.44475533
Primary Effluent	3.4011E-11	9.251003956
Primary Effluent (sonicated)	2.62624E-11	7.143377633
Secondary Effluent	1.31664E-11	3.581259342
Secondary Effluent (sonicated)	1.24796E-11	3.394451487
MLSS - conventional	2.94852E-10	80.19981804
Digester Feed - Sludge	4.83523E-10	131.5181804
Primary Digester - Sludge	7.78418E-10	211.7295656
Secondary Digester - Sludge	9.57289E-10	260.3826372
Digester Feed centrate - Sludge	9.52289E-10	259.0226372
Primary Digester centrate - sludge	3.31419E-11	9.014598095
Secondary Digester centrate - sludge	2.5273E-11	6.874248714
Mass Balance Calculations		
Influent Flow (gpd)	3,235,428	
Effluent Flow (gpd)	2,855,288	
WAS (gpd)	51,205	
MLSS Concentration (mg/L)	4,273	
Digester Feed Solids Concentration (mg/L)	12,838	
Primary Digester Solids Concentration (mg/L)	16,503	
Secondary Digester Solids Concentration (mg/L)	8,722	
Digester Volume (L) - each	425,480	
Digester Volume (gallons) - each	112,412	
Volume Transferred (gallons/day)	5,621	
Solid-Based E2 Activity (M)		
Digester Feed	4.70356E-10	
Primary Digester Solids	7.45276E-10	
Secondary Digester Solids	9.32016E-10	

Facility A and B – April, 2001
Plate 1 – Absorbance at 540 nm
Plate 2 – Absorbance at 690 nm
Columns 1 and 2 – Whole Sample
Columns 11 and 12 – Sonicated Sample

Raw Influent	1	2	3	4	5	6	7	8	9	10	11	12
A	1.253	1.4	0.041	0.041	0.041	0.041	0.042	0.042	0.041	0.04	1.864	2.017
B	1.229	1.36	0.041	0.041	0.041	0.04	0.041	0.041	0.041	0.04	2.051	2.037
C	1.25	1.521	0.04	0.058	0.039	0.039	0.04	0.04	0.042	0.039	2.052	2.098
D	1.125	1.409	0.041	0.04	0.039	0.039	0.041	0.04	0.041	0.039	2.119	2.094
E	1.291	1.389	0.041	0.041	0.04	0.041	0.041	0.041	0.041	0.04	2.104	2.181
F	1.329	1.404	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.04	2.171	2.199
G	1.345	1.406	0.04	0.04	0.039	0.039	0.041	0.041	0.04	0.043	2.253	2.189
H	1.328	1.425	0.041	0.041	0.039	0.039	0.041	0.041	0.042	0.039	2.147	2.171
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.663	0.605	0.041	0.041	0.04	0.04	0.041	0.041	0.04	0.04	0.673	0.732
B	0.577	0.601	0.04	0.04	0.04	0.039	0.041	0.04	0.041	0.039	0.691	0.691
C	0.548	0.673	0.04	0.057	0.039	0.039	0.039	0.039	0.041	0.039	0.705	0.701
D	0.566	0.566	0.04	0.04	0.039	0.039	0.04	0.039	0.04	0.039	0.727	0.706
E	0.575	0.583	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.039	0.73	0.775
F	0.63	0.582	0.04	0.04	0.04	0.039	0.04	0.04	0.04	0.039	0.756	0.771
G	0.666	0.623	0.039	0.04	0.039	0.039	0.04	0.04	0.04	0.043	0.805	0.795
H	0.596	0.6	0.04	0.04	0.039	0.039	0.04	0.04	0.041	0.038	0.76	0.784

Primary Effluent	1	2	3	4	5	6	7	8	9	10	11	12
A	1.408	1.367	0.04	0.042	0.042	0.04	0.041	0.042	0.041	0.04	1.279	1.234
B	1.381	1.341	0.041	0.04	0.041	0.039	0.04	0.04	0.044	0.039	1.347	1.365
C	1.467	1.368	0.04	0.039	0.038	0.039	0.04	0.039	0.04	0.039	1.512	1.63
D	1.351	1.393	0.039	0.039	0.038	0.038	0.039	0.039	0.04	0.039	1.299	1.605
E	1.364	1.304	0.04	0.04	0.04	0.041	0.041	0.041	0.041	0.039	1.178	1.57
F	1.342	1.34	0.04	0.041	0.039	0.039	0.04	0.04	0.04	0.04	1.373	1.691
G	1.503	1.395	0.039	0.04	0.039	0.038	0.04	0.04	0.04	0.038	1.37	1.52
H	1.497	1.491	0.04	0.039	0.039	0.038	0.04	0.041	0.043	0.039	1.388	1.644
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.731	0.671	0.04	0.042	0.042	0.04	0.04	0.041	0.04	0.039	0.729	0.69
B	0.692	0.599	0.041	0.04	0.04	0.039	0.04	0.04	0.043	0.039	0.713	0.71
C	0.71	0.593	0.039	0.039	0.038	0.039	0.04	0.039	0.039	0.039	0.863	1.021
D	0.606	0.559	0.039	0.039	0.038	0.038	0.039	0.039	0.04	0.039	0.644	0.975
E	0.584	0.529	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.039	0.644	0.917
F	0.562	0.557	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.762	1.053
G	0.739	0.618	0.039	0.039	0.039	0.038	0.039	0.04	0.04	0.038	0.737	0.891
H	0.694	0.757	0.039	0.039	0.039	0.038	0.04	0.04	0.042	0.039	0.651	0.87

Secondary Effluent Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.255	1.278	0.04	0.04	0.04	0.04	0.041	0.041	0.041	0.039	1.256	1.382
B	1.254	1.354	0.045	0.04	0.04	0.04	0.04	0.04	0.044	0.04	1.491	1.411
C	1.367	1.283	0.039	0.04	0.039	0.038	0.039	0.04	0.04	0.038	1.329	1.358
D	1.299	1.424	0.039	0.04	0.038	0.039	0.04	0.039	0.04	0.038	1.63	1.644
E	1.31	1.371	0.04	0.041	0.04	0.042	0.041	0.041	0.04	0.039	1.562	1.557
F	1.217	1.515	0.04	0.04	0.04	0.039	0.04	0.04	0.042	0.039	1.523	1.391
G	1.371	1.538	0.04	0.039	0.04	0.038	0.045	0.041	0.04	0.039	1.463	1.625
H	1.331	1.387	0.04	0.039	0.038	0.039	0.043	0.041	0.044	0.038	1.615	1.525
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.717	0.759	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.04	0.686	0.781
B	0.707	0.75	0.045	0.041	0.04	0.04	0.041	0.041	0.043	0.04	0.857	0.787
C	0.779	0.583	0.04	0.04	0.04	0.039	0.04	0.04	0.04	0.039	0.722	0.736
D	0.691	0.702	0.04	0.041	0.039	0.039	0.04	0.04	0.04	0.038	1.038	1.044
E	0.692	0.634	0.041	0.041	0.04	0.042	0.041	0.041	0.04	0.04	0.987	0.905
F	0.605	0.742	0.04	0.04	0.041	0.039	0.04	0.04	0.041	0.039	0.912	0.776
G	0.746	0.83	0.04	0.04	0.042	0.039	0.044	0.041	0.04	0.04	0.913	1.035
H	0.708	0.742	0.04	0.04	0.039	0.04	0.043	0.041	0.043	0.039	0.963	0.824

Permeate – Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.373	1.59	0.04	0.043	0.039	0.04	0.043	0.042	0.04	0.04	1.484	1.606
B	1.634	1.687	0.042	0.041	0.041	0.04	0.04	0.042	0.042	0.04	1.821	1.477
C	1.636	1.606	0.04	0.039	0.038	0.038	0.039	0.039	0.039	0.039	1.535	1.357
D	1.578	1.675	0.039	0.039	0.048	0.049	0.041	0.045	0.042	0.038	1.503	1.12
E	1.713	1.615	0.04	0.04	0.039	0.042	0.044	0.043	0.04	0.039	1.382	1.295
F	1.722	1.623	0.04	0.042	0.039	0.039	0.042	0.041	0.04	0.04	1.589	1.103
G	1.552	1.479	0.041	0.039	0.038	0.039	0.04	0.04	0.04	0.038	1.494	1.657
H	1.723	1.711	0.04	0.04	0.038	0.039	0.04	0.041	0.042	0.038	1.81	1.785
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.889	1.059	0.041	0.043	0.04	0.04	0.042	0.042	0.041	0.041	0.838	0.909
B	1.081	1.071	0.042	0.041	0.041	0.041	0.041	0.042	0.042	0.041	1.1	0.799
C	1.042	1.007	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.878	0.681
D	0.984	1.053	0.04	0.04	0.046	0.049	0.041	0.045	0.042	0.039	0.82	0.529
E	1.069	0.996	0.04	0.041	0.04	0.042	0.043	0.043	0.041	0.04	0.786	0.658
F	1.067	0.984	0.04	0.042	0.04	0.04	0.042	0.041	0.041	0.04	1.002	0.454
G	0.92	0.879	0.041	0.04	0.039	0.04	0.04	0.04	0.04	0.039	0.844	0.908
H	1.019	1.038	0.04	0.041	0.039	0.04	0.04	0.041	0.042	0.039	1.053	0.993

PAC effluent Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.509	1.605	0.041	0.041	0.043	0.04	0.042	0.042	0.041	0.04	0.968	1.45
B	1.591	1.648	0.044	0.041	0.041	0.041	0.042	0.041	0.042	0.04	1.297	1.142
C	1.663	1.775	0.04	0.041	0.039	0.039	0.042	0.043	0.043	0.039	1.504	1.169
D	1.623	1.716	0.04	0.042	0.04	0.039	0.041	0.04	0.041	0.039	1.325	1.244
E	1.7	1.602	0.043	0.041	0.04	0.042	0.041	0.041	0.041	0.04	0.986	1.037
F	1.385	1.378	0.041	0.041	0.041	0.041	0.041	0.041	0.042	0.04	0.988	1.16
G	1.53	1.663	0.04	0.04	0.039	0.039	0.042	0.041	0.041	0.039	1.137	1.101
H	1.41	1.686	0.04	0.04	0.039	0.04	0.044	0.041	0.041	0.039	1.148	1.449
	1	2	3	4	5	6	7	8	9	10	11	12
A	1.01	1.049	0.041	0.041	0.043	0.04	0.041	0.041	0.041	0.04	0.428	0.827
B	1.051	1.065	0.043	0.041	0.04	0.04	0.041	0.041	0.041	0.04	0.688	0.581
C	1.071	1.119	0.04	0.04	0.039	0.039	0.042	0.042	0.043	0.039	0.957	0.664
D	1.047	1.091	0.04	0.041	0.04	0.039	0.04	0.04	0.04	0.039	0.796	0.742
E	1.073	1.024	0.042	0.04	0.04	0.041	0.041	0.04	0.04	0.04	0.493	0.519
F	0.825	0.804	0.04	0.041	0.041	0.04	0.041	0.04	0.042	0.04	0.475	0.614
G	0.952	1.075	0.04	0.04	0.039	0.039	0.041	0.04	0.041	0.039	0.502	0.536
H	0.77	1.024	0.04	0.04	0.039	0.04	0.043	0.04	0.041	0.039	0.554	0.851

GAC effluent Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.801	1.637	0.04	0.041	0.04	0.04	0.041	0.041	0.04	0.046	1.661	1.78
B	1.729	1.707	0.04	0.041	0.04	0.039	0.04	0.041	0.041	0.039	1.65	1.562
C	1.806	1.749	0.039	0.039	0.04	0.038	0.039	0.039	0.039	0.04	1.688	1.512
D	1.764	1.764	0.04	0.039	0.043	0.04	0.044	0.039	0.041	0.038	1.802	1.473
E	1.682	1.657	0.04	0.04	0.039	0.04	0.041	0.041	0.041	0.039	1.539	1.645
F	1.723	1.448	0.04	0.042	0.039	0.041	0.041	0.04	0.041	0.04	1.148	1.586
G	1.685	1.698	0.039	0.04	0.038	0.038	0.041	0.041	0.039	0.039	1.354	1.373
H	1.778	1.73	0.041	0.04	0.047	0.042	0.04	0.045	0.041	0.039	1.763	1.672
	1	2	3	4	5	6	7	8	9	10	11	12
A	1.126	1.065	0.041	0.041	0.04	0.04	0.042	0.041	0.041	0.046	0.977	1.077
B	1.118	1.087	0.041	0.041	0.041	0.04	0.041	0.041	0.041	0.04	0.97	0.9
C	1.134	1.122	0.04	0.04	0.04	0.039	0.04	0.04	0.04	0.04	1.039	0.842
D	1.125	1.14	0.04	0.04	0.042	0.041	0.043	0.04	0.041	0.039	1.15	0.847
E	1.016	1.062	0.04	0.04	0.04	0.04	0.041	0.041	0.041	0.04	0.969	0.964
F	1.071	0.864	0.04	0.042	0.04	0.042	0.041	0.04	0.041	0.04	0.608	0.93
G	1.065	1.07	0.04	0.04	0.039	0.039	0.041	0.041	0.04	0.039	0.811	0.73
H	1.084	1.076	0.041	0.04	0.047	0.042	0.04	0.045	0.041	0.039	1.04	0.955

MLSS Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.724	1.817	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.039	1.764	1.862
B	1.808	1.888	0.039	0.043	0.041	0.04	0.041	0.04	0.04	0.038	2.02	1.978
C	1.899	1.966	0.038	0.04	0.04	0.037	0.038	0.039	0.039	0.037	1.976	1.953
D	1.927	1.978	0.038	0.038	0.04	0.038	0.039	0.038	0.039	0.037	2.066	2.021
E	1.961	1.981	0.039	0.04	0.039	0.04	0.04	0.04	0.04	0.039	2.018	2.189
F	1.984	2.029	0.039	0.04	0.04	0.038	0.04	0.04	0.04	0.039	2.038	1.969
G	1.886	1.948	0.039	0.039	0.038	0.038	0.041	0.039	0.039	0.038	2.032	2.048
H	1.883	1.845	0.039	0.039	0.038	0.038	0.039	0.04	0.04	0.038	1.936	2.073
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.654	0.676	0.04	0.04	0.041	0.04	0.041	0.041	0.041	0.04	0.57	0.66
B	0.639	0.652	0.04	0.043	0.041	0.04	0.042	0.04	0.04	0.039	0.652	0.666
C	0.665	0.672	0.039	0.041	0.041	0.038	0.039	0.039	0.039	0.038	0.656	0.628
D	0.649	0.647	0.039	0.039	0.04	0.039	0.039	0.039	0.04	0.038	0.685	0.649
E	0.683	0.669	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.665	0.738
F	0.691	0.692	0.04	0.04	0.041	0.039	0.04	0.04	0.041	0.04	0.649	0.579
G	0.676	0.677	0.04	0.039	0.039	0.039	0.041	0.04	0.04	0.039	0.667	0.668
H	0.668	0.654	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.618	0.723

MLSS – Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.244	1.718	0.045	0.042	0.042	0.041	0.042	0.042	0.045	0.041	1.767	1.889
B	1.693	1.773	0.041	0.041	0.04	0.04	0.044	0.041	0.042	0.042	2.011	2.05
C	1.8	1.859	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.047	2.032	2.115
D	1.818	1.925	0.04	0.04	0.039	0.039	0.04	0.04	0.041	0.039	2.056	2.129
E	1.87	1.93	0.043	0.041	0.04	0.041	0.041	0.041	0.041	0.04	2.161	2.159
F	1.879	1.911	0.041	0.041	0.04	0.04	0.041	0.041	0.042	0.04	2.19	2.157
G	1.848	1.924	0.042	0.044	0.039	0.039	0.041	0.041	0.041	0.039	2.244	2.092
H	1.661	1.802	0.042	0.041	0.041	0.04	0.041	0.041	0.041	0.039	2.05	2.012
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.565	0.69	0.044	0.041	0.041	0.04	0.041	0.041	0.045	0.04	0.571	0.681
B	0.64	0.694	0.041	0.04	0.04	0.04	0.044	0.04	0.041	0.042	0.661	0.68
C	0.674	0.687	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.046	0.677	0.692
D	0.65	0.692	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.67	0.714
E	0.709	0.712	0.042	0.041	0.04	0.04	0.04	0.04	0.04	0.04	0.729	0.724
F	0.73	0.675	0.041	0.04	0.04	0.04	0.04	0.04	0.041	0.04	0.718	0.727
G	0.711	0.711	0.041	0.043	0.039	0.039	0.04	0.04	0.04	0.039	0.777	0.724
H	0.534	0.606	0.042	0.041	0.041	0.04	0.041	0.041	0.041	0.039	0.696	0.706

MLSS Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.617	1.627	0.044	0.041	0.04	0.04	0.044	0.043	0.041	0.04	1.778	1.931
B	1.866	1.932	0.04	0.041	0.04	0.041	0.041	0.041	0.041	0.04	1.937	2.007
C	1.868	1.952	0.039	0.039	0.039	0.039	0.039	0.04	0.043	0.039	1.96	2.05
D	1.852	1.941	0.039	0.039	0.038	0.039	0.04	0.039	0.04	0.038	2.059	2.109
E	1.982	1.99	0.04	0.052	0.044	0.044	0.041	0.041	0.04	0.04	2.038	2.153
F	2.057	2.039	0.04	0.04	0.04	0.04	0.041	0.042	0.041	0.04	2.143	2.178
G	1.998	1.854	0.039	0.04	0.039	0.04	0.041	0.043	0.041	0.039	2.091	2.179
H	1.939	1.912	0.04	0.04	0.039	0.039	0.04	0.047	0.046	0.04	2.016	2.172
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.634	0.645	0.044	0.041	0.04	0.04	0.043	0.042	0.042	0.04	0.631	0.648
B	0.63	0.662	0.04	0.041	0.04	0.042	0.041	0.041	0.041	0.04	0.641	0.693
C	0.67	0.684	0.04	0.04	0.039	0.04	0.04	0.04	0.042	0.04	0.672	0.725
D	0.567	0.664	0.04	0.04	0.039	0.04	0.04	0.04	0.041	0.039	0.722	0.736
E	0.672	0.67	0.04	0.049	0.043	0.043	0.041	0.041	0.041	0.04	0.726	0.771
F	0.691	0.736	0.04	0.04	0.04	0.04	0.041	0.042	0.041	0.04	0.776	0.79
G	0.674	0.616	0.04	0.04	0.04	0.04	0.041	0.043	0.041	0.04	0.754	0.819
H	0.691	0.675	0.04	0.041	0.039	0.039	0.04	0.048	0.045	0.04	0.721	0.76

Primary Digester – Supernatant Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.708	1.847	0.044	0.042	0.04	0.04	0.042	0.044	0.043	0.042	1.522	1.88
B	1.834	1.877	0.042	0.042	0.04	0.04	0.042	0.042	0.043	0.04	1.923	1.712
C	1.936	1.946	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	1.934	1.819
D	1.793	1.801	0.042	0.043	0.039	0.039	0.04	0.04	0.041	0.039	1.98	1.627
E	2.048	1.999	0.041	0.041	0.04	0.041	0.041	0.045	0.041	0.04	1.973	1.743
F	2.01	1.982	0.04	0.041	0.041	0.04	0.041	0.041	0.041	0.04	1.959	1.442
G	1.967	1.949	0.039	0.042	0.039	0.039	0.04	0.043	0.044	0.043	1.726	1.444
H	1.929	1.938	0.044	0.041	0.042	0.039	0.041	0.044	0.047	0.043	1.977	1.595
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.681	0.727	0.043	0.042	0.04	0.04	0.041	0.044	0.043	0.042	0.609	0.723
B	0.645	0.687	0.041	0.041	0.04	0.04	0.041	0.042	0.043	0.04	0.746	0.608
C	0.713	0.723	0.041	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.746	0.717
D	0.602	0.597	0.042	0.043	0.039	0.039	0.04	0.039	0.04	0.039	0.773	0.541
E	0.744	0.74	0.041	0.04	0.04	0.041	0.041	0.044	0.04	0.04	0.749	0.74
F	0.764	0.771	0.04	0.04	0.041	0.04	0.041	0.041	0.041	0.04	0.758	0.56
G	0.779	0.794	0.039	0.042	0.039	0.039	0.04	0.043	0.044	0.042	0.798	0.73
H	0.743	0.79	0.045	0.041	0.041	0.04	0.041	0.044	0.045	0.044	0.789	0.728

Primary Digester – Sludge Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.789	1.869	0.042	0.041	0.04	0.04	0.042	0.042	0.041	0.04	1.737	1.877
B	1.876	2.207	0.041	0.041	0.041	0.041	0.041	0.041	0.041	0.04	1.983	1.613
C	1.956	1.717	0.045	0.04	0.046	0.039	0.04	0.04	0.04	0.039	1.894	1.536
D	1.943	1.93	0.04	0.039	0.04	0.039	0.04	0.041	0.042	0.04	1.831	1.75
E	1.974	1.89	0.042	0.041	0.04	0.041	0.041	0.042	0.042	0.04	1.822	1.376
F	1.972	1.722	0.041	0.041	0.044	0.04	0.041	0.044	0.046	0.041	1.756	1.512
G	1.895	2.06	0.042	0.041	0.041	0.039	0.043	0.041	0.04	0.039	1.51	1.368
H	1.881	1.681	0.042	0.04	0.039	0.039	0.042	0.043	0.042	0.039	1.71	1.35
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.677	0.686	0.042	0.041	0.04	0.04	0.041	0.041	0.041	0.04	0.63	0.692
B	0.673	0.68	0.041	0.041	0.04	0.044	0.041	0.041	0.041	0.04	0.701	0.514
C	0.67	0.522	0.045	0.04	0.046	0.039	0.04	0.04	0.04	0.039	0.689	0.502
D	0.666	0.661	0.04	0.039	0.04	0.039	0.04	0.041	0.041	0.04	0.72	0.657
E	0.702	0.609	0.041	0.041	0.04	0.041	0.041	0.041	0.042	0.039	0.731	0.598
F	0.732	0.459	0.04	0.041	0.044	0.04	0.041	0.043	0.046	0.04	0.742	0.746
G	0.7	0.772	0.042	0.041	0.041	0.039	0.043	0.04	0.04	0.039	0.757	0.679
H	0.721	0.57	0.041	0.04	0.039	0.039	0.041	0.043	0.041	0.039	0.687	0.635

Secondary Digester – Supernatant Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.744	1.807	0.041	0.041	0.041	0.041	0.043	0.042	0.041	0.041	1.748	1.932
B	1.847	1.907	0.055	0.041	0.04	0.04	0.043	0.042	0.042	0.042	2.045	1.832
C	1.94	1.999	0.04	0.042	0.039	0.04	0.04	0.04	0.04	0.039	2.072	2.028
D	2.035	1.911	0.041	0.041	0.039	0.039	0.04	0.04	0.041	0.039	2.07	1.84
E	2.021	2.062	0.041	0.041	0.04	0.041	0.041	0.041	0.041	0.04	2.094	2.028
F	2.085	2.173	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.04	2.126	1.996
G	2.121	2.022	0.04	0.04	0.039	0.039	0.04	0.042	0.041	0.042	2.073	1.939
H	2.066	1.926	0.041	0.047	0.039	0.039	0.04	0.042	0.043	0.039	2.038	2.016
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.601	0.594	0.041	0.041	0.041	0.041	0.043	0.041	0.041	0.041	0.502	0.6
B	0.593	0.585	0.054	0.041	0.04	0.04	0.042	0.041	0.041	0.042	0.672	0.514
C	0.618	0.629	0.04	0.041	0.039	0.039	0.04	0.04	0.04	0.039	0.696	0.692
D	0.648	0.558	0.041	0.041	0.039	0.039	0.04	0.04	0.041	0.039	0.725	0.547
E	0.613	0.653	0.041	0.041	0.04	0.041	0.041	0.041	0.041	0.04	0.713	0.711
F	0.663	0.714	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.04	0.745	0.752
G	0.687	0.628	0.04	0.04	0.039	0.039	0.04	0.042	0.041	0.041	0.748	0.748
H	0.657	0.569	0.041	0.045	0.04	0.039	0.04	0.041	0.042	0.04	0.682	0.762

Secondary Digester – Sludge Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.818	1.864	0.044	0.041	0.04	0.041	0.042	0.043	0.041	0.041	1.953	2.013
B	2.011	1.987	0.046	0.041	0.041	0.04	0.041	0.043	0.043	0.043	2.01	2.079
C	2.078	2.086	0.05	0.04	0.039	0.038	0.04	0.04	0.041	0.052	2.013	2.099
D	1.972	2.057	0.04	0.039	0.039	0.039	0.04	0.04	0.041	0.039	2.221	2.08
E	2.182	2.044	0.041	0.041	0.04	0.041	0.041	0.044	0.043	0.04	2.163	2.03
F	2.125	2.096	0.04	0.041	0.04	0.04	0.041	0.041	0.043	0.04	2.183	2.145
G	2.139	2.062	0.04	0.04	0.041	0.043	0.042	0.046	0.04	0.039	2.15	2.112
H	2.126	2.075	0.042	0.042	0.04	0.039	0.041	0.042	0.044	0.04	2.072	2.151
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.625	0.643	0.043	0.041	0.04	0.041	0.042	0.043	0.041	0.041	0.65	0.693
B	0.663	0.622	0.045	0.041	0.04	0.04	0.041	0.043	0.042	0.043	0.635	0.763
C	0.693	0.703	0.049	0.04	0.039	0.039	0.04	0.04	0.041	0.049	0.653	0.759
D	0.635	0.682	0.04	0.039	0.039	0.039	0.04	0.04	0.041	0.039	0.81	0.737
E	0.734	0.667	0.041	0.04	0.04	0.041	0.041	0.044	0.042	0.04	0.769	0.7
F	0.693	0.687	0.04	0.04	0.04	0.039	0.04	0.041	0.042	0.04	0.783	0.774
G	0.725	0.695	0.04	0.04	0.041	0.043	0.042	0.045	0.04	0.039	0.757	0.788
H	0.727	0.71	0.042	0.042	0.04	0.039	0.041	0.042	0.043	0.04	0.7	0.77

MLSS centrate – Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.165	1.341	0.041	0.041	0.04	0.04	0.042	0.042	0.041	0.041	1.409	1.549
B	1.303	1.5	0.041	0.041	0.04	0.041	0.042	0.041	0.042	0.041	1.459	1.246
C	1.635	1.553	0.04	0.041	0.041	0.038	0.041	0.041	0.041	0.039	1.343	1.341
D	1.515	1.252	0.042	0.044	0.039	0.039	0.039	0.046	0.043	0.04	1.47	1.392
E	1.379	1.399	0.04	0.041	0.04	0.041	0.041	0.044	0.043	0.04	1.457	1.31
F	1.249	1.643	0.041	0.041	0.042	0.04	0.041	0.041	0.041	0.04	1.114	1.444
G	1.488	1.697	0.04	0.04	0.04	0.04	0.041	0.041	0.04	0.04	1.036	1.05
H	1.306	1.437	0.044	0.04	0.039	0.038	0.04	0.044	0.043	0.039	0.985	1.046
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.656	0.737	0.04	0.04	0.04	0.04	0.041	0.041	0.04	0.04	0.838	0.951
B	0.73	0.827	0.04	0.04	0.04	0.041	0.041	0.04	0.041	0.04	0.898	0.617
C	0.834	0.857	0.04	0.04	0.041	0.038	0.04	0.04	0.04	0.038	0.74	0.742
D	0.942	0.579	0.041	0.043	0.039	0.039	0.039	0.044	0.042	0.039	0.911	0.771
E	0.706	0.739	0.04	0.04	0.04	0.04	0.04	0.043	0.041	0.039	0.811	0.673
F	0.638	0.794	0.041	0.04	0.041	0.039	0.04	0.04	0.04	0.039	0.79	1.056
G	0.791	0.797	0.04	0.04	0.039	0.039	0.04	0.04	0.039	0.04	0.664	0.671
H	0.727	0.686	0.043	0.04	0.039	0.038	0.04	0.044	0.041	0.039	0.624	0.649

MLSS centrate- 2nd stage of Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	0.04	0.04	0.041	0.041	0.039	0.748	0.745	0.042	0.041	0.788	0.792	0.041
B	0.039	0.04	0.041	0.042	0.04	0.767	0.789	0.042	0.045	0.781	0.771	0.042
C	0.039	0.039	0.041	0.04	0.04	0.815	0.797	0.042	0.042	0.772	0.781	0.04
D	0.039	0.039	0.04	0.04	0.039	0.759	0.796	0.045	0.042	0.773	0.782	0.044
E	0.04	0.04	0.041	0.042	0.042	0.764	0.854	0.045	0.043	0.784	0.78	0.044
F	0.042	0.041	0.041	0.041	0.041	0.795	0.862	0.044	0.042	0.794	0.769	0.045
G	0.044	0.043	0.042	0.042	0.04	0.78	0.818	0.042	0.043	0.775	0.762	0.043
H	0.04	0.04	0.041	0.041	0.039	0.741	0.773	0.042	0.042	0.775	0.772	0.04
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.041	0.04	0.041	0.041	0.04	0.479	0.47	0.043	0.042	0.537	0.542	0.041
B	0.04	0.041	0.041	0.042	0.041	0.486	0.495	0.042	0.045	0.537	0.526	0.041
C	0.039	0.039	0.041	0.04	0.04	0.526	0.507	0.042	0.041	0.532	0.536	0.041
D	0.04	0.04	0.04	0.041	0.04	0.49	0.498	0.044	0.042	0.533	0.536	0.044
E	0.041	0.041	0.042	0.041	0.042	0.488	0.507	0.044	0.043	0.539	0.533	0.044
F	0.043	0.041	0.041	0.041	0.041	0.5	0.53	0.044	0.042	0.545	0.525	0.044
G	0.044	0.043	0.042	0.042	0.041	0.494	0.504	0.042	0.043	0.533	0.523	0.044
H	0.041	0.04	0.041	0.041	0.039	0.475	0.486	0.043	0.042	0.535	0.525	0.041

MLSS centrate - 6th stage of Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.451	1.486	0.041	0.041	0.043	0.041	0.042	0.042	0.042	0.045	2.097	2.141
B	1.496	1.592	0.041	0.041	0.041	0.039	0.043	0.041	0.041	0.041	2.155	2.08
C	1.473	1.754	0.041	0.04	0.039	0.038	0.04	0.04	0.04	0.039	2.204	2.156
D	1.618	1.578	0.04	0.04	0.039	0.039	0.04	0.04	0.041	0.039	1.774	1.092
E	1.641	1.554	0.041	0.041	0.04	0.041	0.043	0.044	0.05	0.04	2.162	1.945
F	1.21	1.371	0.04	0.041	0.041	0.044	0.041	0.041	0.043	0.041	1.014	1.003
G	1.53	1.553	0.04	0.042	0.042	0.039	0.04	0.041	0.043	0.039	0.982	1.055
H	1.77	1.862	0.041	0.045	0.039	0.039	0.041	0.041	0.052	0.089	0.959	1.042
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.966	1.009	0.041	0.041	0.043	0.041	0.042	0.042	0.042	0.045	0.794	0.926
B	0.968	1.029	0.04	0.04	0.041	0.04	0.042	0.041	0.041	0.041	0.751	0.721
C	1.003	1.084	0.041	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.807	0.786
D	1.005	1.005	0.04	0.04	0.039	0.039	0.04	0.04	0.041	0.039	0.644	0.791
E	1.023	0.911	0.04	0.041	0.04	0.041	0.042	0.042	0.047	0.04	0.882	0.697
F	0.571	0.729	0.04	0.041	0.04	0.044	0.041	0.041	0.043	0.041	0.631	0.638
G	0.878	0.936	0.04	0.041	0.042	0.039	0.04	0.041	0.043	0.039	0.604	0.683
H	1.103	1.14	0.041	0.045	0.039	0.04	0.041	0.041	0.05	0.086	0.609	0.677

Primary Digester centrate – supernatant Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.286	1.508	0.04	0.041	0.04	0.04	0.041	0.041	0.041	0.046	1.26	0.99
B	1.272	1.439	0.041	0.04	0.04	0.04	0.04	0.04	0.041	0.042	1.265	1.044
C	1.518	1.504	0.039	0.039	0.038	0.038	0.039	0.039	0.039	0.038	1.413	1.025
D	1.304	1.483	0.04	0.039	0.038	0.038	0.039	0.039	0.04	0.039	1.146	0.984
E	1.829	1.554	0.047	0.041	0.04	0.064	0.042	0.04	0.041	0.04	1.327	0.96
F	1.413	1.263	0.04	0.04	0.039	0.039	0.04	0.04	0.042	0.04	1.113	1.025
G	0.755	0.847	0.04	0.044	0.041	0.043	0.04	0.043	0.042	0.039	1.371	0.707
H	0.831	0.831	0.041	0.042	0.041	0.038	0.044	0.043	0.041	0.039	1.347	0.831
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.543	0.669	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.045	0.661	0.634
B	0.532	0.568	0.041	0.04	0.04	0.04	0.04	0.041	0.041	0.042	0.597	0.674
C	0.611	0.615	0.039	0.039	0.038	0.039	0.039	0.039	0.039	0.039	0.969	0.653
D	0.631	0.684	0.041	0.039	0.039	0.039	0.039	0.039	0.04	0.039	0.785	0.619
E	0.855	0.886	0.046	0.04	0.04	0.059	0.041	0.04	0.04	0.04	0.922	0.604
F	0.706	0.532	0.04	0.04	0.039	0.039	0.04	0.04	0.042	0.04	0.764	0.648
G	0.397	0.473	0.04	0.043	0.041	0.044	0.04	0.042	0.041	0.039	0.694	0.384
H	0.475	0.466	0.041	0.041	0.042	0.039	0.044	0.042	0.041	0.039	0.899	0.479

Primary Digester centrate – sludge Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.57	1.499	0.041	0.041	0.04	0.04	0.041	0.042	0.041	0.04	0.103	1.289
B	1.372	1.549	0.042	0.041	0.04	0.04	0.041	0.042	0.041	0.041	0.04	1.027
C	1.589	1.402	0.04	0.04	0.039	0.04	0.04	0.04	0.04	0.045	0.04	0.758
D	1.594	1.363	0.041	0.039	0.043	0.039	0.04	0.04	0.041	0.039	0.049	1.142
E	1.508	1.492	0.041	0.041	0.04	0.041	0.043	0.041	0.041	0.04	0.04	1.334
F	1.522	1.366	0.041	0.048	0.045	0.04	0.041	0.044	0.042	0.043	0.04	1.361
G	1.459	1.453	0.04	0.042	0.041	0.039	0.04	0.04	0.04	0.044	0.041	1.223
H	1.762	1.541	0.042	0.043	0.04	0.04	0.04	0.041	0.043	0.039	0.041	1.612
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.825	0.879	0.041	0.041	0.04	0.04	0.041	0.042	0.041	0.04	0.098	0.688
B	0.808	0.814	0.042	0.041	0.04	0.04	0.041	0.042	0.041	0.041	0.04	0.555
C	0.801	0.6	0.04	0.04	0.039	0.04	0.04	0.04	0.04	0.046	0.04	0.471
D	0.844	0.59	0.041	0.04	0.044	0.039	0.04	0.04	0.041	0.039	0.05	0.636
E	0.729	0.747	0.041	0.041	0.04	0.041	0.043	0.041	0.041	0.04	0.04	0.754
F	0.83	0.666	0.041	0.047	0.044	0.04	0.041	0.044	0.042	0.042	0.041	0.78
G	0.72	0.712	0.04	0.041	0.042	0.04	0.04	0.041	0.04	0.044	0.041	0.426
H	0.968	0.802	0.042	0.042	0.04	0.04	0.04	0.041	0.042	0.039	0.041	0.305

Secondary Digester centrate – supernatnant Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.29	1.497	0.04	0.041	0.04	0.04	0.042	0.041	0.042	0.039	1.394	1.539
B	1.325	1.298	0.04	0.042	0.04	0.04	0.041	0.04	0.041	0.039	1.549	1.567
C	1.446	1.495	0.041	0.039	0.038	0.038	0.039	0.042	0.039	0.038	1.451	0.935
D	1.477	0.966	0.039	0.039	0.039	0.038	0.039	0.039	0.04	0.038	1.426	1.164
E	1.293	1.643	0.04	0.04	0.041	0.04	0.041	0.041	0.04	0.041	1.199	1.407
F	1.353	1.288	0.041	0.046	0.039	0.039	0.041	0.04	0.04	0.039	0.857	0.896
G	1.175	1.354	0.04	0.04	0.039	0.038	0.039	0.04	0.04	0.039	0.947	0.873
H	1.363	1.484	0.04	0.04	0.04	0.042	0.039	0.04	0.041	0.039	0.819	0.948
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.624	0.751	0.04	0.041	0.04	0.04	0.042	0.041	0.042	0.04	0.697	0.945
B	0.842	0.643	0.04	0.042	0.04	0.041	0.041	0.04	0.041	0.039	0.84	0.954
C	0.686	0.759	0.04	0.039	0.039	0.039	0.039	0.042	0.04	0.039	0.902	0.622
D	0.715	0.432	0.039	0.039	0.04	0.039	0.04	0.04	0.04	0.038	0.823	0.642
E	0.66	0.902	0.04	0.04	0.041	0.04	0.04	0.041	0.04	0.041	0.703	0.944
F	0.704	0.677	0.041	0.046	0.039	0.039	0.041	0.04	0.041	0.04	0.514	0.56
G	0.546	0.653	0.04	0.04	0.039	0.039	0.04	0.04	0.04	0.039	0.591	0.535
H	0.606	0.729	0.04	0.04	0.04	0.043	0.04	0.041	0.041	0.039	0.488	0.6

Secondary Digester centrate - sludge	1	2	3	4	5	6	7	8	9	10	11	12
A	1.332	1.419	0.041	0.041	0.04	0.04	0.041	0.042	0.041	0.041	1.251	1.154
B	1.217	1.178	0.041	0.041	0.04	0.039	0.041	0.041	0.041	0.04	1.329	1.235
C	1.27	1.237	0.04	0.04	0.039	0.039	0.039	0.04	0.04	0.039	1.273	1.095
D	1.151	1.199	0.039	0.04	0.039	0.039	0.04	0.039	0.04	0.039	1.206	1.087
E	0.821	0.838	0.041	0.041	0.041	0.041	0.041	0.041	0.041	0.04	0.987	0.985
F	0.761	0.807	0.04	0.041	0.04	0.04	0.041	0.043	0.042	0.042	0.982	1.005
G	0.815	0.811	0.04	0.041	0.041	0.046	0.04	0.042	0.042	0.039	0.956	0.919
H	0.723	0.826	0.041	0.04	0.041	0.041	0.042	0.042	0.042	0.039	0.97	0.947
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.618	0.81	0.041	0.041	0.04	0.04	0.041	0.041	0.041	0.04	0.609	0.616
B	0.557	0.573	0.04	0.04	0.04	0.04	0.041	0.04	0.041	0.04	0.68	0.738
C	0.661	0.61	0.04	0.04	0.039	0.039	0.039	0.04	0.04	0.039	0.653	0.607
D	0.592	0.641	0.04	0.039	0.039	0.039	0.04	0.039	0.04	0.039	0.582	0.586
E	0.466	0.48	0.041	0.04	0.041	0.04	0.041	0.041	0.04	0.04	0.621	0.613
F	0.422	0.463	0.04	0.04	0.04	0.04	0.04	0.042	0.042	0.041	0.617	0.634
G	0.478	0.459	0.04	0.04	0.041	0.047	0.04	0.042	0.041	0.039	0.604	0.569
H	0.41	0.478	0.041	0.04	0.041	0.04	0.042	0.041	0.041	0.039	0.619	0.6

Estradiol Control	1	2	3	4	5	6	7	8	9	10	11	12
A	2.228	2.157	2.04	1.981	1.978	1.266	1.096	1.018	0.951	0.984	0.959	1.073
B	2.19	2.209	2.083	2.029	1.958	1.286	1.103	1.012	0.982	0.949	0.972	1.002
C	2.147	2.27	2.169	2.007	1.751	1.405	1.197	1.031	0.993	0.975	0.992	0.99
D	0.04	0.031	0.034	0.033	0.032	0.032	0.033	0.034	0.034	0.032	0.032	0.038
E	1.421	1.205	1.324	1.639	1.641	1.406	1.163	1.473	1.678	1.296	1.334	1.311
Milli Q Blank	1.014	0.991	1.002	0.964	0.97	1.002	1.009	0.993	0.962	0.975	1.008	1.077
G	0.039	0.04	0.041	0.041	0.039	0.041	0.041	0.043	0.041	0.04	0.041	0.041
Solvent Blank	1.033	0.978	1.134	1.065	1.057	0.989	0.96	0.989	0.977	0.958	1.042	1.051

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.682	0.599	0.554	0.558	0.601	0.543	0.576	0.586	0.561	0.599	0.589	0.699
B	0.637	0.598	0.565	0.555	0.589	0.551	0.533	0.54	0.558	0.554	0.588	0.627
C	0.6	0.628	0.561	0.522	0.532	0.553	0.564	0.551	0.55	0.55	0.598	0.623
D	0.039	0.03	0.032	0.032	0.031	0.031	0.032	0.032	0.032	0.03	0.031	0.033
E	0.624	0.66	0.74	0.744	0.809	0.803	0.611	0.91	0.691	0.74	0.749	0.663
Milli Q Blank	0.603	0.55	0.551	0.541	0.538	0.558	0.558	0.554	0.54	0.552	0.6	0.69
G	0.039	0.039	0.041	0.041	0.039	0.041	0.04	0.041	0.04	0.039	0.041	0.04
Solvent Blank	0.52	0.45	0.611	0.578	0.58	0.544	0.531	0.551	0.576	0.543	0.605	0.59

Calculations with Standard Curve Plate

Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
Estradiol Concentration (M)	2.50E-08	1.25E-08	6.25E-09	3.12408E-09	1.56388E-09	7.81938E-10	3.89134E-10	1.94567E-10	9.91189E-11	4.77239E-11	2.56975E-11	1.10132E-11
Average Absorbance	2.12E+00	2.17E+00	2.11E+00	2.03E+00	1.89E+00	1.34E+00	1.14E+00	1.03E+00	9.89E-01	9.71E-01	9.52E-01	9.42E-01

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Milli Q Blank	0.603	0.55	0.551	0.541	0.538	0.558	0.558	0.554	0.54	0.552	0.6	0.69
G	0.039	0.039	0.041	0.041	0.039	0.041	0.04	0.041	0.04	0.039	0.041	0.04
Solvent Blank	0.52	0.45	0.611	0.578	0.58	0.544	0.531	0.551	0.576	0.543	0.605	0.59

Calculations with Standard Curve Plate

Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
Estradiol Concentration (M)	2.50E-08	1.25E-08	6.25E-09	3.12408E-09	1.56388E-09	7.81938E-10	3.89134E-10	1.94567E-10	9.91189E-11	4.77239E-11	2.56975E-11	1.10132E-11
Average Absorbance	2.12E+00	2.17E+00	2.11E+00	2.03E+00	1.89E+00	1.34E+00	1.14E+00	1.03E+00	9.89E-01	9.71E-01	9.52E-01	9.42E-01

Mass Balance for Facility A and B - April, 2001		
Facility A	**Average E2 Conc. (M)**	**Average E2 Conc. (ng/L)**
Raw Influent	1.11548E-10	30.34116603
Raw Influent (sonicated)	3.30559E-10	89.91216814
Primary Effluent	1.15638E-10	31.45341345
Primary Effluent (sonicated)	8.92922E-11	24.28748395
Pilot - Permeate	3.09781E-11	8.426048328
PAC effluent (sonicated)	2.7713E-11	7.537939505
GAC effluent (sonicated)	3.63105E-11	9.876458063
MLSS - 6th stage pilot	5.06434E-10	137.750137
Mass Balance Calculations		
Influent Flow (gpd)	20,162	
Effluent Flow (gpd)	19,842	
WAS (gpd)	320	
MLSS Concentration (mg/L)	10,752	
Solid-based estrogenic activity (M)	4.75E-10	
Facility B		
Raw Influent	1.11548E-10	30.34116603
Raw Influent (sonicated)	3.30559E-10	89.91216814
Primary Effluent	1.15638E-10	31.45341345
Primary Effluent (sonicated)	8.92922E-11	24.28748395
Secondary Effluent	4.47657E-11	12.17628176
Secondary Effluent (sonicated)	4.24306E-11	11.54113506
MLSS - conventional	1.0025E-09	272.6793814
Digester Feed - Sludge	1.64398E-09	447.1618135
Primary Digester - Sludge	2.64662E-09	719.880523
Secondary Digester - Sludge	3.25478E-09	885.3009666
Digester Feed centrate - Sludge	3.23778E-09	880.6769666
Primary Digester centrate - sludge	1.12682E-10	30.64963352
Secondary Digester centrate - sludge	8.59281E-11	23.37244563
Mass Balance Calculations		
Influent Flow (gpd)	3,045,963	
Effluent Flow (gpd)	2,655,449	
WAS (gpd)	51,205	
MLSS Concentration (mg/L)	4,883	
Digester Feed Solids Concentration (mg/L)	14,672	
Primary Digester Solids Concentration (mg/L)	18,861	
Secondary Digester Solids Concentration (mg/L)	9,968	
Digester Volume (L) - each	425,480	
Digester Volume (gallons) - each	112,412	
Volume Transferred (gallons/day)	5,621	
Solid-Based E2 Activity (M)		
Digester Feed	1.59921E-09	
Primary Digester Solids	2.53394E-09	
Secondary Digester Solids	3.16885E-09	

Facility A and B – May, 2001
Plate 1 – Absorbance at 540 nm
Plate 2 – Absorbance at 690 nm
Columns 1 and 2 – Whole Sample
Columns 11 and 12 – Sonicated Sample

Raw Influent	1	2	3	4	5	6	7	8	9	10	11	12
A	1.501	0.04	1.436	0.04	1.39	0.039	1.495	0.041	1.45	0.04	1.74	0.04
B	1.555	0.039	1.491	0.04	0.946	0.039	1.678	0.04	1.647	0.039	1.8	0.04
C	1.452	0.038	1.541	0.039	1.704	0.038	1.683	0.042	1.75	0.04	1.89	0.04
D	1.526	0.041	1.58	0.039	1.706	0.042	1.793	0.046	1.748	0.044	1.08	0.04
E	1.541	0.039	1.612	0.041	1.79	0.063	1.816	0.047	1.756	0.046	1.86	0.04
F	1.564	0.039	1.621	0.042	1.673	0.041	1.822	0.043	1.016	0.046	1.75	0.043
G	0.921	0.038	1.506	0.039	1.723	0.038	1.771	0.041	1.708	0.038	2	0.04
H	1.579	0.04	1.55	0.042	1.618	0.038	1.68	0.04	0.83	0.038	1.78	0.041
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.652	0.04	0.591	0.04	0.567	0.039	0.552	0.041	0.568	0.04	0.65	0.04
B	0.638	0.039	0.586	0.04	0.561	0.039	0.567	0.04	0.549	0.039	0.59	0.04
C	0.604	0.038	0.568	0.039	0.597	0.038	0.585	0.042	0.579	0.04	0.58	0.04
D	0.605	0.041	0.579	0.039	0.59	0.042	0.625	0.045	0.547	0.044	0.67	0.04
E	0.61	0.039	0.592	0.041	0.583	0.057	0.608	0.047	0.56	0.045	0.55	0.04
F	0.637	0.039	0.597	0.041	0.583	0.041	0.594	0.043	0.502	0.046	0.63	0.043
G	0.597	0.038	0.589	0.039	0.588	0.038	0.568	0.041	0.548	0.038	0.6	0.04
H	0.658	0.039	0.609	0.041	0.553	0.038	0.546	0.04	0.625	0.039	0.58	0.04

Primary Effluent	1	2	3	4	5	6	7	8	9	10	11	12
A	1.077	0.039	0.04	0.041	0.04	1.42	0.042	0.042	0.041	0.05	0.04	1.67
B	1.073	0.039	0.04	0.041	0.042	1.639	0.04	0.04	0.041	0.039	0.04	1.819
C	1.05	0.038	0.039	0.041	0.038	1.192	0.039	0.04	0.041	0.038	0.04	1.786
D	0.934	0.039	0.039	0.039	0.038	1.755	0.04	0.041	0.041	0.038	0.04	1.815
E	1.023	0.039	0.041	0.041	0.04	1.339	0.041	0.041	0.041	0.043	0.04	1.858
F	1.313	0.04	0.04	0.041	0.04	1.701	0.041	0.041	0.041	0.04	0.04	1.878
G	1.598	0.039	0.04	0.04	0.039	1.723	0.04	0.041	0.04	0.039	0.04	1.903
H	1.683	0.04	0.04	0.04	0.039	1.694	0.042	0.041	0.042	0.04	0.04	1.754
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.813	0.039	0.04	0.04	0.04	0.616	0.041	0.041	0.04	0.05	0.04	0.727
B	0.61	0.039	0.04	0.04	0.041	0.573	0.04	0.039	0.04	0.039	0.04	0.606
C	0.789	0.038	0.039	0.041	0.038	0.672	0.039	0.039	0.04	0.038	0.04	0.625
D	0.686	0.039	0.039	0.039	0.038	0.61	0.039	0.039	0.04	0.038	0.04	0.592
E	0.672	0.039	0.04	0.04	0.039	0.659	0.04	0.04	0.04	0.042	0.04	0.6
F	0.679	0.04	0.04	0.04	0.04	0.539	0.04	0.04	0.04	0.039	0.04	0.605
G	0.63	0.038	0.039	0.039	0.039	0.584	0.039	0.04	0.039	0.039	0.04	0.621
H	0.739	0.04	0.04	0.04	0.039	0.743	0.041	0.04	0.041	0.04	0.04	0.761

Secondary Effluent – Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.339	0.039	0.04	0.041	0.039	1.34	0.041	0.041	0.04	0.039	0.04	1.554
B	1.214	0.039	0.041	0.045	0.039	1.433	0.04	0.04	0.041	0.039	0.04	1.613
C	1.224	0.038	0.042	0.039	0.041	1.544	0.039	0.039	0.039	0.038	0.04	1.715
D	1.471	0.038	0.043	0.04	0.039	1.348	0.04	0.039	0.04	0.038	0.04	1.717
E	1.566	0.039	0.04	0.041	0.04	1.256	0.041	0.041	0.04	0.039	0.04	1.382
F	1.108	0.047	0.041	0.04	0.04	1.411	0.041	0.04	0.04	0.04	0.04	1.752
G	1.634	0.039	0.041	0.04	0.038	1.573	0.041	0.04	0.039	0.04	0.04	1.92
H	1.601	0.039	0.041	0.043	0.039	1.542	0.04	0.041	0.042	0.04	0.04	1.786
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.81	0.04	0.04	0.041	0.04	0.676	0.041	0.041	0.041	0.04	0.04	0.702
B	0.662	0.04	0.041	0.044	0.04	0.662	0.04	0.04	0.041	0.04	0.04	0.708
C	0.701	0.039	0.043	0.04	0.041	0.736	0.04	0.04	0.04	0.039	0.04	0.649
D	0.596	0.039	0.043	0.041	0.039	0.643	0.04	0.04	0.04	0.039	0.04	0.67
E	0.625	0.04	0.04	0.041	0.04	0.755	0.041	0.041	0.04	0.04	0.04	0.728
F	0.676	0.048	0.041	0.04	0.04	0.606	0.041	0.041	0.041	0.04	0.04	0.62
G	0.647	0.04	0.041	0.04	0.039	0.614	0.041	0.04	0.04	0.04	0.04	0.762
H	0.691	0.04	0.042	0.043	0.04	0.667	0.04	0.041	0.041	0.041	0.04	0.729

Permeate – Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.881	0.04	0.047	0.044	0.04	1.659	0.041	0.042	0.041	0.04	0.04	2.111
B	1.462	0.041	0.04	0.04	0.04	1.747	0.042	0.043	0.042	0.04	0.04	1.701
C	1.64	0.039	0.04	0.04	0.039	1.761	0.041	0.041	0.04	0.039	0.04	1.673
D	1.529	0.038	0.041	0.04	0.039	1.701	0.04	0.04	0.041	0.039	0.04	1.677
E	1.723	0.039	0.04	0.041	0.04	1.811	0.041	0.041	0.041	0.04	0.04	1.69
F	1.659	0.039	0.04	0.041	0.04	1.808	0.042	0.04	0.041	0.042	0.04	1.692
G	1.785	0.039	0.04	0.04	0.039	1.869	0.04	0.04	0.04	0.039	0.04	1.772
H	1.436	0.043	0.04	0.04	0.039	1.804	0.042	0.041	0.041	0.042	0.04	1.957
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.944	0.041	0.047	0.044	0.04	0.733	0.041	0.041	0.041	0.04	0.04	1.033
B	0.713	0.041	0.04	0.04	0.04	0.618	0.041	0.042	0.041	0.04	0.04	0.654
C	0.637	0.039	0.04	0.04	0.039	0.613	0.041	0.041	0.04	0.039	0.04	0.604
D	0.753	0.039	0.041	0.039	0.039	0.598	0.04	0.04	0.04	0.039	0.04	0.609
E	0.628	0.039	0.04	0.04	0.04	0.622	0.04	0.041	0.04	0.04	0.04	0.596
F	0.633	0.039	0.04	0.04	0.04	0.629	0.041	0.04	0.04	0.041	0.04	0.604
G	0.815	0.039	0.039	0.04	0.038	0.599	0.04	0.04	0.04	0.039	0.04	0.64
H	0.776	0.042	0.04	0.04	0.039	0.636	0.041	0.04	0.041	0.042	0.04	0.935

GAC and PAC – Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.402	0.039	0.04	0.042	0.04	1.465	0.04	0.041	0.042	0.039	0.04	1.786
B	1.396	0.043	0.04	0.04	0.039	1.528	0.04	0.04	0.04	0.041	0.04	1.699
C	1.34	0.038	0.04	0.043	0.038	1.619	0.039	0.039	0.041	0.039	0.04	1.785
D	1.428	0.039	0.041	0.039	0.038	1.63	0.039	0.041	0.042	0.038	0.04	1.615
E	1.371	0.04	0.046	0.04	0.039	1.597	0.041	0.041	0.04	0.04	0.04	1.794
F	1.423	0.039	0.04	0.04	0.039	1.569	0.04	0.04	0.04	0.039	0.04	1.778
G	1.39	0.038	0.039	0.04	0.039	1.583	0.04	0.04	0.04	0.038	0.04	1.851
H	1.493	0.039	0.04	0.04	0.04	1.616	0.039	0.04	0.042	0.039	0.04	1.809
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.713	0.04	0.041	0.042	0.041	0.653	0.041	0.042	0.042	0.04	0.04	0.805
B	0.618	0.043	0.041	0.041	0.04	0.526	0.041	0.041	0.041	0.042	0.04	0.543
C	0.568	0.039	0.041	0.043	0.039	0.564	0.04	0.04	0.042	0.04	0.04	0.605
D	0.592	0.04	0.042	0.04	0.039	0.58	0.04	0.041	0.042	0.039	0.04	0.595
E	0.582	0.041	0.045	0.041	0.04	0.603	0.042	0.041	0.041	0.04	0.04	0.55
F	0.615	0.04	0.041	0.041	0.04	0.571	0.041	0.041	0.041	0.04	0.04	0.543
G	0.604	0.039	0.04	0.041	0.04	0.575	0.04	0.041	0.041	0.039	0.04	0.588
H	0.717	0.04	0.04	0.04	0.041	0.622	0.04	0.041	0.042	0.04	0.04	0.72

MLSS – Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	1.284	0.039	0.04	0.04	0.039	1.287	0.041	0.041	0.04	0.039	0.04	1.614
B	1.196	0.039	0.041	0.044	0.039	1.454	0.042	0.04	0.04	0.039	0.04	1.733
C	1.302	0.038	0.039	0.039	0.038	1.466	0.039	0.039	0.039	0.038	0.04	1.721
D	1.265	0.05	0.039	0.039	0.038	1.463	0.039	0.039	0.04	0.038	0.04	1.771
E	1.323	0.042	0.04	0.041	0.039	1.485	0.04	0.041	0.04	0.039	0.04	1.791
F	1.3	0.039	0.041	0.04	0.039	1.381	0.041	0.044	0.04	0.04	0.04	1.764
G	1.249	0.041	0.039	0.04	0.039	1.474	0.039	0.045	0.04	0.039	0.04	1.8
H	1.382	0.039	0.04	0.039	0.039	1.336	0.039	0.04	0.041	0.039	0.04	1.874
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.752	0.039	0.04	0.041	0.04	0.725	0.041	0.041	0.041	0.04	0.04	0.866
B	0.605	0.039	0.041	0.044	0.04	0.623	0.043	0.041	0.041	0.04	0.04	0.577
C	0.603	0.039	0.039	0.04	0.039	0.607	0.039	0.04	0.04	0.039	0.04	0.557
D	0.597	0.051	0.039	0.039	0.039	0.564	0.04	0.04	0.04	0.039	0.04	0.551
E	0.602	0.042	0.04	0.041	0.04	0.628	0.041	0.041	0.041	0.04	0.04	0.543
F	0.577	0.04	0.041	0.041	0.04	0.575	0.041	0.044	0.041	0.041	0.04	0.55
G	0.59	0.042	0.04	0.041	0.039	0.594	0.04	0.044	0.04	0.039	0.04	0.6
H	0.733	0.04	0.04	0.04	0.039	0.604	0.04	0.041	0.041	0.04	0.04	0.829

MLSS – Facility A	1	2	3	4	5	6	7	8	9	10	11	12
A	1.915	0.04	0.041	0.041	0.04	1.172	0.041	0.042	0.04	0.039	0.04	1.761
B	1.259	0.039	0.04	0.04	0.039	1.197	0.04	0.04	0.042	0.043	0.04	1.341
C	1.19	0.038	0.041	0.039	0.038	1.22	0.039	0.039	0.04	0.04	0.04	1.139
D	1.232	0.038	0.041	0.039	0.038	1.247	0.04	0.039	0.04	0.038	0.04	1.183
E	1.246	0.039	0.04	0.041	0.04	1.1	0.041	0.041	0.04	0.039	0.04	1.727
F	1.318	0.039	0.04	0.041	0.039	1.182	0.042	0.04	0.041	0.039	0.04	1.787
G	1.351	0.038	0.039	0.039	0.039	1.236	0.04	0.04	0.039	0.038	0.04	1.829
H	1.295	0.039	0.04	0.039	0.038	1.279	0.039	0.04	0.041	0.038	0.04	1.881
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.603	0.039	0.041	0.04	0.04	0.611	0.04	0.041	0.04	0.039	0.04	0.668
B	0.561	0.039	0.04	0.04	0.039	0.641	0.04	0.04	0.041	0.042	0.04	0.83
C	0.544	0.038	0.04	0.039	0.038	0.592	0.039	0.039	0.039	0.04	0.04	0.754
D	0.566	0.038	0.04	0.039	0.038	0.605	0.04	0.039	0.04	0.038	0.04	0.724
E	0.569	0.039	0.039	0.04	0.039	0.7	0.04	0.04	0.04	0.039	0.04	0.533
F	0.587	0.039	0.04	0.04	0.039	0.603	0.04	0.04	0.04	0.039	0.04	0.533
G	0.607	0.038	0.039	0.039	0.039	0.62	0.039	0.039	0.039	0.038	0.04	0.591
H	0.648	0.038	0.039	0.039	0.038	0.683	0.039	0.04	0.04	0.038	0.04	0.763

Digester B – Supernatant **Facility B**	1	2	3	4	5	6	7	8	9	10	11	12
A	1.335	0.038	0.04	0.041	0.041	1.258	0.041	0.041	0.04	0.039	0.04	1.746
B	1.369	0.038	0.039	0.041	0.039	1.315	0.04	0.043	0.04	0.039	0.04	1.683
C	1.351	0.037	0.039	0.039	0.04	1.283	0.039	0.039	0.039	0.038	0.04	1.744
D	1.311	0.037	0.038	0.038	0.038	1.263	0.04	0.039	0.04	0.038	0.04	1.769
E	1.349	0.038	0.039	0.04	0.039	1.237	0.04	0.041	0.041	0.041	0.04	1.835
F	1.437	0.039	0.039	0.039	0.039	1.206	0.046	0.04	0.04	0.039	0.04	1.837
G	1.424	0.037	0.04	0.039	0.038	1.229	0.04	0.041	0.043	0.038	0.04	1.771
H	1.454	0.038	0.045	0.039	0.038	1.202	0.04	0.04	0.042	0.042	0.04	1.786
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.749	0.039	0.04	0.041	0.041	0.659	0.04	0.041	0.04	0.039	0.04	0.714
B	0.701	0.039	0.039	0.041	0.039	0.714	0.04	0.042	0.04	0.039	0.04	0.594
C	0.645	0.038	0.039	0.039	0.04	0.714	0.039	0.039	0.039	0.038	0.04	0.605
D	0.71	0.038	0.039	0.039	0.038	0.641	0.039	0.039	0.04	0.038	0.04	0.633
E	0.639	0.039	0.039	0.04	0.039	0.616	0.04	0.04	0.04	0.04	0.04	0.658
F	0.681	0.039	0.039	0.039	0.039	0.587	0.045	0.04	0.04	0.039	0.04	0.632
G	0.673	0.038	0.04	0.039	0.038	0.62	0.04	0.041	0.041	0.038	0.04	0.63
H	0.701	0.038	0.046	0.039	0.038	0.604	0.04	0.041	0.041	0.041	0.04	0.666

Digester B – Sludge **Facility B**	1	2	3	4	5	6	7	8	9	10	11	12
A	1.3	0.039	0.042	0.041	0.04	1.401	0.045	0.042	0.041	0.04	0.04	1.589
B	1.387	0.04	0.042	0.04	0.04	1.668	0.041	0.041	0.041	0.04	0.04	1.671
C	1.404	0.038	0.04	0.04	0.039	1.616	0.039	0.04	0.04	0.039	0.04	1.625
D	1.477	0.04	0.043	0.041	0.039	1.588	0.041	0.04	0.041	0.039	0.04	1.725
E	1.449	0.039	0.04	0.041	0.045	1.632	0.041	0.041	0.041	0.04	0.04	1.688
F	1.475	0.041	0.041	0.04	0.04	1.602	0.041	0.04	0.041	0.04	0.04	1.702
G	1.517	0.039	0.04	0.04	0.039	1.559	0.04	0.04	0.043	0.039	0.04	1.712
H	1.477	0.04	0.04	0.04	0.042	1.586	0.041	0.041	0.042	0.039	0.04	1.837
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.587	0.04	0.042	0.041	0.041	0.567	0.044	0.042	0.041	0.04	0.04	0.591
B	0.574	0.04	0.042	0.041	0.04	0.63	0.041	0.041	0.041	0.04	0.04	0.597
C	0.559	0.039	0.04	0.04	0.039	0.556	0.04	0.04	0.04	0.039	0.04	0.568
D	0.571	0.04	0.042	0.041	0.039	0.547	0.041	0.04	0.041	0.039	0.04	0.592
E	0.569	0.04	0.04	0.041	0.044	0.578	0.041	0.041	0.041	0.04	0.04	0.58
F	0.564	0.041	0.041	0.041	0.04	0.576	0.041	0.04	0.041	0.04	0.04	0.566
G	0.576	0.039	0.04	0.04	0.04	0.558	0.04	0.04	0.043	0.04	0.04	0.56
H	0.643	0.04	0.04	0.04	0.043	0.603	0.041	0.041	0.041	0.039	0.04	0.644

Digester D – Supernatant Facility B	1	2	3	4	5	6	7	8	9	10	11	12
A	2.077	0.038	1.749	0.04	1.544	0.042	1.677	0.04	1.791	0.039	1.8	0.039
B	1.981	0.04	1.799	0.039	1.776	0.04	1.729	0.04	1.645	0.039	1.85	0.04
C	2.091	0.037	1.973	0.039	1.899	0.037	1.88	0.039	1.928	0.039	1.76	0.039
D	2.096	0.037	1.826	0.04	1.916	0.038	1.897	0.04	1.934	0.037	1.95	0.039
E	2.122	0.039	1.879	0.044	1.928	0.041	1.947	0.041	1.996	0.04	1.9	0.04
F	2.092	0.04	1.915	0.045	1.967	0.042	2.01	0.039	1.965	0.039	1.85	0.04
G	2.046	0.04	1.844	0.042	1.901	0.039	1.814	0.039	1.869	0.04	1.88	0.043
H	2.017	0.04	1.665	0.046	1.791	0.038	1.832	0.04	1.799	0.039	1.86	0.04
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.648	0.039	0.543	0.041	0.516	0.042	0.49	0.041	0.503	0.04	0.67	0.04
B	0.538	0.041	0.521	0.04	0.578	0.04	0.567	0.04	0.502	0.04	0.64	0.04
C	0.595	0.038	0.571	0.039	0.552	0.039	0.499	0.039	0.565	0.04	0.57	0.04
D	0.575	0.038	0.555	0.041	0.591	0.039	0.56	0.041	0.583	0.039	0.63	0.04
E	0.587	0.04	0.584	0.044	0.607	0.042	0.568	0.041	0.643	0.041	0.56	0.041
F	0.573	0.041	0.61	0.045	0.582	0.042	0.611	0.04	0.61	0.04	0.62	0.041
G	0.561	0.041	0.59	0.042	0.589	0.04	0.485	0.04	0.587	0.041	0.61	0.043
H	0.644	0.041	0.578	0.046	0.569	0.039	0.539	0.041	0.565	0.04	0.64	0.041

Digester D – Sludge **Facility B**	1	2	3	4	5	6	7	8	9	10	11	12
A	1.948	0.038	1.485	0.04	0.039	0.039	1.508	0.044	1.784	0.039	1.79	0.041
B	2.053	0.039	1.687	0.04	0.039	0.038	2.063	0.04	1.963	0.042	1.85	0.04
C	2.063	0.037	1.714	0.039	0.038	0.038	1.857	0.041	1.987	0.044	1.89	0.04
D	2.076	0.037	1.721	0.039	0.038	0.038	1.817	0.04	1.857	0.046	1.75	0.04
E	2.067	0.039	1.776	0.04	0.045	0.054	1.817	0.04	1.879	0.049	1.75	0.04
F	2.056	0.039	1.855	0.041	0.04	0.041	1.871	0.04	1.909	0.046	1.66	0.042
G	2.08	0.039	1.79	0.039	0.039	0.039	1.784	0.042	1.801	0.039	1.6	0.039
H	1.987	0.039	1.764	0.039	0.041	0.039	1.755	0.044	1.861	0.041	1.44	0.04
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.558	0.039	0.559	0.041	0.04	0.04	0.516	0.045	0.648	0.04	0.71	0.041
B	0.603	0.039	0.595	0.04	0.04	0.039	0.557	0.041	0.545	0.042	0.55	0.04
C	0.591	0.039	0.574	0.039	0.039	0.039	0.584	0.041	0.588	0.045	0.61	0.041
D	0.586	0.039	0.513	0.039	0.039	0.039	0.599	0.041	0.509	0.047	0.56	0.04
E	0.571	0.04	0.555	0.041	0.044	0.051	0.592	0.04	0.566	0.049	0.57	0.04
F	0.55	0.04	0.584	0.041	0.04	0.041	0.57	0.04	0.556	0.046	0.56	0.042
G	0.594	0.041	0.546	0.04	0.04	0.039	0.505	0.042	0.494	0.039	0.51	0.04
H	0.565	0.04	0.58	0.04	0.041	0.04	0.551	0.044	0.544	0.041	0.51	0.041

Digester D – Sludge (Total) **Facility B**	1	2	3	4	5	6	7	8	9	10	11	12
A	2.096	0.04	1.726	0.041	0.04	0.041	1.737	0.043	1.807	0.042	0.04	0.041
B	2.076	0.04	1.797	0.041	0.04	0.039	1.783	0.044	1.795	0.041	0.04	0.052
C	2.104	0.043	1.709	0.04	0.039	0.038	1.931	0.04	1.968	0.041	0.04	0.041
D	2.129	0.038	1.803	0.039	0.039	0.038	1.739	0.04	1.953	0.038	0.04	0.041
E	2.167	0.039	1.872	0.041	0.04	0.041	1.878	0.041	1.995	0.041	0.04	0.042
F	2.143	0.04	1.84	0.041	0.04	0.04	1.962	0.043	2.025	0.04	0.04	0.042
G	2.13	0.038	1.848	0.04	0.038	0.038	1.861	0.043	1.964	0.039	0.04	0.04
H	2.207	0.038	1.962	0.04	0.039	0.039	1.986	0.04	2.101	0.039	0.04	0.041
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.68	0.04	0.621	0.041	0.04	0.041	0.663	0.042	0.663	0.042	0.04	0.041
B	0.592	0.04	0.584	0.041	0.04	0.04	0.56	0.044	0.559	0.041	0.04	0.049
C	0.589	0.043	0.59	0.04	0.039	0.039	0.61	0.04	0.61	0.041	0.04	0.042
D	0.579	0.039	0.55	0.039	0.039	0.039	0.545	0.04	0.609	0.039	0.04	0.041
E	0.607	0.039	0.579	0.041	0.04	0.041	0.587	0.041	0.624	0.041	0.04	0.042
F	0.614	0.04	0.588	0.04	0.04	0.039	0.611	0.043	0.647	0.04	0.04	0.042
G	0.607	0.039	0.561	0.039	0.039	0.039	0.528	0.042	0.625	0.039	0.04	0.04
H	0.771	0.039	0.677	0.039	0.039	0.039	0.68	0.04	0.813	0.039	0.04	0.041

Standard Curve - Baseline Calculation												
Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
		2.086	2.002	1.682	1.297	0.819	0.745	0.77	0.774	0.753	0.75	0.76
		2.022	2.09	1.852	1.137	0.985	0.774	0.78	0.864	0.763	0.77	0.728
		2.096	2.155	1.968	1.15	0.878	0.823	0.758	0.74	0.753	0.71	0.749
Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
Averages		2.77	2.79	2.46	1.60	1.20	1.05	1.03	1.06	1.01	1.00	0.75
Standard Deviation		0.04	0.08	0.14	0.09	0.08	0.04	0.01	0.06	0.01	0.03	0.02
Standard Curve - Plate Calculations												
Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
		2.086	2.002	1.682	1.297	0.819	0.745	0.77	0.774	0.753	0.75	0.76
		2.022	2.09	1.852	1.137	0.985	0.774	0.78	0.864	0.763	0.77	0.728
		2.096	2.155	1.968	1.15	0.878	0.823	0.758	0.74	0.753	0.71	0.749
Averages at 520 nm		2.068	2.08233	1.834	1.19467	0.894	0.781	0.769	0.7927	0.756	0.74	0.746
Average at 690 nm for Plate 11 (MilliQ)	0.5335625											
Averages at 690 nm		0.667	0.60033	0.564333	0.547	0.51733	0.507	0.52	0.5393	0.519	0.51	0.516
Corrected Turbidity		1.934563	2.01556	1.803229	1.18123	0.91023	0.807	0.783	0.7869	0.771	0.76	0.764
Hexane Abs.	0.4959375											

Mass Balance for Facility A and B - May, 2001		
Facility A	Average E2 Conc.(M)	Average E2 Conc. (ng/L)
Raw Influent	5.24934E-11	14.27819578
Raw Influent (sonicated)	1.55557E-10	42.31160853
Primary Effluent	5.44177E-11	14.80160633
Primary Effluent (sonicated)	4.20199E-11	11.42940421
Pilot - Permeate	1.45779E-11	3.965199213
PAC effluent (sonicated)	1.30414E-11	3.547265649
GAC effluent (sonicated)	1.70873E-11	4.647744971
MLSS - 6th stage pilot	2.38322E-10	64.82359387
Mass Balance Calculations		
Influent Flow (gpd)	20,162	
Effluent Flow (gpd)	19,842	
MLSS Concentration (mg/L)	8,640	
Solid-based estrogenic activity (M)	2.24E-10	
Facility B		
Raw Influent	5.24934E-11	14.27819578
Raw Influent (sonicated)	1.55557E-10	42.31160853
Primary Effluent	5.44177E-11	14.80160633
Primary Effluent (sonicated)	4.20199E-11	11.42940421
Secondary Effluent	2.10662E-11	5.730014946
Secondary Effluent (sonicated)	1.99674E-11	5.431122379
MLSS - conventional	4.71764E-10	128.3197089
Digester Feed - Sludge	7.73636E-10	210.4290887
Primary Digester - Sludge	1.24547E-09	338.7673049
Secondary Digester - Sludge	1.53166E-09	416.6122196
Digester Feed centrate - Sludge	1.52366E-09	414.4362196
Primary Digester centrate - sludge	5.3027E-11	14.42335695
Secondary Digester centrate - sludge	4.04368E-11	10.99879794
Mass Balance Calculations		
Influent Flow (gpd)	3,558,971	
Effluent Flow (gpd)	3,140,817	
WAS (gpd)	40,964	
MLSS Concentration (mg/L)	3,924	
Digester Feed Solids Concentration (mg/L)	11,790	
Primary Digester Solids Concentration (mg/L)	15,156	
Secondary Digester Solids Concentration (mg/L)	8,010	
Digester Volume (L) - each	425,480	
Digester Volume (gallons) - each	112,412	
Volume Transferred (gallons/day)	5,621	
Solid-Based E2 Activity (M)		
Digester Feed	7.5257E-10	
Primary Digester Solids	1.19244E-09	
Secondary Digester Solids	1.49123E-09	

Facility C – March, 2001 Key
Plate 1 – Absorbance at 540 nm
Plate 2 – Absorbance at 690 nm

12	College Station - Raw Influent	College Station - MLSS	College Station - 2ndary Effluent
13	College Station - ATAD Feed	College Station - ATAD 1	College Station - ATAD 2
14	College Station - ATAD 3	College Station - DSHT 1	College Station - DSHT 2
15	Dewatered Sludge Filtrate		

Plate 12 – Facility C	1	2	3	4	5	6	7	8	9	10	11	12
A	1.08	0.866	0.041	0.043	0.04	0.953	0.888	0.042	0.041	0.04	0.865	0.973
B	1.257	1.022	0.04	0.042	0.04	0.96	0.867	0.04	0.043	0.04	0.874	0.842
C	0.992	0.927	0.039	0.04	0.038	1.026	0.972	0.04	0.039	0.039	0.924	0.901
D	1.029	1.08	0.04	0.039	0.04	1.02	1.041	0.04	0.041	0.042	0.987	0.905
E	1.01	1.057	0.04	0.04	0.04	1.067	1.014	0.041	0.04	0.045	0.907	0.897
F	1.045	0.897	0.04	0.042	0.041	0.934	0.94	0.04	0.04	0.04	0.914	0.895
G	0.941	0.985	0.039	0.039	0.038	0.943	0.955	0.04	0.04	0.039	0.915	0.865
H	0.967	0.945	0.044	0.04	0.038	1.053	0.998	0.04	0.044	0.039	0.853	1.007
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.634	0.48	0.041	0.043	0.04	0.552	0.501	0.041	0.04	0.039	0.488	0.595
B	0.642	0.542	0.04	0.041	0.039	0.52	0.443	0.04	0.042	0.039	0.476	0.45
C	0.546	0.489	0.039	0.039	0.038	0.564	0.533	0.039	0.039	0.038	0.502	0.489
D	0.579	0.616	0.039	0.039	0.04	0.534	0.558	0.039	0.04	0.042	0.529	0.494
E	0.579	0.601	0.039	0.04	0.039	0.573	0.544	0.04	0.04	0.044	0.506	0.475
F	0.623	0.661	0.039	0.041	0.041	0.48	0.481	0.039	0.04	0.04	0.486	0.483
G	0.541	0.55	0.039	0.039	0.038	0.507	0.525	0.039	0.039	0.038	0.49	0.469
H	0.592	0.55	0.042	0.04	0.038	0.635	0.611	0.04	0.044	0.039	0.472	0.614

Plate 13 – Facility C	1	2	3	4	5	6	7	8	9	10	11	12
A	1.076	1.059	0.04	0.04	0.039	1.09	1.16	0.041	0.04	0.039	1.091	1.141
B	0.984	1.036	0.041	0.04	0.039	1.033	1.012	0.04	0.041	0.039	0.96	1.038
C	0.942	1.012	0.041	0.039	0.038	1.022	1.041	0.039	0.039	0.038	1.082	1.064
D	0.982	1.073	0.051	0.039	0.038	0.974	1.149	0.039	0.04	0.041	1.026	1.121
E	1.002	1.028	0.04	0.04	0.039	1.205	1.084	0.041	0.04	0.039	1.068	1.095
F	1.008	1.068	0.04	0.04	0.04	1.056	1.177	0.04	0.04	0.039	1.099	1.062
G	0.97	1.155	0.04	0.039	0.039	1.211	1.145	0.04	0.04	0.039	1.044	1.067
H	1.067	1.053	0.04	0.04	0.04	1.09	1.148	0.041	0.043	0.042	1.106	1.052
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.737	0.712	0.04	0.041	0.04	0.726	0.787	0.042	0.041	0.04	0.723	0.782
B	0.615	0.64	0.041	0.041	0.04	0.612	0.578	0.041	0.041	0.04	0.523	0.629
C	0.566	0.606	0.041	0.04	0.039	0.535	0.577	0.04	0.04	0.039	0.627	0.626
D	0.595	0.654	0.047	0.04	0.039	0.498	0.643	0.04	0.04	0.041	0.555	0.643
E	0.607	0.61	0.04	0.04	0.04	0.712	0.59	0.041	0.04	0.04	0.596	0.589
F	0.643	0.656	0.04	0.04	0.041	0.594	0.685	0.04	0.041	0.04	0.641	0.6
G	0.607	0.763	0.041	0.04	0.039	0.76	0.682	0.041	0.04	0.039	0.603	0.633
H	0.73	0.689	0.04	0.041	0.041	0.698	0.691	0.042	0.044	0.043	0.719	0.68

Plate 14 – Facility C	1	2	3	4	5	6	7	8	9	10	11	12
A	1.566	1.586	0.041	0.041	0.04	2.148	2.124	0.043	0.042	0.041	1.039	1.11
B	1.404	1.602	0.04	0.042	0.04	2.377	2.142	0.042	0.042	0.041	1.024	1.053
C	1.602	1.795	0.04	0.04	0.038	2.376	2.364	0.04	0.04	0.042	1.057	1.053
D	1.442	1.599	0.039	0.039	0.039	2.361	2.426	0.04	0.041	0.04	1.137	0.984
E	1.761	1.75	0.041	0.041	0.04	2.478	2.332	0.042	0.041	0.041	1.075	1.07
F	1.606	1.775	0.04	0.041	0.04	2.306	2.391	0.041	0.041	0.041	1.112	1.026
G	1.389	1.302	0.04	0.041	0.039	2.323	2.299	0.041	0.041	0.04	1.118	1.063
H	1.089	1.259	0.04	0.041	0.04	2.238	2.129	0.044	0.043	0.04	1.024	0.905
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.678	0.789	0.041	0.041	0.04	0.631	0.606	0.042	0.041	0.04	0.638	0.714
B	0.703	0.684	0.04	0.042	0.04	0.714	0.555	0.041	0.041	0.04	0.571	0.622
C	0.66	0.771	0.039	0.039	0.039	0.694	0.71	0.04	0.04	0.041	0.578	0.609
D	0.626	0.72	0.039	0.039	0.039	0.656	0.749	0.04	0.041	0.039	0.647	0.56
E	0.697	0.728	0.04	0.04	0.039	0.786	0.656	0.041	0.041	0.04	0.574	0.659
F	0.73	0.779	0.04	0.04	0.039	0.661	0.717	0.04	0.041	0.04	0.628	0.59
G	0.634	0.648	0.039	0.041	0.039	0.739	0.676	0.041	0.04	0.039	0.675	0.653
H	0.536	0.766	0.04	0.04	0.039	0.818	0.667	0.044	0.041	0.039	0.654	0.565

Plate 15 – Facility C	1	2	3	4	5	6	7	8	9	10	11	12
A	0.895	0.842	0.042	0.042	0.041	0.041	0.042	0.043	0.05	0.042	0.044	0.052
B	0.859	0.905	0.043	0.042	0.04	0.041	0.041	0.041	0.042	0.04	0.041	0.047
C	0.936	0.873	0.04	0.04	0.039	0.04	0.042	0.04	0.04	0.04	0.039	0.04
D	0.876	0.924	0.049	0.04	0.041	0.039	0.041	0.04	0.047	0.039	0.039	0.041
E	0.942	0.873	0.041	0.041	0.04	0.041	0.041	0.041	0.041	0.04	0.04	0.042
F	0.97	0.904	0.041	0.041	0.041	0.04	0.041	0.041	0.041	0.041	0.04	0.043
G	0.996	0.857	0.041	0.04	0.039	0.039	0.041	0.041	0.041	0.039	0.039	0.041
H	0.998	0.981	0.041	0.04	0.044	0.039	0.04	0.041	0.041	0.04	0.04	0.04
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.519	0.494	0.041	0.041	0.04	0.04	0.041	0.042	0.049	0.041	0.043	0.048
B	0.474	0.524	0.043	0.041	0.04	0.041	0.041	0.041	0.041	0.04	0.041	0.045
C	0.549	0.501	0.04	0.04	0.039	0.04	0.042	0.04	0.04	0.04	0.039	0.04
D	0.519	0.532	0.048	0.04	0.041	0.039	0.041	0.04	0.045	0.039	0.039	0.041
E	0.521	0.472	0.04	0.041	0.04	0.041	0.041	0.041	0.041	0.04	0.04	0.041
F	0.554	0.498	0.041	0.04	0.041	0.04	0.041	0.04	0.041	0.041	0.04	0.042
G	0.574	0.473	0.041	0.04	0.039	0.039	0.04	0.041	0.041	0.039	0.039	0.041
H	0.608	0.611	0.041	0.04	0.043	0.039	0.04	0.041	0.041	0.04	0.04	0.04

Estradiol Control	1	2	3	4	5	6	7	8	9	10	11	12
A	2.075	2.133	2.102	2.081	1.83	1.385	1.043	0.955	0.912	0.895	0.882	0.882
B	2.081	2.155	2.169	2.071	1.857	1.379	1.074	0.984	0.959	0.871	0.911	0.928
C	0.041	0.033	0.034	0.033	0.035	0.032	0.036	0.036	0.033	0.033	0.031	0.036
Negative Control	0.939	0.874	0.954	0.971	0.891	0.832	0.814	0.879	0.881	0.837	0.888	0.864
E	0.043	0.04	0.041	0.041	0.041	0.041	0.041	0.042	0.041	0.042	0.04	0.043
Milli Q Blank	0.945	0.956	0.928	0.984	0.932	0.969	1.034	0.957	0.951	0.968	0.891	0.897
G	0.039	0.04	0.041	0.04	0.04	0.048	0.051	0.042	0.042	0.044	0.045	0.041
Solvent Blank	0.943	0.947	0.929	0.973	0.96	0.985	0.953	0.981	1.015	0.959	0.952	0.939
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.558	0.571	0.587	0.569	0.562	0.566	0.537	0.539	0.509	0.488	0.567	0.567
B	0.527	0.518	0.534	0.527	0.5	0.492	0.486	0.527	0.488	0.488	0.504	0.567
C	0.041	0.031	0.032	0.032	0.033	0.031	0.035	0.033	0.032	0.031	0.03	0.035
Negative Control	0.522	0.442	0.489	0.508	0.414	0.367	0.378	0.41	0.431	0.395	0.473	0.482
E	0.042	0.04	0.04	0.04	0.04	0.04	0.04	0.041	0.04	0.042	0.04	0.043
Milli Q Blank	0.539	0.512	0.551	0.528	0.482	0.546	0.572	0.506	0.494	0.52	0.474	0.504
G	0.039	0.039	0.04	0.04	0.04	0.047	0.049	0.042	0.041	0.043	0.044	0.041
Solvent Blank	0.539	0.516	0.501	0.522	0.518	0.542	0.524	0.541	0.573	0.531	0.534	0.55
Calculations with Standard Curve Plate												
Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
Estradiol Concentration (M)	2.50E-08	1.25E-08	6.25E-09	3.12408E-09	1.56388E-09	7.81938E-10	3.89134E-10	1.94567E-10	9.91189E-11	4.77239E-11	2.56975E-11	1.10132E-11
Average Absorbance	2.05E+00	2.12E+00	2.09E+00	2.05E+00	1.83E+00	1.37E+00	1.07E+00	9.56E-01	9.56E-01	9.14E-01	8.80E-01	8.57E-01
Milli-Q water Blank	0.519											
Solvent Blank Average	0.5325											
Negative Control (540 nm)	0.8853											
Negative Control (690 nm)	0.4425											
Negative Control	0.9753											

Mass Balance Facility C – March, 2001	**Average E2 Conc. (M)**	**Average E2 Conc.(ng/L)**
Raw Influent	4.39335E-11	11.94990211
Secondary Effluent	1.9469E-11	5.2955782
MLSS	3.91417E-11	10.64653048
Digester Feed	3.55843E-10	96.78924544
Digester 1	3.60693E-10	98.10845565
Digester 2	5.81096E-10	158.0580386
Digester 3	7.70637E-10	209.6131694
DSHT 1	2.28208E-09	620.7251042
DSHT 2	1.50738E-09	410.0076606
ATAD Feed - Centrate	1.11389E-10	
ATAD Digester 1 Centrate	5.29029E-11	
ATAD Digester 2 Centrate	5.60366E-11	
ATAD Digester 3 Centrate	4.54584E-11	
DSHT 1 Centrate	2.82034E-10	
DSHT 2 Centrate	3.23317E-10	
Mass Balance Calculations		
Influent Flow (gpd)	5,878,093	
Effluent Flow (gpd)	5,731,945	
Total Activated Sludge Volume (gallons)	3,946,000	
SRT (days)	9	
WAS (gpd)	146,148	
Feed Sludge Transferred (gpd)	17,760	
MLSS (mg/L)	3,293	
WAS (mg/L)	9,878	
Digester Feed (mg/L)	42,787	
Digester 1 (mg/L)	38,749	
Digester 2 (mg/L)	36,397	
Digester 3 (mg/L)	35,300	
DSHT 1 (mg/L)	28,008	
DSHT 2 (mg/L)	26,636	
Solid Based E2 Activity		
MLSS	1.96726E-11	
Digester Feed	2.44454E-10	
Digester 1	3.0779E-10	
Digester 2	5.25059E-10	
Digester 3	7.25178E-10	
DSHT 1	2.00004E-09	
DSHT 2	1.18406E-09	

Facility C – April, 2001

Raw	1	2	3	4	5	6	7	8	9	10	11	12
A	0.835	0.77	0.042	0.042	0.046	0.598	0.676	0.044	0.044	0.042	0.686	0.79
B	0.757	0.906	0.044	0.044	0.042	0.767	0.805	0.044	0.043	0.041	0.831	0.745
C	0.797	0.883	0.041	0.041	0.041	0.82	0.713	0.041	0.041	0.04	0.789	0.753
D	0.773	0.895	0.042	0.041	0.041	0.826	0.79	0.041	0.041	0.039	0.728	0.694
E	0.832	0.827	0.042	0.042	0.043	0.851	0.766	0.042	0.042	0.041	0.758	0.743
F	0.813	0.843	0.042	0.043	0.041	0.789	0.697	0.043	0.042	0.042	0.748	0.7
G	0.838	0.826	0.04	0.041	0.04	0.762	0.62	0.042	0.041	0.04	0.81	0.749
H	0.687	0.842	0.041	0.04	0.04	0.694	0.489	0.042	0.042	0.04	0.716	0.782
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.595	0.54	0.042	0.041	0.046	0.39	0.456	0.043	0.042	0.042	0.467	0.555
B	0.532	0.658	0.042	0.043	0.041	0.53	0.571	0.042	0.042	0.041	0.587	0.518
C	0.565	0.633	0.041	0.041	0.041	0.577	0.482	0.04	0.04	0.039	0.546	0.523
D	0.543	0.639	0.041	0.041	0.04	0.583	0.544	0.041	0.041	0.039	0.486	0.464
E	0.594	0.577	0.041	0.042	0.042	0.599	0.523	0.041	0.041	0.04	0.512	0.507
F	0.582	0.595	0.042	0.042	0.04	0.555	0.48	0.042	0.041	0.041	0.509	0.469
G	0.609	0.584	0.04	0.041	0.04	0.531	0.426	0.041	0.04	0.04	0.569	0.518
H	0.475	0.604	0.041	0.04	0.04	0.475	0.357	0.042	0.041	0.04	0.498	0.545

ATAD Feed	1	2	3	4	5	6	7	8	9	10	11	12
A	0.719	0.767	0.04	0.041	0.04	0.841	0.863	0.042	0.041	0.039	1.085	0.749
B	0.757	0.807	0.04	0.04	0.04	0.777	0.851	0.04	0.041	0.04	0.913	0.781
C	0.708	0.825	0.039	0.039	0.039	0.861	0.873	0.04	0.039	0.038	0.916	0.848
D	0.726	0.831	0.039	0.039	0.044	0.851	0.839	0.041	0.041	0.038	0.916	0.814
E	0.683	0.814	0.042	0.041	0.04	0.901	0.765	0.041	0.041	0.04	0.953	0.796
F	0.684	0.743	0.04	0.04	0.039	0.796	0.829	0.041	0.041	0.04	1.06	0.887
G	0.614	0.794	0.039	0.04	0.039	0.867	0.839	0.041	0.041	0.039	1.285	0.828
H	0.704	0.714	0.041	0.041	0.039	0.823	0.786	0.046	0.042	0.039	0.844	0.842
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.493	0.53	0.041	0.041	0.04	0.597	0.614	0.042	0.041	0.04	0.516	0.491
B	0.526	0.563	0.041	0.041	0.04	0.535	0.592	0.041	0.041	0.04	0.575	0.487
C	0.485	0.576	0.04	0.04	0.039	0.598	0.61	0.04	0.04	0.039	0.56	0.464
D	0.497	0.58	0.04	0.039	0.045	0.59	0.574	0.041	0.041	0.039	0.555	0.457
E	0.461	0.567	0.043	0.041	0.04	0.634	0.514	0.042	0.041	0.04	0.535	0.493
F	0.466	0.509	0.041	0.041	0.04	0.553	0.573	0.041	0.041	0.041	0.502	0.498
G	0.411	0.555	0.04	0.04	0.039	0.616	0.593	0.041	0.041	0.04	0.581	0.521
H	0.49	0.495	0.041	0.041	0.039	0.584	0.553	0.046	0.042	0.04	0.505	0.558

ATAD R1	1	2	3	4	5	6	7	8	9	10	11	12
A	0.782	0.788	0.04	0.041	0.041	0.745	0.625	0.043	0.041	0.043	0.857	0.767
B	0.728	0.859	0.041	0.042	0.041	0.912	0.835	0.04	0.047	0.039	0.801	0.775
C	0.735	0.795	0.044	0.04	0.038	0.92	0.826	0.039	0.039	0.038	0.806	0.778
D	0.701	0.856	0.04	0.039	0.039	0.905	0.822	0.039	0.041	0.038	0.81	0.778
E	0.718	0.843	0.042	0.044	0.04	0.47	1.528	0.041	0.04	0.039	0.827	0.802
F	0.678	0.773	0.04	0.04	0.039	0.789	0.689	0.041	0.041	0.04	0.844	0.847
G	0.755	0.501	0.04	0.04	0.039	0.657	0.565	0.04	0.04	0.039	0.834	0.811
H	0.757	0.794	0.04	0.041	0.042	0.657	0.724	0.041	0.041	0.041	0.82	0.876
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.556	0.552	0.041	0.041	0.042	0.513	0.413	0.042	0.041	0.043	0.601	0.527
B	0.506	0.612	0.041	0.042	0.042	0.572	0.583	0.041	0.048	0.04	0.546	0.527
C	0.508	0.557	0.045	0.04	0.039	0.654	0.568	0.04	0.04	0.039	0.544	0.529
D	0.481	0.604	0.041	0.04	0.039	0.632	0.565	0.04	0.041	0.039	0.546	0.53
E	0.497	0.589	0.042	0.044	0.04	0.284	1.094	0.041	0.041	0.04	0.573	0.557
F	0.463	0.537	0.041	0.041	0.04	0.544	0.403	0.041	0.041	0.04	0.594	0.596
G	0.524	0.311	0.04	0.04	0.039	0.424	0.36	0.041	0.041	0.04	0.588	0.567
H	0.531	0.565	0.041	0.042	0.044	0.425	0.488	0.041	0.041	0.042	0.579	0.625

ATAD R2	1	2	3	4	5	6	7	8	9	10	11	12
A	0.81	0.865	0.041	0.042	0.041	0.655	0.601	0.043	0.042	0.041	0.834	0.865
B	0.855	0.806	0.043	0.041	0.04	0.708	0.683	0.042	0.042	0.041	0.794	0.821
C	0.93	0.843	0.042	0.041	0.04	0.742	0.834	0.046	0.041	0.04	0.846	0.834
D	0.865	0.927	0.04	0.041	0.039	0.855	0.971	0.04	0.04	0.038	0.831	0.884
E	0.938	0.914	0.041	0.044	0.04	0.883	0.958	0.041	0.041	0.039	0.864	0.868
F	0.875	0.834	0.041	0.042	0.04	0.879	0.906	0.041	0.042	0.04	0.804	0.854
G	0.858	0.855	0.041	0.04	0.039	0.885	0.963	0.042	0.041	0.041	0.797	0.897
H	0.845	0.932	0.043	0.042	0.042	0.844	0.687	0.04	0.041	0.039	0.927	0.892
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.575	0.617	0.041	0.041	0.04	0.416	0.362	0.042	0.041	0.041	0.598	0.619
B	0.612	0.565	0.042	0.041	0.04	0.469	0.359	0.041	0.041	0.041	0.561	0.581
C	0.663	0.589	0.041	0.041	0.04	0.447	0.513	0.045	0.04	0.04	0.598	0.579
D	0.612	0.668	0.04	0.041	0.039	0.572	0.691	0.04	0.04	0.039	0.575	0.619
E	0.67	0.657	0.041	0.043	0.041	0.56	0.592	0.041	0.041	0.039	0.604	0.596
F	0.603	0.582	0.041	0.041	0.04	0.589	0.62	0.04	0.041	0.04	0.545	0.579
G	0.602	0.57	0.041	0.04	0.039	0.615	0.594	0.042	0.041	0.04	0.544	0.628
H	0.597	0.62	0.042	0.042	0.042	0.592	0.456	0.04	0.04	0.04	0.664	0.633

ATAD R3	1	2	3	4	5	6	7	8	9	10	11	12
A	0.983	0.947	0.041	0.041	0.04	0.637	0.77	0.041	0.041	0.04	0.859	0.86
B	0.954	0.794	0.041	0.041	0.04	0.858	0.771	0.04	0.041	0.041	0.922	0.834
C	0.977	0.856	0.039	0.039	0.038	0.872	0.848	0.04	0.041	0.038	0.812	0.85
D	1	0.871	0.039	0.039	0.038	0.917	1.1	0.039	0.04	0.038	0.833	0.832
E	0.989	0.879	0.04	0.041	0.04	0.894	0.972	0.041	0.041	0.04	0.751	1.016
F	0.907	0.856	0.04	0.04	0.039	0.813	0.967	0.04	0.041	0.048	0.818	0.865
G	0.884	0.895	0.041	0.04	0.039	0.814	0.931	0.04	0.04	0.039	0.821	0.911
H	0.909	0.822	0.04	0.04	0.039	0.856	0.955	0.042	0.041	0.041	0.823	0.89
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.693	0.623	0.041	0.041	0.04	0.355	0.484	0.042	0.041	0.041	0.621	0.622
B	0.681	0.546	0.041	0.041	0.04	0.522	0.467	0.041	0.041	0.041	0.676	0.597
C	0.662	0.551	0.04	0.04	0.039	0.518	0.506	0.04	0.041	0.039	0.57	0.609
D	0.664	0.602	0.039	0.039	0.039	0.527	0.62	0.04	0.041	0.039	0.586	0.589
E	0.689	0.57	0.04	0.041	0.04	0.582	0.635	0.041	0.041	0.04	0.517	0.746
F	0.657	0.54	0.04	0.04	0.04	0.535	0.618	0.041	0.041	0.05	0.581	0.63
G	0.575	0.514	0.042	0.04	0.039	0.513	0.619	0.041	0.04	0.039	0.589	0.673
H	0.663	0.552	0.04	0.04	0.039	0.564	0.621	0.042	0.041	0.041	0.59	0.651

DSHT 1	1	2	3	4	5	6	7	8	9	10	11	12
A	1.384	1.514	0.041	0.042	0.041	2.064	2.195	0.043	0.043	0.041	1.255	1.203
B	1.772	1.66	0.044	0.043	0.041	2.093	2.246	0.041	0.042	0.041	1.357	1.647
C	1.908	2.115	0.042	0.044	0.043	2.165	2.25	0.041	0.041	0.039	1.562	2.101
D	1.985	1.729	0.043	0.042	0.043	2.205	2.118	0.041	0.042	0.043	1.626	1.819
E	1.55	2.054	0.041	0.041	0.04	2.208	2.185	0.042	0.041	0.04	1.511	1.529
F	1.553	1.712	0.041	0.042	0.04	2.222	2.172	0.041	0.043	0.04	1.286	1.457
G	1.805	2.023	0.041	0.041	0.041	2.257	2.153	0.041	0.041	0.04	1.074	1.176
H	1.894	2.01	0.043	0.04	0.04	2.092	2.159	0.042	0.042	0.041	1.02	0.932
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.458	0.632	0.041	0.041	0.04	0.591	0.583	0.042	0.042	0.04	0.623	0.661
B	0.595	0.58	0.042	0.041	0.04	0.558	0.629	0.04	0.041	0.04	0.48	0.585
C	0.574	0.584	0.041	0.042	0.042	0.542	0.61	0.04	0.04	0.039	0.479	0.593
D	0.612	0.571	0.041	0.041	0.043	0.569	0.558	0.04	0.041	0.042	0.51	0.57
E	0.566	0.527	0.04	0.041	0.04	0.586	0.531	0.041	0.04	0.04	0.542	0.551
F	0.573	0.577	0.04	0.042	0.04	0.595	0.536	0.04	0.042	0.04	0.531	0.561
G	0.655	0.544	0.04	0.04	0.04	0.63	0.538	0.04	0.04	0.039	0.495	0.555
H	0.478	0.532	0.042	0.04	0.04	0.588	0.545	0.041	0.041	0.04	0.517	0.443

DSHT 2	1	2	3	4	5	6	7	8	9	10	11	12
A	1.495	1.907	0.046	0.047	0.046	1.499	1.727	0.046	0.048	0.044	0.916	0.945
B	1.448	1.426	0.048	0.05	0.049	1.758	2.134	0.046	0.048	0.046	1.178	1.338
C	1.642	1.467	0.048	0.048	0.044	2.068	2.133	0.046	0.043	0.043	1.464	2.434
D	1.431	1.221	0.048	0.048	0.045	2.113	2.004	0.042	0.041	0.04	1.624	2.231
E	1.317	1.733	0.044	0.045	0.044	1.854	1.945	0.044	0.044	0.039	1.667	2.297
F	1.831	1.49	0.045	0.048	0.044	1.886	2.058	0.045	0.044	0.041	1.601	2.37
G	1.356	1.334	0.044	0.046	0.042	1.94	1.795	0.044	0.045	0.042	1.456	1.522
H	1.887	1.532	0.044	0.043	0.042	1.555	2.038	0.045	0.047	0.043	1.175	1.311
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.588	0.61	0.043	0.044	0.043	0.522	0.562	0.044	0.045	0.043	0.527	0.547
B	0.589	0.561	0.045	0.046	0.046	0.598	0.598	0.043	0.044	0.043	0.504	0.655
C	0.556	0.562	0.044	0.045	0.042	0.555	0.525	0.045	0.042	0.042	0.546	0.998
D	0.603	0.473	0.045	0.044	0.042	0.547	0.574	0.041	0.04	0.039	0.569	0.83
E	0.579	0.498	0.042	0.043	0.043	0.583	0.555	0.042	0.043	0.039	0.566	0.863
F	0.648	0.549	0.043	0.046	0.043	0.6	0.552	0.043	0.042	0.041	0.529	0.965
G	0.628	0.569	0.042	0.044	0.041	0.547	0.588	0.042	0.043	0.041	0.573	0.649
H	0.639	0.547	0.043	0.041	0.041	0.521	0.557	0.043	0.046	0.041	0.444	0.541

Estradiol Control	1	2	3	4	5	6	7	8	9	10	11	12
A	2.023	2.005	2.004	1.431	1.111	0.962	0.89	0.844	0.814	0.866	0.66	0.722
B	2.01	2.045	1.901	1.482	1.1	0.891	0.785	0.796	0.805	0.756	0.799	0.741
C	0.042	0.035	0.039	0.24	0.238	0.051	0.039	0.24	0.042	0.041	0.034	0.047
Negative Control	0.867	0.828	0.847	0.755	0.748	0.747	0.851	0.793	0.746	0.771	0.699	0.71
E	0.042	0.049	0.046	0.046	0.043	0.042	0.044	0.043	0.044	0.041	0.041	0.042
Milli Q Blank	0.849	0.826	0.809	0.808	0.779	0.77	0.808	0.814	0.742	0.736	0.737	0.686
G	0.044	0.04	0.044	0.047	0.043	0.041	0.044	0.043	0.042	0.04	0.04	0.052
Solvent Blank	0.784	0.845	0.767	0.756	0.756	0.793	0.719	0.728	0.676	0.662	0.71	0.724
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.577	0.593	0.717	0.59	0.634	0.644	0.633	0.603	0.584	0.631	0.458	0.505
B	0.575	0.556	0.567	0.591	0.575	0.553	0.53	0.556	0.572	0.53	0.572	0.527
C	0.041	0.033	0.036	0.227	0.23	0.043	0.037	0.23	0.039	0.038	0.033	0.041
Negative Control	0.632	0.596	0.611	0.527	0.519	0.518	0.609	0.556	0.515	0.539	0.477	0.495
E	0.041	0.046	0.044	0.044	0.042	0.041	0.043	0.042	0.042	0.041	0.041	0.042
Milli Q Blank	0.617	0.595	0.582	0.576	0.55	0.546	0.573	0.58	0.519	0.515	0.517	0.475
G	0.043	0.039	0.042	0.046	0.042	0.04	0.043	0.041	0.041	0.04	0.04	0.051
Solvent Blank	0.548	0.611	0.543	0.532	0.534	0.565	0.499	0.507	0.464	0.454	0.498	0.508

Continued from previous page

Calculations with Standard Curve Plate												
Estradiol Concentration (ng/L)	6,810	3,405	1,703	851	426	213	106	53	27	13	7	3
Estradiol Concentration (M)	2.50E-08	1.25E-08	6.25E-09	3.12408E-09	1.56388E-09	7.81938E-10	3.89134E-10	1.94567E-10	9.91189E-11	4.77239E-11	2.56975E-11	1.10132E-11
Average Absorbance	1.99E+00	2.00E+00	1.86E+00	1.42E+00	1.05E+00	8.82E-01	8.10E-01	7.94E-01	7.85E-01	7.84E-01	7.68E-01	7.69E-01
Milli-Q water Blank (690 nm)	0.55375											
Solvent Blank (690 nm)	0.521916667											
Negative Control (540 nm)	0.780166667											
Negative Control (690 nm)	0.5495											
Negative Control Turbidity	0.752583333											

Facility C – April, 2001	**Average E2 Conc. (M)**	**Average E2 Conc. (ng/L)**
Raw Influent	1.49374E-10	40.62966718
Secondary Effluent	6.61947E-11	18.00496588
MLSS	1.33082E-10	36.19820364
Digester Feed	1.20987E-09	329.0834345
Digester 1	1.22636E-09	333.5687492
Digester 2	1.97573E-09	537.3973311
Digester 3	2.62016E-09	712.684776
DSHT 1	7.75906E-09	2110.465354
DSHT 2	5.1251E-09	1394.026046
ATAD Feed - Centrate	3.78722E-10	103.0125196
ATAD Digester 1 Centrate	1.7987E-10	48.92458557
ATAD Digester 2 Centrate	1.90525E-10	51.82267392
ATAD Digester 3 Centrate	1.54558E-10	42.03988303
DSHT 1 Centrate	9.58915E-10	260.8248202
DSHT 2 Centrate	1.09928E-09	299.0035064
Mass Balance Calculations		
Influent Flow (gpd)	4,996,379	
Effluent Flow (gpd)	4,642,875	
Total Activated Sludge Volume (gallons)	3,946,000	
SRT (days)	9	
WAS (gpd)	146,148	
Feed Sludge Transferred (gpd)	17,760	
MLSS (mg/L)	3,763	
WAS (mg/L)	11,290	
Digester Feed (mg/L)	48,899	
Digester 1 (mg/L)	44,285	
Digester 2 (mg/L)	41,597	
Digester 3 (mg/L)	40,342	
DSHT 1 (mg/L)	32,010	
DSHT 2 (mg/L)	30,442	
Solid Based E2 Activity		
MLSS	6.68869E-11	
Digester Feed	8.31143E-10	
Digester 1	1.04649E-09	
Digester 2	1.7852E-09	
Digester 3	2.46561E-09	
DSHT 1	6.80015E-09	
DSHT 2	4.02582E-09	

Facility C – April, 2001
Plate 1 – Absorbance at 540 nm
Plate 2 – Absorbance at 690 nm

Raw	1	2	3	4	5	6	7	8	9	10	11	12
A	0.952	0.888	0.042	0.042	0.041	0.721	0.796	0.046	0.045	0.041	0.806	0.896
B	0.866	1.022	0.047	0.048	0.044	0.89	0.922	0.044	0.044	0.041	0.951	0.857
C	0.917	1.007	0.046	0.044	0.042	0.95	0.845	0.042	0.042	0.041	0.919	0.869
D	0.904	1.032	0.043	0.042	0.042	0.96	0.932	0.042	0.041	0.039	0.874	0.83
E	0.954	0.97	0.044	0.043	0.042	0.997	0.907	0.043	0.042	0.042	0.904	0.876
F	0.938	0.979	0.042	0.045	0.044	0.91	0.811	0.043	0.042	0.042	0.885	0.838
G	0.954	0.948	0.041	0.042	0.041	0.884	0.72	0.043	0.042	0.041	0.94	0.869
H	0.812	0.946	0.045	0.043	0.042	0.809	0.553	0.044	0.043	0.04	0.826	0.897
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.589	0.532	0.042	0.042	0.041	0.39	0.454	0.044	0.043	0.041	0.466	0.54
B	0.527	0.644	0.044	0.046	0.043	0.52	0.56	0.043	0.043	0.041	0.573	0.505
C	0.562	0.619	0.044	0.042	0.041	0.566	0.474	0.042	0.041	0.04	0.533	0.51
D	0.549	0.627	0.042	0.041	0.041	0.57	0.534	0.041	0.041	0.039	0.477	0.455
E	0.584	0.568	0.043	0.042	0.042	0.586	0.512	0.042	0.042	0.042	0.501	0.493
F	0.586	0.584	0.042	0.044	0.043	0.544	0.473	0.042	0.042	0.041	0.497	0.458
G	0.608	0.574	0.041	0.041	0.04	0.521	0.421	0.042	0.042	0.04	0.552	0.505
H	0.49	0.588	0.044	0.042	0.041	0.468	0.378	0.043	0.042	0.04	0.484	0.533

ATAD Feed	1	2	3	4	5	6	7	8	9	10	11	12
A	0.932	0.926	0.041	0.052	0.043	0.991	1	0.044	0.044	0.041	1.772	0.992
B	0.902	0.945	0.042	0.042	0.042	0.922	0.991	0.041	0.042	0.041	1.278	1.063
C	0.856	0.976	0.044	0.042	0.039	1.024	1.009	0.04	0.04	0.04	1.275	1.287
D	0.955	0.98	0.042	0.041	0.04	1.036	1	0.041	0.041	0.038	1.277	1.204
E	0.874	0.944	0.041	0.041	0.04	1.076	0.913	0.041	0.041	0.042	1.403	1.065
F	0.846	0.873	0.041	0.041	0.04	0.934	0.969	0.041	0.041	0.04	1.631	1.292
G	0.782	0.92	0.04	0.041	0.043	0.995	0.954	0.041	0.042	0.04	1.964	1.081
H	0.839	0.831	0.042	0.041	0.04	0.936	0.887	0.045	0.042	0.039	1.307	1.04
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.561	0.547	0.041	0.049	0.041	0.606	0.611	0.042	0.041	0.04	0.581	0.528
B	0.533	0.55	0.041	0.041	0.04	0.525	0.564	0.04	0.041	0.04	0.566	0.488
C	0.488	0.568	0.041	0.04	0.038	0.588	0.581	0.039	0.039	0.039	0.543	0.462
D	0.572	0.571	0.04	0.04	0.039	0.597	0.557	0.04	0.04	0.038	0.539	0.452
E	0.501	0.549	0.04	0.04	0.039	0.635	0.5	0.04	0.04	0.041	0.522	0.466
F	0.489	0.5	0.04	0.04	0.039	0.545	0.557	0.04	0.04	0.04	0.492	0.475
G	0.445	0.542	0.039	0.04	0.043	0.605	0.572	0.04	0.041	0.039	0.632	0.492
H	0.501	0.491	0.041	0.04	0.039	0.567	0.532	0.043	0.041	0.039	0.578	0.529

ATAD R1	1	2	3	4	5	6	7	8	9	10	11	12
A	0.939	0.92	0.042	0.042	0.041	0.894	0.8	0.043	0.042	0.048	1.002	0.938
B	1.026	0.984	0.042	0.041	0.041	1.489	1.008	0.041	0.042	0.04	0.999	0.932
C	0.872	0.934	0.04	0.041	0.038	1.07	1.045	0.039	0.039	0.038	0.974	0.943
D	0.843	1.096	0.041	0.04	0.039	1.089	0.973	0.039	0.041	0.038	0.985	0.954
E	0.85	1.034	0.041	0.044	0.04	0.674	1.801	0.041	0.041	0.04	0.979	0.964
F	0.811	0.887	0.041	0.04	0.039	0.93	0.963	0.041	0.042	0.04	0.982	1.002
G	0.947	0.703	0.04	0.041	0.039	0.814	0.727	0.041	0.041	0.039	0.967	0.949
H	0.871	0.902	0.041	0.041	0.042	0.818	0.889	0.041	0.041	0.04	0.943	1.009
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.591	0.55	0.042	0.042	0.041	0.502	0.435	0.042	0.042	0.047	0.593	0.554
B	0.609	0.594	0.041	0.042	0.041	0.552	0.583	0.041	0.041	0.04	0.554	0.515
C	0.506	0.547	0.04	0.041	0.039	0.636	0.594	0.04	0.04	0.039	0.529	0.52
D	0.489	0.622	0.041	0.04	0.04	0.627	0.544	0.04	0.041	0.039	0.53	0.525
E	0.491	0.571	0.041	0.044	0.04	0.308	1.183	0.042	0.041	0.04	0.556	0.548
F	0.465	0.516	0.041	0.041	0.04	0.532	0.394	0.041	0.042	0.04	0.576	0.587
G	0.572	0.342	0.04	0.041	0.039	0.411	0.365	0.041	0.041	0.039	0.571	0.556
H	0.53	0.551	0.041	0.041	0.041	0.421	0.49	0.041	0.041	0.04	0.568	0.614

ATAD R2	1	2	3	4	5	6	7	8	9	10	11	12
A	0.991	0.991	0.042	0.045	0.042	0.85	0.845	0.044	0.043	0.042	0.935	1.047
B	1.023	1.013	0.045	0.043	0.042	0.997	1.144	0.045	0.044	0.041	0.92	0.956
C	1.183	1.046	0.044	0.043	0.041	1.245	1.123	0.046	0.043	0.041	0.994	0.999
D	1.009	1.084	0.041	0.042	0.039	1.109	1.193	0.041	0.04	0.04	0.986	1.045
E	1.094	1.072	0.042	0.041	0.04	1.198	1.495	0.042	0.042	0.04	1.022	1.035
F	1.036	0.999	0.042	0.042	0.041	1.15	1.114	0.042	0.042	0.041	0.978	1.039
G	1.013	0.955	0.041	0.042	0.041	1.076	1.188	0.042	0.041	0.041	0.95	1.061
H	0.907	0.901	0.044	0.043	0.042	1.031	0.885	0.041	0.042	0.041	1.043	1.017
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.611	0.614	0.041	0.05	0.041	0.437	0.352	0.042	0.042	0.041	0.591	0.668
B	0.634	0.538	0.042	0.042	0.041	0.514	0.341	0.042	0.042	0.04	0.545	0.584
C	0.641	0.572	0.042	0.041	0.04	0.437	0.493	0.044	0.041	0.04	0.592	0.578
D	0.589	0.657	0.04	0.041	0.039	0.577	0.675	0.04	0.04	0.039	0.562	0.607
E	0.646	0.648	0.041	0.04	0.04	0.526	0.567	0.04	0.04	0.039	0.59	0.575
F	0.565	0.571	0.04	0.041	0.04	0.567	0.594	0.04	0.041	0.04	0.535	0.565
G	0.583	0.448	0.04	0.041	0.039	0.601	0.468	0.041	0.04	0.04	0.53	0.613
H	0.523	0.388	0.042	0.041	0.041	0.587	0.438	0.04	0.04	0.04	0.632	0.601

ATAD R3	1	2	3	4	5	6	7	8	9	10	11	12
A	1.118	1.416	0.041	0.044	0.042	1.09	0.947	0.044	0.041	0.041	1.089	1.001
B	1.091	1.041	0.043	0.042	0.041	1.376	0.997	0.042	0.043	0.041	1.049	1.026
C	1.127	1.116	0.04	0.04	0.038	1.493	1.288	0.04	0.043	0.041	0.969	1.042
D	1.177	1.208	0.042	0.041	0.04	1.529	1.93	0.04	0.041	0.038	1.003	1.037
E	1.194	1.143	0.042	0.041	0.041	1.326	1.447	0.041	0.041	0.041	0.867	0.949
F	1.159	0.93	0.042	0.045	0.04	1.038	1.544	0.044	0.043	0.04	0.917	0.97
G	1.103	1.614	0.039	0.041	0.04	1.13	1.156	0.047	0.041	0.04	0.855	0.988
H	0.93	1.144	0.04	0.04	0.039	1.18	1.358	0.043	0.043	0.043	0.869	0.914
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.588	0.614	0.041	0.043	0.042	0.381	0.456	0.043	0.041	0.041	0.726	0.658
B	0.651	0.534	0.042	0.042	0.041	0.486	0.445	0.041	0.042	0.041	0.665	0.659
C	0.582	0.518	0.04	0.04	0.039	0.493	0.591	0.04	0.042	0.04	0.573	0.664
D	0.527	0.596	0.042	0.041	0.04	0.526	0.539	0.04	0.042	0.039	0.584	0.651
E	0.651	0.557	0.042	0.041	0.041	0.56	0.59	0.041	0.041	0.04	0.494	0.551
F	0.625	0.422	0.042	0.044	0.04	0.439	0.485	0.043	0.042	0.04	0.552	0.624
G	0.475	0.482	0.04	0.041	0.039	0.495	0.62	0.047	0.04	0.04	0.515	0.644
H	0.529	0.52	0.04	0.04	0.039	0.534	0.588	0.042	0.042	0.042	0.533	0.577

DSHT 1	1	2	3	4	5	6	7	8	9	10	11	12
A	1.925	2.188	0.042	0.043	0.042	2.187	2.134	0.047	0.048	0.044	1.819	1.731
B	2.184	2.186	0.045	0.045	0.043	2.101	2.169	0.043	0.043	0.042	1.88	2.06
C	2.103	2.178	0.044	0.045	0.044	2.193	2.18	0.042	0.042	0.042	1.98	2.102
D	2.138	2.165	0.043	0.042	0.04	2.159	2.011	0.042	0.042	0.043	2.011	2.077
E	2.103	2.095	0.042	0.042	0.042	2.182	2.136	0.042	0.042	0.042	2.03	2.001
F	2.063	2.108	0.043	0.042	0.041	2.145	2.113	0.042	0.043	0.041	1.888	1.97
G	2.184	2.014	0.042	0.043	0.042	2.219	2.128	0.044	0.042	0.04	1.655	1.748
H	1.982	1.923	0.044	0.042	0.042	2.012	2.099	0.044	0.043	0.042	1.556	1.466
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.607	0.652	0.041	0.041	0.041	0.594	0.52	0.044	0.045	0.042	0.608	0.638
B	0.652	0.588	0.042	0.042	0.041	0.538	0.557	0.041	0.042	0.041	0.472	0.567
C	0.526	0.576	0.042	0.042	0.042	0.524	0.544	0.041	0.041	0.04	0.476	0.578
D	0.572	0.56	0.041	0.04	0.039	0.501	0.537	0.041	0.04	0.041	0.501	0.555
E	0.576	0.511	0.04	0.041	0.04	0.535	0.477	0.041	0.04	0.041	0.531	0.534
F	0.588	0.558	0.041	0.04	0.039	0.505	0.465	0.041	0.042	0.04	0.52	0.538
G	0.637	0.44	0.04	0.041	0.04	0.573	0.498	0.041	0.041	0.039	0.485	0.528
H	0.457	0.379	0.042	0.04	0.04	0.579	0.508	0.042	0.041	0.04	0.507	0.434

DSHT 2	1	2	3	4	5	6	7	8	9	10	11	12
A	2	2.278	0.046	0.047	0.046	2.058	2.105	0.047	0.048	0.045	1.328	1.362
B	1.964	2.006	0.048	0.05	0.048	2.182	2.158	0.046	0.046	0.045	1.761	1.93
C	1.957	2.081	0.048	0.048	0.044	2.149	2.104	0.046	0.043	0.043	2.003	2.563
D	2.042	1.978	0.048	0.048	0.045	2.148	2.243	0.042	0.043	0.043	2.077	2.422
E	1.891	1.995	0.044	0.046	0.044	2.152	2.162	0.047	0.043	0.04	2.105	2.426
F	2.091	1.997	0.047	0.048	0.046	2.073	2.093	0.045	0.044	0.042	2.045	2.472
G	1.986	2.015	0.046	0.046	0.043	2.103	2.175	0.044	0.046	0.043	1.99	2.047
H	1.993	1.846	0.046	0.043	0.043	2.023	2.008	0.046	0.048	0.044	1.757	1.86
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.607	0.627	0.044	0.044	0.043	0.538	0.592	0.044	0.045	0.043	0.518	0.541
B	0.559	0.558	0.045	0.046	0.045	0.59	0.578	0.044	0.043	0.042	0.491	0.645
C	0.475	0.524	0.045	0.045	0.042	0.507	0.498	0.044	0.042	0.042	0.534	1.041
D	0.543	0.483	0.044	0.045	0.043	0.509	0.59	0.041	0.041	0.041	0.556	0.917
E	0.565	0.461	0.042	0.044	0.043	0.584	0.561	0.044	0.042	0.04	0.553	0.926
F	0.54	0.517	0.045	0.045	0.044	0.544	0.51	0.043	0.042	0.041	0.523	0.983
G	0.606	0.544	0.044	0.044	0.041	0.499	0.585	0.042	0.044	0.041	0.56	0.634
H	0.492	0.386	0.044	0.042	0.042	0.489	0.485	0.044	0.046	0.042	0.449	0.538

Facility C – May, 2001	**Average E2 Conc. (M)**	**Average E2 Conc. (ng/L)**
Raw Influent	7.02935E-11	19.11984338
Secondary Effluent	3.11505E-11	8.472925119
MLSS	6.26266E-11	17.03444877
Digester Feed	5.69349E-10	154.8627927
Digester 1	5.77109E-10	156.973529
Digester 2	9.29753E-10	252.8928617
Digester 3	1.23302E-09	335.381071
DSHT 1	3.65132E-09	993.1601667
DSHT 2	2.41181E-09	656.012257
ATAD Feed - Centrate	1.78222E-10	48.47647979
ATAD Digester 1 Centrate	8.46446E-11	23.02333438
ATAD Digester 2 Centrate	8.96586E-11	24.38714067
ATAD Digester 3 Centrate	7.27334E-11	19.78347437
DSHT 1 Centrate	4.51254E-10	122.7410919
DSHT 2 Centrate	5.17307E-10	140.7075324
Mass Balance Calculations		
Influent Flow (gpd)	5,878,093	
Effluent Flow (gpd)	5,731,945	
Total Activated Sludge Volume (gallons)	3,946,000	
SRT (days)	9	
WAS (gpd) - CALCULATED	146,148	
Feed Sludge Transferred (gpd)	17,760	
MLSS (mg/L)	3,024	
WAS (mg/L)	9,072	
Digester Feed (mg/L)	39,294	
Digester 1 (mg/L)	35,586	
Digester 2 (mg/L)	33,426	
Digester 3 (mg/L)	32,418	
DSHT 1 (mg/L)	25,722	
DSHT 2 (mg/L)	24,462	
Solid Based E2 Activity		
MLSS	3.14762E-11	
Digester Feed	3.91126E-10	
Digester 1	4.92464E-10	
Digester 2	8.40095E-10	
Digester 3	1.16029E-09	
DSHT 1	3.20007E-09	
DSHT 2	1.8945E-09	

Appendix B (Chapter 4)
Fluorescence Quenching of Pyrene by Colloidal Organic Carbon from Activated Sludge Systems

Sample Calculation for Fluoresence Quenching

1. Measure fluorescence intensity of sample (Observed FI).
2. Measure absorbance of the sample at the excitation and emission wavelengths (Abs. at Excit. And Abs. at Emission).
3. Use inner filter effect correction formula from MacDonald et al. (reference 28 in Chapter 4) to find the true FI (Ideal FI).
4. Subtract the Ideal FI of the background (Foback) and spiked sample containing no colloidal material (Fospike) to calculate the FI of pyrene (Fo).
5. Repeat steps 1 through 4 for solutions containing different concentrations of colloidal organic carbon (COC) to calculate the FI of pyrene in the presence of static quenchers (F).
6. Take the ratio of Fo/F as a function of COC concentration and construct a Stern-Volmer Plot.

7.

< 1.5 um CAS for Pyrene (June, 2002)							
Controls (Fospike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI (Fo)**		
	101.3	0.0166	0.0012	102.9540315	95.84710247		
	82.9	0.0226	0.0063	85.22815062	78.35709405		
	70	0.0201	0.0054	71.71280933	64.8892848		
	60.7	0.0179	0.0041	61.96015847	55.55397245		
	53.1	0.0171	0.0046	54.18553709	47.88691269		
	47.2	0.0164	0.0039	48.0951076	41.89036407		
Controls (Foback)							
	7	0.0161	0.0007	7.106928985			
	6.7	0.0189	0.0076	6.871056568			
	6.7	0.0149	0.0049	6.823524535			
	6.3	0.0141	0.0042	6.406186015			
	6.2	0.0135	0.0039	6.298624401			
	6.1	0.0138	0.0048	6.204743528			
COC Samples						**COC (mg/L)**	**Fo/F**
Fspike	97	0.0398	0.0087	101.7697566	94.37061948	2.001997004	1.015645579
	78.9	0.0343	0.0056	82.04547414	73.06865539	3.204155014	1.072376296
	66.1	0.0421	0.0176	70.1597249	60.0006783	4.005994006	1.081475854
	57.3	0.0454	0.0189	61.10975964	50.29910804	4.578932344	1.104472318
	50.8	0.0478	0.0203	54.39128021	43.17457777	5.008743442	1.109146057
	45.2	0.0511	0.0224	48.66687147	36.98662366	5.343104597	1.132581456
Blanks							
Fback	7.2	0.0217	0.0068	7.399137136			
	8.7	0.0277	0.0047	8.976818754			
	9.6	0.0396	0.0172	10.1590466			
	10.1	0.0489	0.0189	10.8106516			
	10.5	0.0449	0.021	11.21670245			

	10.9	0.0473	0.0216	11.68024781			

< 0.22 um CAS for Pyrene (June, 2002)							
Controls (Fo spike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI (Fo)**		
	101.3	0.0166	0.0012	102.9540315	95.84710247		
	82.9	0.0226	0.0063	85.22815062	78.35709405		
	70	0.0201	0.0054	71.71280933	64.8892848		
	60.7	0.0179	0.0041	61.96015847	55.55397245		
	53.1	0.0171	0.0046	54.18553709	47.88691269		
	47.2	0.0164	0.0039	48.0951076	41.89036407		
Controls (Fo back)							
	7	0.0161	0.0007	7.106928985			
	6.7	0.0189	0.0076	6.871056568			
	6.7	0.0149	0.0049	6.823524535			
	6.3	0.0141	0.0042	6.406186015			
	6.2	0.0135	0.0039	6.298624401			
	6.1	0.0138	0.0048	6.204743528			
COC Samples						**COC (mg/L)**	**Fo/F**
Fspike	102.5	0.0239	0.0012	104.9644825	100.1263762	1.409552338	0.957261274
	84.2	0.0107	0.0015	85.07975387	78.1306667	2.255959515	1.00289806
	70.2	0.0263	0.0077	72.55383437	64.3266057	2.820512821	1.008747222
	61	0.0291	0.0091	63.32028273	54.12376663	3.223903321	1.026424728
	54.4	0.0313	0.0103	56.66849702	46.06821972	3.526521775	1.039478256
	48.5	0.0338	0.0114	50.71124528	40.11096799	3.761936487	1.044361833
Blanks							
Fback							
	4.8	0.0096	0.0002	4.838106283			
	6.8	0.0169	0.0062	6.949087172			
	8	0.0213	0.0079	8.227228674			
	8.9	0.0249	0.0089	9.196516095			
	9.3	0.0281	0.1004	10.60027729			
	9.9	0.0306	0.0113	10.31604004			

< 100 kD CAS for Pyrene (June, 2002)							
Controls (Fo spike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Correcte FI (Fo)**		
	100.5	0.0145	0.0016	101.9612292	96.72374695		
	81.8	0.0128	0.0014	82.82612992	77.68777157		
	68.2	0.0117	0.0016	68.99116469	64.05891218		
	59.6	0.0112	0.0016	60.26017347	55.2283056		
	52.5	0.0105	0.0012	53.02106462	47.98502407		
Controls (Fo back)	5.2	0.0072	0.0019	5.23748222			
	5.1	0.0075	0.0019	5.138358341			
	4.9	0.0071	0.0014	4.932252508			
	5	0.007	0.0013	5.031867874			
	5	0.0075	0.0016	5.036040542			
COC Samples						**COC (mg/L)**	**Fo/F**
Fspike	90.3	0.0342	0.0144	94.75020157	87.05358631	1.913795973	1.111082852
	74.2	0.047	0.0221	79.52789242	69.85296285	3.062991077	1.112161437
	63	0.0538	0.027	68.34706005	57.53758047	3.82950383	1.113340388
	55.8	0.0592	0.0317	61.17264562	49.48243643	4.377200495	1.116119366
	50.2	0.0616	0.0317	55.17043155	42.89332343	4.78807561	1.118706135
Blanks (Fback)							
	7.4	0.0268	0.0133	7.696615264			
	9.1	0.0407	0.0206	9.674929569			
	10	0.0504	0.0269	10.80947958			
	10.7	0.0571	0.0305	11.69020919			
	11.2	0.0594	0.0314	12.27710812			

< 30 kD CAS for Pyrene (June, 2002)							
Controls (Fo spike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI (Fo)**		
	100.5	0.0145	0.0016	101.9612292	96.72374695		
	81.8	0.0128	0.0014	82.82612992	77.68777157		
	68.2	0.0117	0.0016	68.99116469	64.05891218		
	59.6	0.0112	0.0016	60.26017347	55.2283056		
	52.5	0.0105	0.0012	53.02106462	47.98502407		
Controls (Fo back)	5.2	0.0072	0.0019	5.23748222			
	5.1	0.0075	0.0019	5.138358341			
	4.9	0.0071	0.0014	4.932252508			
	5	0.007	0.0013	5.031867874			
	5	0.0075	0.0016	5.036040542			
COC Samples						**COC (mg/L)**	**Fo/F**
(Fspike)	98	0.0255	0.0062	101.0449048	93.90689077	1.454484939	1.029996267
	80.6	0.0307	0.0092	83.81329553	75.12624051	2.327873219	1.034096356
	68.4	0.034	0.0112	71.51853768	61.72452409	2.91042291	1.037819459
	60.8	0.0381	0.0134	63.98830709	53.61761613	3.326672376	1.030040304
	54.5	0.0389	0.0137	57.42333968	46.53000529	3.638937464	1.031270548
Blanks (Fback)							
	6.9	0.0248	0.0101	7.138013989			
	8.4	0.0254	0.0092	8.687055026			
	9.4	0.0302	0.0116	9.79401359			
	9.9	0.0334	0.0136	10.37069095			
	10.4	0.0335	0.0134	10.89333439			

< 1.5 um CAS for pyrene (August, 2002)							
Controls (Fo spike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI (Fo)**		
	97.7	0.0267	0.0083	101.0805923	95.30988816		
	80.2	0.0314	0.0121	83.70898934	78.46390443		
	67.4	0.0344	0.0144	70.73628866	65.48032331		
	58.8	0.0383	0.0168	62.11467314	56.76539677		
	51.8	0.0379	0.0167	54.69173296	49.33469135		
Controls (Fo back)							
	5.2	0.1	0.0027	5.770704191			
	5.2	0.009	0.0015	5.245084911			
	5.2	0.0099	0.0026	5.255965351			
	5.3	0.0095	0.0016	5.349276367			
	5.3	0.0099	0.0026	5.357041607			
Samples (Fspike)						**COC (mg/L)**	**Fo/F**
	87.6	0.0267	0.0083	90.63111453	84.17227819	1.364619737	1.132319217
	70	0.0314	0.0121	73.0627089	66.02741543	2.184045812	1.188353412
	58	0.0344	0.0144	60.87099024	53.36530459	2.730602731	1.227020511
	50	0.0383	0.0168	52.81859961	44.85725997	3.121134266	1.265467325
	44.5	0.0379	0.0167	46.98421075	38.79726308	3.414106087	1.271602361
Blanks (Fback)							
	6.3	0.0188	0.0074	6.45883634			
	6.8	0.0247	0.0103	7.035293464			
	7.2	0.0293	0.013	7.50568565			
	7.6	0.0322	0.0148	7.961339641			
	7.8	0.0336	0.0153	8.186947666			

< 0.22 um CAS for pyrene (August, 2002)							
Controls (Fo spike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI (Fo)**		
	97.7	0.0267	0.0083	101.0805923	95.30988816		
	80.2	0.0314	0.0121	83.70898934	78.46390443		
	67.4	0.0344	0.0144	70.73628866	65.48032331		
	58.8	0.0383	0.0168	62.11467314	56.76539677		
	51.8	0.0379	0.0167	54.69173296	49.33469135		
Controls (Fo back)							
	5.2	0.1	0.0027	5.770704191			
	5.2	0.009	0.0015	5.245084911			
	5.2	0.0099	0.0026	5.255965351			
	5.3	0.0095	0.0016	5.349276367			
	5.3	0.0099	0.0026	5.357041607			
COC Samples (Fspike)						**COC (mg/L)**	**Fo/F**
	98	0.0212	0.0043	100.397922	94.29932039	1.327175903	1.010716597
	79.5	0.0218	0.0049	81.54657658	75.02047428	2.124117725	1.045899872
	67.4	0.0221	0.0048	69.1494109	62.30960692	2.655677656	1.050886477
	59.2	0.0227	0.0051	60.79323491	53.64050463	3.035493387	1.058256203
	52.5	0.0223	0.0052	53.89616621	46.10616904	3.320426347	1.070023651
Blanks (Fback)							
	6	0.0137	0.0042	6.098601572			
	6.4	0.0161	0.0049	6.526102301			
	6.7	0.0172	0.0049	6.839803978			
	7	0.0181	0.0049	7.152730283			
	7.6	0.0196	0.0064	7.789997164			

< 100 kD CAS for pyrene (August, 2002)							
Controls (Fo spike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI (Fo)**		
	97.8	0.0188	0.0007	99.57209859	93.82267905		
	78.8	0.017	0.0024	80.21952848	74.35406786		
	66.7	0.0162	0.0026	67.85935957	61.9884268		
	58.1	0.0176	0.0043	59.30003826	53.41326856		
	50.8	0.015	0.0028	51.62947219	45.75732279		
Controls (Fo back)							
	5.7	0.0104	0.0001	5.749419543			
	5.8	0.0107	0.0023	5.865460612			
	5.8	0.0111	0.0028	5.87093277			
	5.8	0.0127	0.0038	5.886769694			
	5.8	0.0115	0.0026	5.872149396			
COC Samples (Fspike)						**COC (mg/L)**	**Fo/F**
	93.2	0.0224	0.0033	95.50024786	88.98022866	0.919454152	1.054421645
	76.1	0.0255	0.007	78.52952768	71.35898923	1.471567452	1.041971988
	64.7	0.0306	0.0105	67.36311	59.86042954	1.83982684	1.035549315
	56.7	0.0326	0.0112	59.19918284	51.32071507	2.102959368	1.040774052
	49.6	0.0294	0.01	51.55070854	43.49918674	2.300358065	1.051912144
Blanks (Fback)							
	6.4	0.0165	0.0036	6.520019199			
	7	0.0193	0.0061	7.170538445			
	7.3	0.0214	0.0072	7.502680464			
	7.6	0.0266	0.0103	7.878467772			
	7.8	0.0245	0.0083	8.051521795			

< 30 kD CAS for pyrene (August, 2002)							
Controls (Fo spike)	**Observed FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI (Fo)**		
	97.8	0.0188	0.0007	99.57209859	93.82267905		
	78.8	0.017	0.0024	80.21952848	74.35406786		
	66.7	0.0162	0.0026	67.85935957	61.9884268		
	58.1	0.0176	0.0043	59.30003826	53.41326856		
	50.8	0.015	0.0028	51.62947219	45.75732279		
Controls (Fo back)							
	5.7	0.0104	0.0001	5.749419543			
	5.8	0.0107	0.0023	5.865460612			
	5.8	0.0111	0.0028	5.87093277			
	5.8	0.0127	0.0038	5.886769694			
	5.8	0.0115	0.0026	5.872149396			
COC Samples (Fspike)						**COC (mg/L)**	**Fo/F**
	98.9	0.0228	0.0018	101.2254844	94.73512153	0.812115161	0.990368488
	80.6	0.0244	0.0055	82.94944262	75.89012264	1.299773605	0.97975949
	67.8	0.0366	0.0116	71.11184588	63.67813142	1.625041625	0.973464915
	59.9	0.027	0.0081	61.97906485	54.25564297	1.857455514	0.984473976
	52.8	0.0271	0.0079	54.62697053	46.79408023	2.031809476	0.977844261
Blanks (Fback)							
	6.4	0.0145	0.0012	6.490362835			
	6.9	0.0185	0.0057	7.059319979			
	7.2	0.0242	0.0088	7.433714457			
	7.5	0.0228	0.0077	7.723421876			
	7.6	0.0234	0.0079	7.832890299			

< 1.5 um MBR for pyrene (August, 2002)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	101.9	0.0176	0.0006	103.6067499	97.65497954		
	83	0.0164	0.0022	84.42518262	78.36123575		
	70	0.0164	0.003	71.26100626	65.08704433		
	61.1	0.015	0.0028	62.09765257	55.92560932		
	53.7	0.0232	0.0096	55.43163765	49.25639634		
Controls (Fo back)							
	5.9	0.0101	0.0005	5.951770343			
	6	0.0105	0.0019	6.063946873			
	6.1	0.0112	0.0026	6.173961927			
	6.1	0.0107	0.0028	6.172043249			
	6.1	0.0117	0.0023	6.17524131			
COC Samples (Fspike)							
						COC (mg/L)	**Fo/F**
	94.1	0.0268	0.007	97.23505363	90.0763922	1.391246464	1.084135112
	76	0.0291	0.0091	78.89084405	70.94419308	2.22666134	1.104547565
	63.9	0.0326	0.0122	66.78569948	58.36189996	2.783882784	1.115231759
	55.8	0.035	0.0135	58.54388639	49.68025366	3.182034447	1.125711026
	49.3	0.0366	0.014	51.83690846	42.7459029	3.480722791	1.152306841
Blanks (Fback)							
	7	0.0185	0.0053	7.158661432			
	7.7	0.0236	0.009	7.946650969			
	8.1	0.0282	0.0118	8.423799521			
	8.5	0.0301	0.0125	8.863632736			
	8.7	0.0318	0.0128	9.091005569			

< 0.22 um MBR for pyrene (August, 2002)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	101.9	0.0176	0.0006	103.6067499	97.65497954		
	83	0.0164	0.0022	84.42518262	78.36123575		
	70	0.0164	0.003	71.26100626	65.08704433		
	61.1	0.015	0.0028	62.09765257	55.92560932		
	53.7	0.0232	0.0096	55.43163765	49.25639634		
Controls (Fo back)							
	5.9	0.0101	0.0005	5.951770343			
	6	0.0105	0.0019	6.063946873			
	6.1	0.0112	0.0026	6.173961927			
	6.1	0.0107	0.0028	6.172043249			
	6.1	0.0117	0.0023	6.17524131			
COC Samples (Fspike)							
						COC (mg/L)	**Fo/F**
	100.8	0.022	0.0019	103.095368	96.19864278	1.199866866	1.015138849
	82	0.024	0.0054	84.34654757	76.888817	1.920362232	1.019149973
	69.2	0.0251	0.0059	71.29834855	63.50673735	2.400932401	1.024884084
	60.8	0.0255	0.0067	62.72156896	54.60475329	2.744314397	1.024189397
	54.4	0.0267	0.0069	56.20075419	47.85859468	3.00191523	1.02920691
Blanks (Fback)							
	6.8	0.0143	0.0015	6.896725198			
	7.3	0.0176	0.0052	7.457730573			
	7.6	0.0202	0.006	7.791611205			
	7.9	0.0215	0.0068	8.11681567			
	8.1	0.0233	0.0073	8.342159513			

< 100 kD MBR for pyrene (August, 2002)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	104.1	0.0182	0.0018	106.0411825	99.98163215		
	84	0.0163	0.0001	85.24780662	79.09937992		
	72.4	0.0166	0.0029	73.71187671	67.54111242		
	62.9	0.0207	0.0067	64.56606011	58.39401695		
	55.3	0.0148	0.0024	56.16802009	49.99533746		
Controls (Fo back)							
	6	0.0102	0.0015	6.059550346			
	6.1	0.0097	0.0001	6.148426697			
	6.1	0.0109	0.0024	6.170764289			
	6.1	0.011	0.0025	6.172043153			
	6.1	0.0112	0.0024	6.172682633			
COC Samples (Fspike)							
						COC (mg/L)	**Fo/F**
	102.1	0.0247	0.0057	105.1306014	98.19589936	0.794641371	1.018185411
	82.1	0.0267	0.0058	84.72106771	77.25638081	1.271807165	1.023855623
	70.9	0.0282	0.0088	73.50541336	65.67901223	1.59007659	1.028351526
	61.8	0.0288	0.009	64.12413384	55.9702322	1.817489771	1.04330489
	54.1	0.0301	0.0098	56.25681948	47.88088456	1.988092264	1.04416069
Blanks (Fback)							
	6.8	0.0163	0.0048	6.934702084			
	7.3	0.0197	0.004	7.464686903			
	7.6	0.0224	0.0081	7.826401127			
	7.9	0.0244	0.0083	8.153901639			
	8.1	0.0257	0.0088	8.375934915			

< 30 kD MBR for pyrene (August, 2002)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	104.1	0.0182	0.0018	106.0411825	99.98163215		
	84	0.0163	0.0001	85.24780662	79.09937992		
	72.4	0.0166	0.0029	73.71187671	67.54111242		
	62.9	0.0207	0.0067	64.56606011	58.39401695		
	55.3	0.0148	0.0024	56.16802009	49.99533746		
Controls (Fo back)							
	6	0.0102	0.0015	6.059550346			
	6.1	0.0097	0.0001	6.148426697			
	6.1	0.0109	0.0024	6.170764289			
	6.1	0.011	0.0025	6.172043153			
	6.1	0.0112	0.0024	6.172682633			
COC Samples (Fspike)							
						COC (mg/L)	**Fo/F**
	103.7	0.0236	0.0035	106.4136316	99.49400181	0.728074555	1.004901103
	84.5	0.0264	0.0042	87.02619917	79.47570229	1.165268345	0.995264938
	72.3	0.0273	0.0078	74.80945283	66.78458025	1.456876457	1.011327947
	63.4	0.0285	0.0081	65.70256729	57.33963853	1.665239319	1.018388299
	56.7	0.029	0.0086	58.82014482	50.24450733	1.821550504	0.995040854
Blanks (Fback)							
	6.8	0.0155	0.0035	6.919629771			
	7.4	0.0184	0.0032	7.550496881			
	7.8	0.0221	0.0075	8.024872571			
	8.1	0.0242	0.0088	8.362928764			
	8.3	0.0251	0.0086	8.575637495			

Appendix C. (Chapter 5)

Sorption of 17β-Estradiol and 17α-Ethinylestradiol by Colloidal Organic Carbon Derived from Biological Wastewater Treatment Systems

< 1.5 um CAS for E2 (May, 2002)							
Controls (Fo)	**Observed FI**	**Abs. At Excitation**	**Abs. At Emission**	**Ideal FI**	**Corrected FI**		
40619	6.89	0.0531	0.0325	7.6	1.4		
40620	5.60	0.0561	0.0344	6.2			
Sample Number	**Observed FI**	**Abs. At Excitation**	**Abs. At Emission**	**Ideal FI**	**Corrected FI**	**COC (mg/L)**	**Fo/F (units)**
4061	6.832	0.0573	0.035	7.6	1.1	0.80	1.218527703
4062	7.122	0.0569	0.0347	7.9	1.5	0.80	0.953540992
4063	7.177	0.0569	0.0347	8.0	1.5	0.80	0.914615639
4064	7.223	0.0611	0.0376	8.1	1.2	1.59	1.163359622
4065	7.385	0.0606	0.0374	8.3	1.4	1.59	1.013987609
4066	7.405	0.061	0.0376	8.3	1.4	1.59	0.993901683
4067	7.518	0.065	0.0405	8.5	1.2	2.39	1.194739984
4068	7.632	0.0648	0.0402	8.6	1.3	2.39	1.080141495
4069	7.572	0.0655	0.0411	8.5	1.2	2.39	1.125727231
40610	7.887	0.0731	0.046	9.0	1.1	3.98	1.256854708
40611	8.032	0.0732	0.0459	9.2	1.3	3.98	1.092654521
40612	8.103	0.0726	0.0457	9.3	1.3	3.98	1.033482188
40613	8.235	0.0765	0.0458	9.5	1.1	4.77	1.264289033
40614	8.328	0.0774	0.049	9.6	1.2	4.77	1.109965113
40615	8.349	0.0766	0.0482	9.6	1.3	4.77	1.104274636
40616	8.424	0.081	0.0515	9.8	1.0	5.57	1.330511806
40617	8.555	0.0804	0.0511	9.9	1.2	5.57	1.171931697
40618	8.538	0.0808	0.0513	9.9	1.2	5.57	1.184947968
Blanks							
406c1a	5.806	0.0571	0.0356	6.4			
406c2a	6.140	0.0621	0.0393	6.9			
406c3a	6.459	0.0673	0.0425	7.3			
406c4a	6.876	0.0764	0.0489	7.9			
406c5a	7.193	0.0811	0.052	8.4			
406c6a	7.453	0.0861	0.0553	8.8			
406c7a	5.654	0.0543	0.034	6.2			
406c8a	6.182	0.0631	0.0399	6.9			
406c9a	6.961	0.0776	0.0497	8.0			

< 1.5 um CAS for EE2 (May, 2002)							
Controls (Fo)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emim.**	**Ideal FI**	**Corrected FI**		
40619	6.67	0.0668	0.0446	7.6	1.1		
40620	5.92	0.0504	0.0285	6.5			
Sample Number	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emim.**	**Ideal FI**	**Corrected FI**	**COC (mg/L)**	**Fo/F (units)**
4061	7.015	0.0526	0.0302	7.7	1.0	0.75	1.0644947
4062	6.978	0.0526	0.0302	7.7	1.0	0.75	1.1082075
4063	7.027	0.052	0.0296	7.7	1.0	0.75	1.0617244
4064	7.297	0.0563	0.0326	8.1	1.0	1.51	1.0985842
4065	7.262	0.0565	0.0327	8.0	1.0	1.51	1.1396993
4066	7.290	0.0567	0.0327	8.1	1.0	1.51	1.1023709
4067	7.505	0.0611	0.0357	8.4	0.9	2.26	1.2725797
4068	7.507	0.0611	0.0358	8.4	0.9	2.26	1.2679568
4069	7.522	0.0612	0.036	8.4	0.9	2.26	1.2396283
40610	8.011	0.0702	0.0417	9.1	0.9	3.76	1.2661834
40611	8.010	0.0696	0.0415	9.1	0.9	3.76	1.2806049
40612	8.029	0.0697	0.0416	9.1	0.9	3.76	1.2458297
40613	8.299	0.074	0.0444	9.5	0.9	4.52	1.2247131
40614	8.270	0.0735	0.044	9.5	0.9	4.52	1.2860617
40615	8.255	0.0743	0.0448	9.5	0.9	4.52	1.2857996
40616	8.537	0.0783	0.0474	9.9	0.7	5.27	1.5121471
40617	8.551	0.0782	0.0473	9.9	0.7	5.27	1.4845833
40618	8.517	0.0782	0.047	9.8	0.7	5.27	1.5744999
Blanks							
406c1a	6.049	0.0548	0.0317	6.670507964	1.102768968		
406c2a	6.346	0.0599	0.0353	7.068596896	1.113922684		
406c3a	6.684	0.0646	0.0383	7.511626291	1.123888144		
406c4a	7.199	0.0734	0.0442	8.229827344	1.14316071		
406c5a	7.453	0.078	0.0475	8.598182584	1.153653909		
406c6a	7.839	0.0826	0.0506	9.124278601	1.163973864		
406c7a	5.969	0.0524	0.0304	6.6	1.1		
406c8a	6.428	0.0608	0.0362	7.2	1.1		
406c9a	7.173	0.0745	0.0455	8.2	1.1		

< 0.22 um CAS for E2 (May, 2003)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
42313a	7.967791667	0.0322	0.0176	8.421408728	2.536485489		
42313b	7.332013333	0.0322	0.0176	7.749434682	1.864511443		
42313c	7.138145	0.0322	0.0176	7.544529164	1.659605925		
42313d	6.996125	0.0322	0.0176	7.394423775	1.509500536		
42313e	6.903845	0.0322	0.0176	7.296890151	1.411966912		
Controls (Fo back)							
42399	5.56793333	0.0322	0.0176	5.884923239			
	5.56793333	0.0322	0.0176	5.884923239			
	5.56793333	0.0322	0.0176	5.884923239			
	5.56793333	0.0322	0.0176	5.884923239			
	5.56793333	0.0322	0.0176	5.884923239			
COC Samples (Fspike)						**COC (mg/L)**	**Fo/F Ratio**
42301a	8.80961	0.03954	0.0225	9.443934684	1.784485978	1.56	1.4214096
42301b	9.085906667	0.04688	0.0274	9.879019573	1.408419876	2.4	1.3238321
42301c	9.28124	0.05422	0.0323	10.23530118	1.0575653	2.925	1.5692704
42301d	9.32226	0.06156	0.0372	10.4271267	0.811556511	3.284210526	1.8600067
42301e	9.493176667	0.0689	0.0421	10.76969947	0.890715736	3.545454545	1.5852049
42302a	9.061001667	0.03954	0.0225	9.713427485	2.053978779	1.56	1.2349132
42302b	9.135228333	0.04688	0.0274	9.932646551	1.462046853	2.4	1.2752748
42302c	9.311028333	0.05422	0.0323	10.2681516	1.090415713	2.925	1.5219938
42302d	9.554706667	0.06156	0.0372	10.68712275	1.071552568	3.284210526	1.4087041
42302e	9.526871667	0.0689	0.0421	10.80792535	0.928941615	3.545454545	1.5199738
Blanks (Fback)							
42303a	7.144983333	0.03954	0.0225	7.659448706			
42303b	7.790558333	0.04688	0.0274	8.470599698			
42303c	8.322253333	0.05422	0.0323	9.177735884			
42303d	8.596696667	0.06156	0.0372	9.615570185			
42303e	8.708036667	0.0689	0.0421	9.878983732			

< 0.22 um CAS for EE2 (May, 2003)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
42313a	7.46	0.0322	0.017	7.878674924	2.536485489		
42313b	7.14	0.0322	0.017	7.541356995	1.864511443		
42313c	6.95	0.0322	0.017	7.341857051	1.659605925		
42313d	6.79	0.0322	0.017	7.174745416	1.509500536		
42313e	6.69	0.0322	0.017	7.070558533	1.411966912		
Controls (Fo back)							
42399	5.72	0.0322	0.017	6.044096107			
COC Samples (Fspike)						**COC (mg/L)**	**Fo/F Ratio**
42301a	8.721	0.0391	0.02188	9.337724583	1.629627612	1.56	1.1257657
42301b	8.836	0.0462	0.02676	9.593142621	1.290801335	2.4	1.1599468
42301c	8.894	0.0533	0.03164	9.790411368	0.919704276	2.925	1.4110633
42301d	8.911	0.0604	0.03652	9.945552586	0.706887545	3.284210526	1.5994755
42301e	9.007	0.0675	0.0414	10.19313426	0.686841721	3.545454545	1.4944672
42302a	8.564	0.0391	0.02188	9.169365723	1.461268752	1.56	1.2554698
42302b	8.720	0.0462	0.02676	9.466442192	1.164100906	2.4	1.2861951
42302c	8.744	0.0533	0.03164	9.625418988	0.754711896	2.925	1.7195448
42302d	8.846	0.0604	0.03652	9.873094562	0.634429521	3.284210526	1.7821512
42302e	8.919	0.0675	0.0414	10.09426574	0.587973196	3.545454545	1.745764
Blanks (Fback)							
42303a	7.199	0.0391	0.02188	7.708096971			
42303b	7.647	0.0462	0.02676	8.302341286			
42303c	8.059	0.0533	0.03164	8.870707092			
42303d	8.277	0.0604	0.03652	9.238665041			
42303e	8.400	0.0675	0.0414	9.506292539			

< 30 kD CAS for E2 (May, 2003)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
42313a	7.97	0.0322	0.0176	8.421408728	2.536485489		
42313b	7.33	0.0322	0.0176	7.749434682	1.864511443		
42313c	7.14	0.0322	0.0176	7.544529164	1.659605925		
42313d	7.00	0.0322	0.0176	7.394423775	1.509500536		
42313e	6.90	0.0322	0.0176	7.296890151	1.411966912		
Controls (Fo back)							
42399	5.56793333	0.0322	0.0176	5.884923239			
Samples (Fspike)						**COC (mg/L)**	**Fo/F Ratio**
42301a	0.039	0.02254	8.860783065	9.337724583	1.979415277	1.05	1.2814317
42301b	0.047	0.02748	8.844364525	9.593142621	1.570530328	1.615384615	1.1871859
42301c	0.054	0.03242	8.856399474	9.790411368	1.152924513	1.96875	1.4394749
42301d	0.061	0.03736	9.10556611	9.945552586	1.112001912	2.210526316	1.3574622
42301e	0.068	0.0423	9.183945638	10.19313426	0.873592377	2.386363636	1.6162766
42302a	0.039	0.02254	8.986930803	9.169365723	2.105563015	1.05	1.204659
42302b	0.047	0.02748	8.885239002	9.466442192	1.611404805	1.615384615	1.157072
42302c	0.054	0.03242	8.803678457	9.625418988	1.100203496	1.96875	1.5084536
42302d	0.061	0.03736	8.975095293	9.873094562	0.981531095	2.210526316	1.5379039
42302e	0.068	0.0423	9.085965421	10.09426574	0.775612159	2.386363636	1.8204548
Blanks (Fback)							
42303a	0.039	0.02254	6.881367789	7.708096971			
42303b	0.047	0.02748	7.273834197	8.302341286			
42303c	0.054	0.03242	7.703474961	8.870707092			
42303d	0.061	0.03736	7.993564198	9.238665041			
42303e	0.068	0.0423	8.310353261	9.506292539			

< 30 kD CAS for EE2 (May, 2003)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
42313a	7.46	0.0322	0.017	7.878674924	1.834578817		
42313b	7.14	0.0322	0.017	7.541356995	1.497260888		
42313c	6.95	0.0322	0.017	7.341857051	1.297760944		
42313d	6.79	0.0322	0.017	7.174745416	1.130649309		
42313e	6.69	0.0322	0.017	7.070558533	1.026462426		
Controls (Fo back)							
42399	5.722503333	0.0322	0.017	6.044096107			
Samples (Fspike)						**COC (mg/L)**	**Fo/F Ratio**
42301a	8.072	0.03846	0.02158	1.069526326	1.559852453	1.05	1.1761233
42301b	7.910	0.04472	0.02556	1.08227099	1.248756736	1.615384615	1.1990013
42301c	7.841	0.05098	0.02954	1.095167105	1.052386227	1.96875	1.2331603
42301d	7.744	0.05724	0.03352	1.108216466	0.809511494	2.210526316	1.3967057
42301e	7.651	0.0635	0.0375	1.12142089	0.701970227	2.386363636	1.4622592
42302a	8.033	0.03846	0.02158	1.069526326	1.517559817	1.05	1.2089005
42302b	7.869	0.04472	0.02556	1.08227099	1.204892293	1.615384615	1.2426512
42302c	7.810	0.05098	0.02954	1.095167105	1.018162255	1.96875	1.2746111
42302d	7.749	0.05724	0.03352	1.108216466	0.815464463	2.210526316	1.3865096
42302e	7.703	0.0635	0.0375	1.12142089	0.760018711	2.386363636	1.3505752
Blanks (Fback)							
42303a	6.614	0.03846	0.02158	1.069526326			
42303b	6.756	0.04472	0.02556	1.08227099			
42303c	6.880	0.05098	0.02954	1.095167105			
42303d	7.013	0.05724	0.03352	1.108216466			
42303e	7.025	0.0635	0.0375	1.12142089			

< 1.5 um MBR for E2 (May, 2002)							
Controls (Fospike and Fback)	**Obse. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corr. FI**		
40919	5.99	0.0505	0.0314	6.6	1.2		
40920	4.90	0.0533	0.0332	5.4			
COC Samples (Fspike)							
						COC (mg/L)	**Fo/F**
40901	6.460	0.0566	0.0351	7.2	1.1	0.48	1.049873161
40902	6.477	0.057	0.0354	7.2	1.1	0.48	1.027936491
40904	7.040	0.0641	0.0403	7.9	1.2	0.96	0.959873725
40905	7.018	0.064	0.0401	7.9	1.2	0.96	0.982530557
40907	7.508	0.0712	0.0454	8.6	1.2	1.45	0.982455472
40908	7.497	0.0742	0.0464	8.6	1.2	1.45	0.960260848
40913	8.837	0.0939	0.061	10.5	1.2	2.89	0.98584757
40914	8.772	0.0929	0.0604	10.5	1.1	2.89	1.073805088
40916	9.174	0.1003	0.0654	11.1	0.9	3.38	1.22770276
40917	9.158	0.1003	0.0655	11.1	0.9	3.38	1.250893061
Blanks (Fback)							
409c1	5.418	0.0604	0.0379	6.1			
409c2	5.906	0.0685	0.0439	6.7			
409c3	6.407	0.076	0.0489	7.4			
409c4	7.296	0.091	0.0559	8.6			
409c5	7.778	0.0982	0.0642	9.4			
409c6	8.297	0.1056	0.0695	10.1			

< 1.5 um MBR for EE2 (May, 2002)							
Controls (Fospike and Foback)	**Obse. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corr. FI**		
40919	6.35	0.0509	0.0284	6.9	0.8		
40920	5.60	0.0533	0.0298	6.2			
COC Samples (Fspike)							
						COC (mg/L)	**Fo/F**
40901	6.914	0.0593	0.034	7.7	0.9	0.48	0.900829769
40902	6.903	0.0593	0.034	7.7	0.9	0.48	0.91335376
40904	7.444	0.0629	0.0369	8.3	0.8	0.96	0.987369564
40905	7.398	0.064	0.0374	8.3	0.8	0.96	1.033565656
40907	7.928	0.0702	0.0418	9.0	0.8	1.45	0.979303879
40908	7.896	0.0702	0.0418	9.0	0.8	1.45	1.026931076
40913	8.891	0.0834	0.0508	10.4	0.7	2.41	1.159628626
40914	8.803	0.0833	0.0507	10.3	0.6	2.41	1.373525125
40916	9.366	0.0897	0.0552	11.1	0.6	2.89	1.216995504
40917	9.343	0.0904	0.0557	11.0	0.6	2.89	1.240761141
40918	9.90429	0.099	0.0619	11.9036	0.7	3.38	1.09445668
40919	9.83595	0.0982	0.0612	11.801	0.6	3.38	1.277350806
Blanks (Fback)							
409c1	9.90429	0.099	0.0619	11.9			
409c2	9.836	0.0982	0.0612	11.8			
409c3	6.123	0.0599	0.0344	6.8			
409c4	6.687	0.0668	0.0391	7.5			
409c5	7.17135	0.0739	0.0443	8.2			
409c6	8.22361	0.0887	0.0546	9.7			
409c7	8.71341	0.0959	0.0595	10.406			
409c8	9.235	0.1033	0.0644	11.187			

< 0.22 um MBR for E2 (May, 2003)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emis.**	**Ideal FI**	**Corrected FI**		
42610a	7.64943	0.0473	0.0282	8.328898573	2.161252545		
42610b	7.187466667	0.0473	0.0282	7.825900866	1.71269614		
42610c	6.9434	0.0473	0.0282	7.560154724	1.5013913		
42610d	6.701186667	0.0473	0.0282	7.29642654	1.292104417		
42610e	6.5332	0.0473	0.0282	7.113518283	1.163637462		
Controls (Fo back)							
42611	5.664491667	0.0473	0.0282	6.167646028			
	5.614491667	0.0473	0.0282	6.113204726			
	5.564491667	0.0473	0.0282	6.058763424			
	5.514491667	0.0473	0.0282	6.004322122			
	5.464491667	0.0473	0.0282	5.94988082			
COC Samples (Fspike)						**COC (mg/L)**	**Fo/F**
42601a	8.078088333	0.05344	0.03236	8.901056168	1.889529142	2.892	1.143804823
42601b	8.337391667	0.05958	0.03652	9.296884335	1.59253377	3.484615385	1.075453577
42601c	8.122036667	0.06572	0.04068	9.165291559	1.102004164	3.855	1.362418899
42601d	8.132291667	0.07186	0.04484	9.286845829	1.092347189	4.108421053	1.182869723
42601e	8.072226667	0.078	0.049	9.328727847	0.844146038	4.292727273	1.378478853
42602a	8.08639	0.05344	0.03236	8.91020358	1.898676553	2.892	1.138294219
42602b	8.1738	0.05958	0.03652	9.114466036	1.410115472	3.484615385	1.214578646
42602c	8.099575	0.06572	0.04068	9.139944749	1.076657354	3.855	1.394493146
42602d	8.114711667	0.07186	0.04484	9.266769969	1.072271329	4.108421053	1.205016288
42602e	8.008761667	0.078	0.049	9.255384055	0.770802246	4.292727273	1.509644617
Blanks (Fback)							
42603a	6.36326	0.05344	0.03236	7.011527027			
42603b	6.909216667	0.05958	0.03652	7.704350564			
42603c	7.14547	0.06572	0.04068	8.063287395			
42603d	7.175746667	0.07186	0.04484	8.19449864			
42603e	7.34178	0.078	0.049	8.484581809			

< 0.22 um MBR for EE2 (May, 2003)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Correct. FI**		
42621a	7.86039	0.0425	0.0218	8.448424338	2.12122495		
42621b	7.477536667	0.0425	0.0218	8.036929816	1.709730429		
42621c	7.219696667	0.0425	0.0218	7.759800853	1.432601466		
42621d	7.068828333	0.0425	0.0218	7.597646087	1.2704467		
42621e	6.94047	0.0425	0.0218	7.459685291	1.132485903		
Controls (Fo back)							
42622	5.886808333	0.0425	0.0218	6.327199388			
Samples						**COC (mg/L)**	**Fo/F**
42612a	8.37304	0.04864	0.02572	9.104766708	1.460621388	1.011	1.4522757
42612b	8.395991667	0.05478	0.02964	9.236587059	1.133433915	1.555384615	1.5084518
42612c	8.343251667	0.06092	0.03356	9.28599802	0.959188431	1.895625	1.4935558
42612d	8.345205	0.06706	0.03748	9.396882729	0.821948994	2.128421053	1.5456515
42612e	8.334461667	0.0732	0.0414	9.494623466	0.69972357	2.297727273	1.6184761
42613a	8.39941	0.04864	0.02572	9.133441204	1.489295884	1.011	1.424314
42613b	8.423338333	0.05478	0.02964	9.266671637	1.163518493	1.555384615	1.4694484
42613c	8.377923333	0.06092	0.03356	9.324587414	0.997777825	1.895625	1.435792
42613d	8.34374	0.06706	0.03748	9.395233107	0.820299372	2.128421053	1.5487598
42613e	8.350576667	0.0732	0.0414	9.512981683	0.718081787	2.297727273	1.5770988
Blanks							
42614a	7.029805	0.04864	0.02572	7.64414532			
42614b	7.365708333	0.05478	0.02964	8.103153144			
42614c	7.481443333	0.06092	0.03356	8.326809589			
42614d	7.615246667	0.06706	0.03748	8.574933735			
42614e	7.720238333	0.0732	0.0414	8.794899896			

< 30 kD MBR for E2 (May, 2003)							
Controls (Fo spike)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emis.**	**Ideal FI**	**Corrected FI**		
42610a	7.64943	0.0473	0.0282	8.328898573	2.161252545		
42610b	7.187466667	0.0473	0.0282	7.825900866	1.71269614		
42610c	6.9434	0.0473	0.0282	7.560154724	1.5013913		
42610d	6.701186667	0.0473	0.0282	7.29642654	1.292104417		
42610e	6.5332	0.0473	0.0282	7.113518283	1.163637462		
Samples (Fspike)						**COC (mg/L)**	**Fo/F**
42604a	7.86918	0.05344	0.03236	8.670864973	1.777716218	0.849	1.215746655
42604b	7.770536667	0.05958	0.03652	8.664793918	1.500074799	1.306153846	1.141740493
42604c	7.625501667	0.06572	0.04068	8.604977905	1.25078996	1.591875	1.200354454
42604d	7.455561667	0.07186	0.04484	8.514039413	1.018735702	1.787368421	1.268341155
42604e	7.440423333	0.078	0.049	8.598579699	0.878571144	1.929545455	1.324465834
42605a	8.037563333	0.05344	0.03236	8.856402621	1.963253866	0.849	1.100852306
42605b	7.753933333	0.05958	0.03652	8.646279822	1.481560703	1.306153846	1.156008078
42605c	7.625501667	0.06572	0.04068	8.604977905	1.25078996	1.591875	1.200354454
4205d	7.347151667	0.07186	0.04484	8.390238276	0.894934565	1.787368421	1.443797645
42605e	7.5	0.078	0.049	8.66742991	0.947421355	1.929545455	1.228215362
Blanks (Fback)							
42606a	6.255826667	0.05344	0.03236	6.893148755			
42606b	6.425278333	0.05958	0.03652	7.16471912			
42606c	6.517085	0.06572	0.04068	7.354187945			
42606d	6.563476667	0.07186	0.04484	7.495303711			
42606e	6.680188333	0.078	0.049	7.720008555			

< 30 kD MBR for EE2 (May, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Correct. FI**		
42621a	7.86039	0.0425	0.0218	8.448424338	2.12122495		
42621b	7.477536667	0.0425	0.0218	8.036929816	1.709730429		
42621c	7.219696667	0.0425	0.0218	7.759800853	1.432601466		
42621d	7.068828333	0.0425	0.0218	7.597646087	1.2704467		
42621e	6.94047	0.0425	0.0218	7.459685291	1.132485903		
Controls (Fo without Probe)							
42622	5.886808333	0.0425	0.0218	6.327199388			
Samples						**COC (mg/L)**	**Fo/F**
42615a	8.08883	0.04836	0.02564	8.792057331	1.49925898	0.849	1.4148489
42615b	7.95316	0.05422	0.02948	8.742135919	1.185616652	1.306153846	1.4420601
42615c	7.80472	0.06008	0.03332	8.675770324	0.957448121	1.591875	1.4962706
42615d	7.65529	0.06594	0.03716	8.605678237	0.755258772	1.787368421	1.6821343
42615e	7.669938333	0.0718	0.041	8.719426453	0.702226417	1.929545455	1.6127076
42616a	8.09518	0.04836	0.02564	8.798959388	1.506161036	0.849	1.4083653
42616b	7.912153333	0.05422	0.02948	8.697061275	1.140542009	1.306153846	1.4990508
42616c	7.802766667	0.06008	0.03332	8.673598988	0.955276784	1.591875	1.4996716
42616d	7.729028333	0.06594	0.03716	8.68857103	0.838151565	1.787368421	1.515772
42616e	7.702658333	0.0718	0.041	8.756623575	0.73942354	1.929545455	1.5315795
Blanks							
42617a	6.709488333	0.04836	0.02564	7.292798351			
42617b	6.874545	0.05422	0.02948	7.556519267			
42617c	6.9434	0.06008	0.03332	7.718322204			
42617d	6.98344	0.06594	0.03716	7.850419465			
42617e	7.052233333	0.0718	0.041	8.017200036			

< 1.5 um CAS for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	98.8	0.0407	0.0076	103.6367704	66.28463676		
	90.1	0.0393	0.0078	94.3934518	55.55011413		
	83.2	0.0384	0.0082	87.11952339	47.75957588		
	46.9	0.0371	0.0081	49.03827052	27.17616312		
	44.6	0.0373	0.0092	46.69626171	24.82282594		
Controls (Fo without Probe)	37	0.00331	0.008	37.35213367			
	37.3	0.0328	0.0085	38.84333768			
	37.8	0.0328	0.0084	39.35994752			
	21	0.0326	0.0084	21.86210741			
	21	0.0327	0.0088	21.87343577			
						COC (mg/L)	**Fo/F**
Samples	106.3	0.0592	0.0167	114.738015	61.37676983	1.364619737	1.0799629
	103.2	0.0683	0.0221	113.0776755	49.73125467	2.184045812	1.1170061
	100.7	0.0756	0.0262	111.6490214	41.16646237	2.730602731	1.1601574
	58.1	0.0785	0.0278	64.71811299	22.9016803	3.121134266	1.1866449
	57.4	0.0828	0.03	64.37029093	19.9263427	3.414106087	1.2457291
Blanks							
	49.9	0.0504	0.0165	53.36124512			
	58.3	0.061	0.0213	63.34642087			
	64	0.0696	0.0257	70.48255902			
	37.7	0.074	0.0282	41.81643268			
	39.8	0.0782	0.0305	44.44394823			

< 0.22 um CAS for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	98.8	0.0407	0.0076	103.6367704	66.28463676		
	90.1	0.0393	0.0078	94.3934518	55.55011413		
	83.2	0.0384	0.0082	87.11952339	47.75957588		
	46.9	0.0371	0.0081	49.03827052	27.17616312		
	44.6	0.0373	0.0092	46.69626171	24.82282594		
Controls (Fo without Probe)	37	0.00331	0.008	37.35213367			
	37.3	0.0328	0.0085	38.84333768			
	37.8	0.0328	0.0084	39.35994752			
	21	0.0326	0.0084	21.86210741			
	21	0.0327	0.0088	21.87343577			
						COC (mg/L)	**Fo/F**
Samples	104.8	0.0485	0.0093	111.0176955	68.30901684	1.327175903	0.9703644
	94.2	0.0524	0.0132	100.5984208	56.33595425	2.124117725	0.9860508
	86.7	0.055	0.0147	92.98307713	47.05560566	2.655677656	1.0149604
	47	0.0562	0.0156	50.51582532	24.63753227	3.035493387	1.1030392
	44.4	0.0568	0.0164	47.79059482	21.66148657	3.320426347	1.1459429
Blanks							
	40.3	0.0468	0.0114	42.70867867			
	41.4	0.0519	0.0148	44.26246657			
	42.7	0.0553	0.0172	45.92747147			
	24	0.0568	0.0181	25.87829305			
	24.2	0.0577	0.0185	26.12910826			

< 100 kD CAS for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	73	0.0429	0.007	76.70073831	51.52292424		
	64.1	0.0396	0.0076	67.16145216	42.08767367		
	58.2	0.0375	0.0077	60.85345695	35.71473348		
	53	0.0365	0.0081	55.3819487	30.1498046		
	50.1	0.0364	0.0089	52.38959976	26.80441151		
Controls (Fo without Probe)							
	24.2	0.0338	0.0066	25.17781407			
	24.1	0.0326	0.0078	25.07377849			
	24.2	0.0314	0.0075	25.13872347			
	24.3	0.0311	0.0074	25.23214411			
	24.5	0.0344	0.0096	25.58518825			
Samples						**COC (mg/L)**	**Fo/F**
	77.28	0.0619	0.0202	83.9519041	54.65012089		
	66.8	0.0576	0.0159	71.92350182	40.79535059	1.471567452	1.031678195
	61.6	0.0604	0.0179	66.65527492	33.95410185	1.83982684	1.051853282
	57.6	0.0622	0.0194	62.54043723	28.78383526	2.102959368	1.047456127
	55.7	0.0633	0.0201	60.59034089	24.47917892	2.300358065	1.094988177
Blanks							
	27.552	0.0473	0.0143	29.30178322			
	29.1	0.051	0.0162	31.12815122			
	30.4	0.0544	0.0182	32.70117307			
	31.3	0.0567	0.0184	33.75660197			
	33.2	0.0618	0.0215	36.11116197			

< 30 kD CAS for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	73	0.0429	0.007	76.70073831	51.52292424		
	64.1	0.0396	0.0076	67.16145216	42.08767367		
	58.2	0.0375	0.0077	60.85345695	35.71473348		
	53	0.0365	0.0081	55.3819487	30.1498046		
	50.1	0.0364	0.0089	52.38959976	26.80441151		
Controls (Fo without Probe)							
	24.2	0.0338	0.0066	25.17781407			
	24.1	0.0326	0.0078	25.07377849			
	24.2	0.0314	0.0075	25.13872347			
	24.3	0.0311	0.0074	25.23214411			
	24.5	0.0344	0.0096	25.58518825			
Samples						**COC (mg/L)**	**Fo/F**
	70.6	0.0518	0.0105	75.13808744	47.26722585	0.812115161	1.090034867
	63.3	0.0551	0.0151	67.92247385	38.22291579	1.299773605	1.101111017
	58.7	0.0568	0.0161	63.16297548	32.1910645	1.625041625	1.109461089
	55	0.0589	0.0178	59.41508921	27.29505266	1.857455514	1.104588622
Blanks	52.5	0.0614	0.0191	56.93808883	24.01700215	2.031809476	1.116059837
	26.4	0.044	0.0105	27.87086159			
	27.9	0.0484	0.0141	29.69955806			
	28.9	0.052	0.017	30.97191099			
	29.9	0.0541	0.0172	32.12003654			
	30.5	0.0569	0.019	32.92108669			

< 1.5 um CAS for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	124.4	0.0453	0.0058	134.734438	97.38230429		
	144.816	0.0433	0.0073	159.7487546	120.9054169		
	84.336	0.0422	0.0083	94.07890616	54.71895864		
	64.176	0.0405	0.0082	71.89459069	50.03248329		
	64.848	0.04	0.0087	73.04731902	51.17388326		
Controls (Fo without Probe)							
	37	0.00331	0.008	37.35213367			
	37.3	0.0328	0.0085	38.84333768			
	37.8	0.0328	0.0084	39.35994752			
	21	0.0326	0.0084	21.86210741			
	21	0.0327	0.0088	21.87343577		**COC (mg/L)**	**Fo/F**
Samples							
	134	0.0646	0.0146	140.9682185	87.60697342	1.364619737	1.111581653
	164.976	0.0745	0.0224	173.4652156	110.1187947	2.184045812	1.097954415
	108.836	0.0809	0.0268	114.4245758	43.94201679	2.730602731	1.245253692
	79.968	0.0834	0.0284	83.91762229	42.1011896	3.121134266	1.188386451
	77.064	0.0863	0.0308	80.87020181	36.42625358	3.414106087	1.404862654
Blanks							
	49.9	0.0504	0.0165	53.36124512			
	58.3	0.061	0.0213	63.34642087			
	64	0.0696	0.0257	70.48255902			
	37.7	0.074	0.0282	41.81643268			
	39.8	0.0782	0.0305	44.44394823			

< 0.22 um CAS for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	124.4	0.0453	0.0058	134.734438	97.38230429		
	144.816	0.0433	0.0073	159.7487546	120.9054169		
	84.336	0.0422	0.0083	94.07890616	54.71895864		
	64.176	0.0405	0.0082	71.89459069	50.03248329		
	64.848	0.04	0.0087	73.04731902	51.17388326		
Controls (Fo without Probe)							
	37	0.00331	0.008	37.35213367			
	37.3	0.0328	0.0085	38.84333768			
	37.8	0.0328	0.0084	39.35994752			
	21	0.0326	0.0084	21.86210741			
	21	0.0327	0.0088	21.87343577		**COC (mg/L)**	**Fo/F**
Samples							
	124.4	0.0527	0.0089	132.3002694	89.59159075	1.327175903	1.086958089
	139.518	0.0571	0.0139	149.8303401	105.5678735	2.124117725	1.145286088
	86.688	0.0575	0.015	93.24024358	47.31277211	2.655677656	1.156536728
	63.168	0.059	0.0161	68.12574334	42.24745029	3.035493387	1.184272256
	58.464	0.0587	0.017	63.09176262	36.96265437	3.320426347	1.384475334
Blanks							
	40.3	0.0468	0.0114	42.70867867			
	41.4	0.0519	0.0148	44.26246657			
	42.7	0.0553	0.0172	45.92747147			
	24	0.0568	0.0181	25.87829305			
	24.2	0.0577	0.0185	26.12910826			

< 100 kD CAS for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	98.6	0.0463	0.0067	103.9	78.753923		
	85.9	0.042	0.0076	90.2	65.1528941		
	76.3	0.0401	0.0084	80.1	54.91315245		
	70.4	0.0382	0.0073	73.6	48.40040982		
	65.1	0.0368	0.0075	68.0	42.41941865		
Controls (Fo without Probe)							
	24.20	0.0338	0.0066	25.2			
	24.10	0.0326	0.0078	25.1			
	24.20	0.0314	0.0075	25.1			
	24.30	0.0311	0.0074	25.2			
	24.50	0.0344	0.0096	25.6		**COC (mg/L)**	**Fo/F**
Samples							
	98.3	0.0581	0.012	105.4673296	75.99113101	0.919454152	0.964918675
	87.4	0.059	0.0148	94.13272163	63.00457041	1.471567452	0.967026427
	80.2	0.062	0.017	86.84462676	54.14345369	1.83982684	0.985983344
	77.1	0.0641	0.0184	83.79102989	50.03442792	2.102959368	1.033760419
	72.3	0.0679	0.0212	79.1135006	43.00233862	2.300358065	1.013741819
Blanks							
	27.716	0.0473	0.0143	29.47619859			
	29.1	0.051	0.0162	31.12815122			
	30.4	0.0544	0.0182	32.70117307			
	31.3	0.0567	0.0184	33.75660197			
	33.2	0.0618	0.0215	36.11116197			

< 30 kD CAS for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**FI (Fo)**		
	98.6	0.0463	0.0067	103.9	78.753923		
	85.9	0.042	0.0076	90.2	65.1528941		
	76.3	0.0401	0.0084	80.1	54.91315245		
	70.4	0.0382	0.0073	73.6	48.40040982		
	65.1	0.0368	0.0075	68.0	42.41941865		
Controls (Fo without Probe)							
	24.20	0.0338	0.0066	25.2			
	24.10	0.0326	0.0078	25.1			
	24.20	0.0314	0.0075	25.1			
	24.30	0.0311	0.0074	25.2			
	24.50	0.0344	0.0096	25.6		**COC (mg/L)**	**Fo/F**
Samples							
	99.3	0.0559	0.0103	106.1106756	78.23981397	0.812115161	1.006570939
	87.9	0.0581	0.0143	94.53404117	64.83448311	1.299773605	1.004911136
	79.2	0.06	0.0164	85.5311145	54.55920351	1.625041625	1.006487429
	73.9	0.0616	0.018	80.07243637	47.95239983	1.857455514	1.009342806
	69.2	0.0619	0.0183	75.02649065	42.10540396	2.031809476	1.007457824
Blanks							
	26.4	0.044	0.0105	27.87086159			
	27.9	0.0484	0.0141	29.69955806			
	28.9	0.052	0.017	30.97191099			
	29.9	0.0541	0.0172	32.12003654			
	30.5	0.0569	0.019	32.92108669			

< 1.5 um MBR for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	53.4	0.0453	0.0073	56.2642641	30.87626797		
	48.6	0.0418	0.0078	51.04792176	25.09884314		
	44.8	0.0397	0.0077	46.9494003	20.90960038		
	42.8	0.0412	0.009	44.9837448	18.94579955		
	39.5	0.0427	0.0105	41.6446023	15.62241477		
Controls (Fo without Probe)							
	24.6	0.0257	0.0069	25.38799613			
	24.9	0.0342	0.0078	25.94907861			
	25	0.0338	0.0077	26.03979993			
	24.9	0.0363	0.009	26.03794524			
	24.8	0.0383	0.0103	26.02218753			
Samples						**COC (mg/L)**	**Fo/F**
	54.6	0.0592	0.0167	58.9341074	28.14282631	1.391246464	1.097127475
	52	0.0683	0.0221	56.97712333	22.29355839	2.22666134	1.125833871
	50	0.0756	0.0262	55.43645551	18.22073858	2.783882784	1.147571504
	48.1	0.0785	0.0278	53.57902297	14.7149868	3.182034447	1.287517265
	47.1	0.0828	0.03	52.81952444	12.88045084	3.480722791	1.212877946
Blanks							
	28.8	0.0519	0.0148	30.79128109			
	32	0.0602	0.0197	34.68356495			
	34	0.0664	0.023	37.21571692			
	35.1	0.0738	0.0267	38.86403617			
	36	0.075	0.0274	39.9390736			

< 0.22 um MBR for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	53.4	0.0453	0.0073	56.2642641	30.87626797		
	48.6	0.0418	0.0078	51.04792176	25.09884314		
	44.8	0.0397	0.0077	46.9494003	20.90960038		
	42.8	0.0412	0.009	44.9837448	18.94579955		
	39.5	0.0427	0.0105	41.6446023	15.62241477		
Controls (Fo without Probe)							
	24.6	0.0257	0.0069	25.38799613			
	24.9	0.0342	0.0078	25.94907861			
	25	0.0338	0.0077	26.03979993			
	24.9	0.0363	0.009	26.03794524			
	24.8	0.0383	0.0103	26.02218753			
Samples						**COC (mg/L)**	**Fo/F**
	68	0.0667	0.0131	73.69479649	44.99739126	1.199866866	0.686179067
	58.5	0.0672	0.0169	63.68226876	33.1990483	1.920362232	0.756010923
	52.6	0.0679	0.019	57.42594763	26.265666	2.400932401	0.796081104
	48.1	0.07	0.0208	52.72565744	21.41978763	2.744314397	0.884499878
	45.3	0.0717	0.0223	49.82126005	17.8562032	3.00191523	0.874901265
Blanks							
	26.9	0.0519	0.0127	28.69740524			
	28.3	0.057	0.0169	30.48322046			
	28.8	0.0598	0.0184	31.16028163			
	28.8	0.0629	0.0198	31.30586981			
	29.3	0.0648	0.0214	31.96505685			

< 100 kD MBR for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	47.3	0.045	0.0057	49.7	27.43052413		
	43.1	0.0421	0.0078	45.3	22.52409164		
	39.5	0.0402	0.0075	41.4	18.63531638		
	37.1	0.046	0.012	39.3	16.49174188		
	35.4	0.0382	0.0075	37.0	14.01395132		
Controls (Fo without Probe)							
	21.40	0.0373	0.005	22.3			
	21.80	0.036	0.0078	22.8			
	21.80	0.0361	0.0082	22.8			
	21.80	0.0371	0.0091	22.8			
	22.10	0.034	0.0075	23.0			
Samples						**COC (mg/L)**	**Fo/F**
	46.1	0.0558	0.0123	49.35892691	44.99739126	24.44662078	0.794641371
	43.9	0.061	0.0173	47.50269623	33.1990483	20.93573882	1.271807165
	41.3	0.063	0.0189	44.85629192	26.265666	17.70940498	1.59007659
	39.6	0.0655	0.0206	43.19743573	21.41978763	14.93208634	1.817489771
	38.3	0.0677	0.0217	41.92239503	17.8562032	12.99895058	1.988092264
Blanks							
	23.5	0.0469	0.0116	24.91230614			
	24.8	0.0524	0.0162	26.56695741			
	25.2	0.0562	0.0178	27.14688694			
	26.1	0.0597	0.0194	28.26534938			
	26.6	0.062	0.021	28.92344446			

< 30 kD MBR for E2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	47.3	0.045	0.0057	49.7	27.43052413		
	43.1	0.0421	0.0078	45.3	22.52409164		
	39.5	0.0402	0.0075	41.4	18.63531638		
	37.1	0.046	0.012	39.3	16.49174188		
	35.4	0.0382	0.0075	37.0	14.01395132		
Controls (Fo without Probe)							
	21.40	0.0373	0.005	22.3			
	21.80	0.036	0.0078	22.8			
	21.80	0.0361	0.0082	22.8			
	21.80	0.0371	0.0091	22.8			
	22.10	0.034	0.0075	23.0			
Samples						**COC (mg/L)**	**Fo/F**
	46	0.0539	0.0094	49.00754987	23.9585139	0.728074555	1.144917596
	42.9	0.0579	0.0155	46.18560356	19.98620165	1.165268345	1.126982107
	40.2	0.0619	0.0178	43.56218113	16.38434317	1.456876457	1.137385624
	38.6	0.0642	0.0189	41.97593818	14.07307998	1.665239319	1.171864432
	37.7	0.0644	0.0196	41.03546739	12.64418209	1.821550504	1.108331976
Blanks							
	23.4	0.0525	0.0154	25.04903598			
	24.5	0.0516	0.0153	26.19940191			
	25.2	0.0569	0.0182	27.17783796			
	25.8	0.0591	0.0187	27.9028582			
	26.2	0.0602	0.0195	28.3912853			

< 1.5 um MBR for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	69.2	0.0471	0.0062	73.0	47.57661482		
	62.1	0.0442	0.0077	65.4	39.43441372		
	56.2	0.0422	0.0082	59.1	33.03986955		
	51.4	0.0416	0.0085	54.0	27.97898788		
	47.9	0.0407	0.0085	50.3	24.26964025		
Controls (Fo without Probe)							
	24.60	0.0257	0.0069	25.4			
	24.90	0.0342	0.0078	25.9			
	25.00	0.0338	0.0077	26.0			
	24.90	0.0363	0.009	26.0			
	24.80	0.0383	0.0103	26.0			
Samples						**COC (mg/L)**	**Fo/F**
	69.9	0.0624	0.0136	75.45635728	44.66507619	1.391246464	1.065186022
	65.3	0.0697	0.0196	71.46859321	36.78502826	2.22666134	1.072023472
	61.4	0.0749	0.0232	67.81553178	30.59981486	2.783882784	1.079740832
	58.3	0.0778	0.0253	64.72597525	25.86193907	3.182034447	1.081859632
	56.2	0.0814	0.0277	62.7834817	22.8444081	3.480722791	1.092388666
Blanks							
	28.8	0.0519	0.0148	30.79128109			
	32	0.0602	0.0197	34.68356495			
	34	0.0664	0.023	37.21571692			
	35.1	0.0738	0.0267	38.86403617			
	36	0.075	0.0274	39.9390736			

< 0.22 um MBR for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	69.2	0.0471	0.0062	73.0	47.57661482		
	62.1	0.0442	0.0077	65.4	39.43441372		
	56.2	0.0422	0.0082	59.1	33.03986955		
	51.4	0.0416	0.0085	54.0	27.97898788		
	47.9	0.0407	0.0085	50.3	24.26964025		
Controls (Fo without Probe)							
	24.60	0.0257	0.0069	25.4			
	24.90	0.0342	0.0078	25.9			
	25.00	0.0338	0.0077	26.0			
	24.90	0.0363	0.009	26.0			
	24.80	0.0383	0.0103	26.0			
Samples						**COC (mg/L)**	**Fo/F**
	69.5	0.057	0.0103	74.35145368	45.65404844	1.199866866	1.04211163
	63.2	0.0606	0.0141	68.13198227	37.64876181	1.920362232	1.047429233
	58	0.0618	0.0167	62.77281814	31.61253651	2.400932401	1.045150855
	53.8	0.0651	0.0185	58.53562266	27.22975286	2.744314397	1.027515308
	50.6	0.0679	0.0189	55.23672846	23.27167162	3.00191523	1.04288341
Blanks							
	26.9	0.0519	0.0127	28.69740524			
	28.3	0.057	0.0169	30.48322046			
	28.8	0.0598	0.0184	31.16028163			
	28.8	0.0629	0.0198	31.30586981			
	29.3	0.0648	0.0214	31.96505685			

< 100 kD MBR for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	60.4	0.0463	0.0036	63.5	41.1533887		
	54.8	0.0441	0.0077	57.7	34.93066076		
	49.4	0.0427	0.0079	51.9	29.1693295		
	45.8	0.0406	0.0073	48.0	25.2047034		
	42.9	0.0395	0.0077	44.9	21.92974675		
Controls (Fo without Probe)							
	21.40	0.0373	0.005	22.3			
	21.80	0.036	0.0078	22.8			
	21.8	0.0361	0.0082	22.77266696			
	21.8	0.0371	0.0091	22.81753746			
	22.1	0.034	0.0075	23.01918236			
Samples						**COC (mg/L)**	**Fo/F**
	58.3	0.0567	0.0091	62.27278565	37.36047951	0.794641371	1.101521962
	53.5	0.0608	0.016	57.80063583	31.23367843	1.271807165	1.118365256
	49.1	0.0643	0.018	53.35002498	26.20313805	1.59007659	1.113199856
	46.3	0.066	0.0194	50.46947042	22.20412104	1.817489771	1.135136282
	44.1	0.0667	0.0202	48.14610671	19.22266225	1.988092264	1.140827762
Blanks							
	23.5	0.0469	0.0116	24.91230614			
	24.8	0.0524	0.0162	26.56695741			
	25.2	0.0562	0.0178	27.14688694			
	26.1	0.0597	0.0194	28.26534938			
	26.6	0.062	0.021	28.92344446			

< 30 kD MBR for EE2 (August, 2002)							
Controls (Fo with Probe)	**Obs. FI**	**Abs. At Excit.**	**Abs. At Emiss.**	**Ideal FI**	**Corrected FI**		
	60.4	0.0463	0.0036	63.5	41.1533887		
	54.8	0.0441	0.0077	57.7	34.93066076		
	49.4	0.0427	0.0079	51.9	29.1693295		
	45.8	0.0406	0.0073	48.0	25.2047034		
	42.9	0.0395	0.0077	44.9	21.92974675		
Controls (Fo without Probe)							
	21.40	0.0373	0.005	22.3			
	21.80	0.036	0.0078	22.8			
	21.8	0.0361	0.0082	22.77266696			
	21.8	0.0371	0.0091	22.81753746			
	22.1	0.034	0.0075	23.01918236			
Samples						**COC (mg/L)**	**Fo/F**
	58.8	0.0544	0.0084	62.61197955	37.56294357	0.728074555	1.095584765
	53.5	0.0604	0.0153	57.73480631	31.5354044	1.165268345	1.107664906
	49.1	0.0631	0.0173	53.24512764	26.06728969	1.456876457	1.119001241
	46.1	0.0641	0.0183	50.09554213	22.19268393	1.665239319	1.13572128
	44	0.0652	0.0194	47.92262271	19.53133742	1.821550504	1.122798008
Blanks							
	23.4	0.0525	0.0154	25.04903598			
	24.5	0.0516	0.0153	26.19940191			
	25.2	0.0569	0.0182	27.17783796			
	25.8	0.0591	0.0187	27.9028582			
	26.2	0.0602	0.0195	28.3912853			

Organic Carbon and Molar Extinction Coefficient Calculations

MBR (August, 2002)						
Size Fraction	Whole	1.5 um	0.22 um	100 kD	30 kD	1 kD
TOC (mg/L)	8.38	7.82	7.05	5.43	5.16	2.24
COC (mg/L)	6.14	5.58	4.81	3.19	2.92	2.24
Abs 280	0.1332	0.1028	0.0771	0.0831	0.08	0.0362
e280	260.3257	221.0753	192.3493	312.6019	328.7671	193.9286
CAS (August, 2002)						
Size Fraction	Whole	1.5 um	0.22 um	100 kD	30 kD	1 kD
TOC (mg/L)	10.9	7.69	7.54	5.9	5.47	2.4
COC (mg/L)	8.66	5.45	5.3	3.66	3.23	2.4
Abs 280	0.2107	0.1111	0.0652	0.0777	0.0742	0.0309
e280	291.963	244.6239	147.6226	254.7541	275.6656	154.5

Appendix D. (Chapter 6)

Evaluation of a Recombinant Yeast Bioassay to Quantify the Bioavailability of Colloid-Associated 17β-Estradiol

Key to Plates Containing COC and 17β-Estradiol

Plate Number	First COC Dilution – Replicate 1
	First COC Dilution – Replicate 2
	Blank
	Second COC Dilution – Replicate 1
	Second COC Dilution – Replicate 2
	Blank
	Third COC Dilution – Replicate 1
	Blank

Key to Plates Containing only COC (Blanks or negative controls)

Plate Number	First COC Dilution
	Blank
	Second COC Dilution
	Blank
	Third COC Dilution
	Blank
	Blank
	Blank

Blank = Left row empty intentionally.

17β-Estradiol Concentration in serially diluted microtiter cells for all COC size fractions

First COC Dilution E2 Concentration (pM)	4762	2381	1190	595	298	149	74	37	19	9	5	2
Second COC Dilution E2 Concentration (pM)	3175	1587	794	397	198	99	50	25	12	6	3	2
Third COC Dilution E2 Concentration (pM)	2381	1190	595	298	149	74	37	19	9	5	2	1

Samples from 1/29/03

Plate Number	Description
1	Whole Sample - E2 Spiked
2	Whole Sample - E2 Spiked
3	Whole Sample - Blank
4	< 1.5 um Sample - E2 Spiked
5	< 1.5 um Sample - E2 Spiked
6	< 1.5 um Sample - Blank
7	< 0.22 um Sample - E2 Spiked
8	< 0.22 um Sample - E2 Spiked
9	< 0.22 um Sample - Blank
10	100 kD Sample - E2 Spiked
11	100 kD Sample - E2 Spiked
12	100 kD Sample - Blank
13	30 kD Sample - E2 Spiked
14	30 kD Sample - E2 Spiked
15	30 kD Sample - Blank
16	10 kD Sample - E2 Spiked
17	10 kD Sample - E2 Spiked
18	10 kD Sample - Blank
19	1 kD Sample - E2 Spiked
20	1 kD Sample - E2 Spiked
21	1 kD Sample - Blank

Samples from 1/29/03 – Summary of E2 Dose-Response and Organic Carbon Parameters

Exper	COC	Repl	Min	Max	EC50	EC50 err	Slope	Slope err	Protein	Polys	Humics	Fulvics
<0.22	1.496667	1	1.08237	2.5114	102.9758	5.51587	2.28645	0.24135	1.52	0.4	0.08	1.24
<0.22	1.496667	1	1.06413	2.53436	89.36354	3.53423	2.1153	0.15483	1.52	0.4	0.08	1.24
<0.22	2.245	2	1.10482	2.59939	80.17716	3.78074	3.56792	0.49199	2.02	0.54	0.1	1.65
<0.22	2.245	2	1.15204	2.73688	87.24302	5.67729	3.24889	0.62746	2.02	0.54	0.1	1.65
<0.22	2.694	3	1.05489	3.58378	123.1461	28.26321	1.48195	0.38093	2.27	0.6	0.12	1.86
<0.22	2.694	3	1.16396	3.4515	148.9557	20.40351	2.31728	0.61125	2.27	0.6	0.12	1.86
<100kDa	1.313333	1	1.13021	2.60244	87.91043	6.52043	1.83462	0.22113	1.36	0.33	0.15	1.32
<100kDa	1.313333	1	1.1341	2.59756	78.94932	3.98538	2.31366	0.23831	1.36	0.33	0.15	1.32
<100kDa	1.97	2	1.05212	2.62702	79.54985	5.83435	2.73818	0.47268	1.82	0.44	0.21	1.79
<100kDa	1.97	2	1.04946	2.61692	122.0638	6.37925	2.43606	0.26851	1.82	0.44	0.21	1.79
<100kDa	2.364	3	1.04983	3.34341	137.5499	30.73865	2.05465	0.76415	2.05	0.49	0.23	2.01
<100kDa	2.364	3	1.059	3.30961	103.8735	7.81628	2.76399	0.46814	2.05	0.49	0.23	2.01
<30kDa	1.126667	1	1.06713	2.45315	71.5403	3.27706	2.03055	0.16651	1.22	0.3	0.15	1.37
<30kDa	1.126667	1	1.10133	2.46156	77.40851	3.50938	2.69254	0.29709	1.22	0.3	0.15	1.37
<30kDa	1.69	2	1.13518	2.64364	77.01194	4.12826	3.75043	0.57343	1.62	0.4	0.2	1.82
<30kDa	1.69	2	1.13497	2.6209	77.19131	5.75146	2.79894	0.49489	1.62	0.4	0.2	1.82
<30kDa	2.028	3	1.13793	2.9734	81.84559	5.82854	4.43917	1.60396	1.82	0.45	0.22	2.05
<30kDa	2.028	3	1.10771	3.42225	88.61096	5.53818	3.13988	0.5304	1.82	0.45	0.22	2.05

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<10kDa	0.91	1	1.08356	2.49737	84.47114	2.34622	2.33798	0.13304	0.9	0.23	0.31	1.34
<10kDa	0.91	1	1.11395	2.50852	83.97299	4.43004	2.39186	0.26479	0.9	0.23	0.31	1.34
<10kDa	1.365	2	1.07221	2.5884	79.19029	4.78536	2.30803	0.28182	1.2	0.31	0.41	1.79
<10kDa	1.365	2	1.0549	2.57876	80.63082	3.38063	2.83027	0.28895	1.2	0.31	0.41	1.79
<10kDa	1.638	3	1.06018	3.63473	126.2167	7.69851	1.78731	0.15464	1.35	0.35	0.46	2.01
<10kDa	1.638	3	1.0972	3.9249	167.7456	45.41422	1.77888	0.61627	1.35	0.35	0.46	2.01
<1kda		1	1.11503	2.52586	91.35907	4.56694	2.5593	0.28187				
<1kda		1	1.08046	2.53696	93.31778	2.46919	2.47861	0.13976				
<1kda		2	1.05327	2.6231	78.27487	3.74462	3.09179	0.38081				
<1kda		2	1.07518	2.66336	85.84803	5.33909	3.07563	0.52492				
<1kda		3	1.09197	3.59375	108.5629	5.89592	2.19386	0.21929				
<1kda		3	1.11001	3.32715	97.6687	25.53945	1.67771	0.61246				

Absorbance at 540 nm – Plates 1 through 20 from 1/29/03

1	1.764	2.8626	2.6289	2.0556	2.83	2.7722	2.2627	1.8574	1.2059	1.1314	1.0639	1.0918
	1.9581	2.7628	2.536	2.9053	2.9086	2.8117	2.3072	1.2147	1.0808	1.0569	1.1305	1.0682
	0.192	0.1513	0.0798	0.0776	0.0621	0.0543	0.0418	0.0402	0.0507	0.0412	0.0418	0.0382
	2.2656	2.3291	2.5862	2.3105	2.9611	3.1938	1.3443	1.2634	1.0835	1.04	1.0436	1.0372
	1.7933	2.0718	3.1148	3.2018	2.2598	3.2184	2.0021	1.9996	1.083	1.033	1.207	1.0863
	0.1819	0.0945	0.0885	0.0939	0.0765	0.055	0.0447	0.0419	0.041	0.0386	0.0378	0.0393
	2.084	2.3004	2.4244	2.4808	2.2168	2.8778	1.7573	1.1629	1.087	1.0576	1.0306	1.0174
	0.2814	0.1599	0.0879	0.0636	0.0674	0.0517	0.0614	0.042	0.0398	0.0393	0.0395	0.0404
2	2.435	2.3073	2.2764	2.7846	2.9507	2.6487	2.2532	1.4991	1.1655	1.6172	1.1238	1.0307
	2.5585	2.2526	1.6303	3.0311	2.9899	2.9772	1.6207	1.2516	1.1233	1.1048	1.1369	1.0851
	0.185	0.1139	0.0754	0.0534	0.0423	0.0406	0.0394	0.0384	0.0394	0.0391	0.0378	0.0389
	1.4792	2.4206	2.4303	2.8291	2.5585	2.1966	2.1861	2.5295	1.0807	1.434	0.9511	1.0581
	2.4209	2.636	2.9438	3.2256	3.0743	2.7667	1.3737	1.2373	1.6474	0.9921	0.9872	1.0163
	0.1105	0.1439	0.1035	0.0859	0.0536	0.0552	0.0444	0.0409	0.0407	0.0384	0.038	0.0441
	2.0507	2.1103	2.1854	2.6349	3.8404	2.0088	2.2976	1.4703	1.1289	1.1954	1.0229	1.2648
	0.1393	0.1501	0.058	0.0679	0.0487	0.0506	0.0448	0.0396	0.0388	0.0393	0.0394	0.0389

1 - FSOCN	1.764	2.8626	2.6289	2.0556	2.83	2.7722	2.2627	1.8574	1.2059	1.1314	1.0639	1.0918
	1.9581	2.7628	2.536	2.9053	2.9086	2.8117	2.3072	1.2147	1.0808	1.0569	1.1305	1.0682
	0.192	0.1513	0.0798	0.0776	0.0621	0.0543	0.0418	0.0402	0.0507	0.0412	0.0418	0.0382
	2.2656	2.3291	2.5862	2.3105	2.9611	3.1938	1.3443	1.2634	1.0835	1.04	1.0436	1.0372
	1.7933	2.0718	3.1148	3.2018	2.2598	3.2184	2.0021	1.9996	1.083	1.033	1.207	1.0863
	0.1819	0.0945	0.0885	0.0939	0.0765	0.055	0.0447	0.0419	0.041	0.0386	0.0378	0.0393
	2.084	2.3004	2.4244	2.4808	2.2168	2.8778	1.7573	1.1629	1.087	1.0576	1.0306	1.0174
	0.2814	0.1599	0.0879	0.0636	0.0674	0.0517	0.0614	0.042	0.0398	0.0393	0.0395	0.0404
2 - FSCON	2.435	2.3073	2.2764	2.7846	2.9507	2.6487	2.2532	1.4991	1.1655	1.6172	1.1238	1.0307
	2.5585	2.2526	1.6303	3.0311	2.9899	2.9772	1.6207	1.2516	1.1233	1.1048	1.1369	1.0851
	0.185	0.1139	0.0754	0.0534	0.0423	0.0406	0.0394	0.0384	0.0394	0.0391	0.0378	0.0389
	1.4792	2.4206	2.4303	2.8291	2.5585	2.1966	2.1861	2.5295	1.0807	1.434	0.9511	1.0581
	2.4209	2.636	2.9438	3.2256	3.0743	2.7667	1.3737	1.2373	1.6474	0.9921	0.9872	1.0163
	0.1105	0.1439	0.1035	0.0859	0.0536	0.0552	0.0444	0.0409	0.0407	0.0384	0.038	0.0441
	2.0507	2.1103	2.1854	2.6349	3.8404	2.0088	2.2976	1.4703	1.1289	1.1954	1.0229	1.2648
	0.1393	0.1501	0.058	0.0679	0.0487	0.0506	0.0448	0.0396	0.0388	0.0393	0.0394	0.0389
4 - FSCON	2.2245	2.8193	2.8788	2.8908	2.9527	2.6763	2.2316	1.2508	1.1366	1.0693	1.027	1.0072
	2.3815	2.8478	2.7559	2.828	2.9032	2.662	1.4617	1.1577	1.1509	1.1263	1.071	0.9949
	0.115	0.1342	0.0668	0.047	0.0477	0.039	0.0377	0.0399	0.0382	0.0388	0.0394	0.0389
	2.4872	2.9112	3.0264	3.0984	2.4381	2.6271	1.5456	1.264	1.129	1.05	1.0436	1.0576
	2.5512	2.99	3.1882	1.9635	3.1977	1.6844	1.6793	2.193	1.1076	1.0893	1.036	0.9998
	0.2072	0.1304	0.0751	0.0541	0.0547	0.049	0.0461	0.0411	0.0381	0.0409	0.0389	0.0396
	1.6799	3.007	2.8907	2.6869	3.2804	1.5415	1.8726	1.1331	1.6084	1.011	1.0003	0.9476
	0.1935	0.1004	0.0738	0.0674	0.05	0.0432	0.0431	0.0405	0.0401	0.0394	0.0393	0.039
5 - FSCON	2.8292	2.3697	3.1076	2.9377	2.9399	1.9757	2.1962	1.0651	1.1138	1.1596	1.0056	0.9725
	2.8206	2.879	3.0452	2.8863	2.9181	2.6797	1.6037	1.1878	1.3305	1.5891	1.0128	1.1229

	0.1619	0.1202	0.0568	0.0407	0.0655	0.0406	0.0372	0.0387	0.0392	0.0375	0.0383	0.0393
	2.5255	2.7188	2.8946	3.2575	3.1841	2.6584	1.9175	1.8375	1.112	1.0479	1.0561	0.9543
	2.7035	2.5434	3.316	2.9343	2.7556	2.1584	1.4168	1.7869	1.0439	1.0574	0.9708	0.936
	0.1111	0.0875	0.0713	0.0443	0.0422	0.0397	0.0395	0.0405	0.0376	0.0394	0.037	0.0391
	2.4848	1.8104	2.3597	2.7452	2.4296	2.2923	1.1248	1.2323	1.0289	1.0074	1.0178	1.0558
	0.1615	0.1013	0.0995	0.063	0.0429	0.0402	0.0464	0.0407	0.0399	0.0386	0.0403	0.0395

7	2.9743	3.0148	2.9215	2.9623	3.0044	2.0237	1.4847	1.1652	1.5941	1.1036	1.0295	1.0349
	2.9245	2.9714	2.9667	3.0949	2.4214	2.7018	2.0908	1.2339	1.6068	1.5498	1.07	1.0303
	0.2255	0.1861	0.1033	0.0555	0.0453	0.043	0.0383	0.0393	0.0388	0.0402	0.041	0.041
	2.8893	3.0997	2.9739	2.9839	3.0329	2.5769	1.4069	1.1939	1.1122	0.9615	0.9851	1.0679
	2.9316	3.083	3.0985	3.0647	3.0848	2.6112	1.3576	1.194	1.2433	1.0794	1.1154	1.0421
	0.2645	0.3123	0.1387	0.0963	0.0458	0.0418	0.0524	0.0406	0.0423	0.0384	0.0378	0.0402
	3.1897	3.5882	3.8945	2.9202	3.1237	1.9168	1.7703	1.0939	1.0573	1.0212	1	0.9691
	0.2666	0.2129	0.0937	0.0656	0.0633	0.0445	0.0401	0.04	0.0412	0.0395	0.0411	0.0403
8	2.9747	2.9645	2.9323	2.9588	2.9601	2.0492	2.2063	1.8296	1.1462	1.1053	1.0511	1.0438
	2.9684	2.8458	2.98	2.9418	2.9328	2.7479	1.59	1.2635	1.1658	1.1569	1.0291	0.9902
	0.1592	0.1165	0.1073	0.0623	0.0527	0.0397	0.0371	0.0379	0.0383	0.0406	0.0386	0.0387
	3.0036	3.1251	3.1329	3.2219	3.2679	2.782	1.5266	1.295	1.1913	1.0554	1.0889	1.0063
	3.1196	3.0733	3.3026	3.2106	3.2735	1.9152	1.4365	1.4646	1.1163	1.2523	1.0671	1.0853
	0.2216	0.21	0.1156	0.053	0.0639	0.0413	0.039	0.0417	0.0386	0.0409	0.0392	0.0381
	3.3577	3.511	3.6396	3.6983	2.1685	1.6267	1.91	1.1643	1.0923	1.2267	1.0016	0.9363
	0.1707	0.1728	0.0924	0.0646	0.0565	0.0452	0.0489	0.0426	0.0402	0.0391	0.0385	0.0396

10	2.9021	2.9092	2.9388	2.9178	2.8889	1.8182	1.4014	1.7605	1.2628	0.9806	1.0052	0.9223
	2.891	2.8555	2.8947	2.8745	2.8685	2.6782	2.3806	1.116	1.5878	1	0.9664	0.9398
	0.141	0.1147	0.0913	0.0923	0.0444	0.0402	0.0381	0.038	0.0387	0.0405	0.0374	0.0386
	2.9921	3.0862	2.9992	3.2107	3.1155	1.9357	1.3825	1.7386	1.0739	1.0607	0.9249	0.9733
	2.9961	3.0319	2.967	3.1783	3.1798	2.598	1.9795	1.0994	1.1637	0.9798	0.8706	0.9238
	0.1631	0.1107	0.0799	0.0619	0.0508	0.0455	0.0403	0.042	0.0387	0.0418	0.0387	0.0395
	3.406	3.133	3.4979	3.7548	2.0302	2.1546	1.2003	1.0422	1.0232	0.9947	0.9692	0.9174
	0.1335	0.2097	0.0548	0.0456	0.0462	0.0436	0.045	0.0403	0.04	0.0386	0.0378	0.0388
11	2.8848	2.9005	2.8882	2.9885	3.0105	2.7003	2.133	1.7466	0.9043	1.4622	0.8736	0.8819
	2.8172	2.9084	2.8879	2.8738	2.9456	2.7073	2.0611	1.7194	1.5799	0.9712	0.9731	1.0375
	0.1372	0.0993	0.1509	0.0501	0.0408	0.0381	0.045	0.0506	0.0391	0.0373	0.0377	0.0374
	2.9483	3.0492	3.0056	3.1734	2.8855	1.5093	1.2413	1.0946	1.5346	1.1161	1.0224	0.9706
	2.9674	3.0862	3.0632	3.0596	2.049	2.2622	1.1669	1.6195	1.0111	0.9372	0.9604	0.9712
	0.1017	0.0833	0.066	0.0482	0.0709	0.0479	0.0433	0.0527	0.0398	0.0384	0.0377	0.0424
	3.4989	3.6872	3.8685	3.4237	3.2919	1.6422	1.2315	1.0004	0.9754	0.9243	0.9553	0.9307
	0.1145	0.0857	0.0598	0.0456	0.0447	0.0426	0.0412	0.0393	0.0387	0.0405	0.0399	0.0384

13	2.8124	2.8155	2.88	2.848	2.7032	2.2688	2.1528	1.3962	1.1311	1.0234	0.9274	1.0902
	2.7955	2.9382	2.8939	2.9116	2.957	2.169	2.1822	1.7408	1.0538	1.0196	0.9981	0.9932
	0.1138	0.1155	0.0676	0.0641	0.051	0.0396	0.04	0.0389	0.0397	0.0383	0.0392	0.0408
	2.8545	2.981	3.0745	3.0873	3.0643	2.1578	1.4345	1.2808	1.106	1.6134	1.0646	1.0916
	2.9414	3.0897	3.0954	3.2657	3.1878	2.7912	1.3436	1.1869	1.1398	1.0816	1.0479	0.9723
	0.1254	0.1057	0.0613	0.0669	0.0562	0.0405	0.0421	0.0399	0.0425	0.0394	0.0395	0.0393
	3.3565	3.6653	3.5746	3.1763	3.3774	2.3281	1.3169	1.1403	1.0788	1.233	1.1337	0.9989
	0.1387	0.1045	0.0616	0.0495	0.054	0.0421	0.0426	0.0413	0.039	0.0402	0.0473	0.0397
14	2.8193	2.9131	2.9587	2.8584	2.3435	2.6771	2.0888	1.202	1.1197	1.267	1.2689	1.0773
	2.8734	2.8991	2.9593	2.984	2.8612	2.7053	2.1447	1.742	1.5895	1.1168	0.8957	1.4785
	0.1144	0.1562	0.0583	0.0623	0.0491	0.0412	0.037	0.0386	0.0394	0.0385	0.0376	0.0394
	2.9772	3.0894	3.0151	3.0781	2.4357	2.6812	1.4278	1.2709	1.0989	1.5531	1.2405	1.1197
	3.0516	3.1298	3.1195	3.1078	2.481	2.7235	1.4065	1.7282	1.1447	1.1499	0.9463	1.0455
	0.1282	0.1122	0.0667	0.0593	0.0524	0.0418	0.0412	0.0459	0.0399	0.0418	0.0369	0.0386
	3.7001	3.3848	3.7707	4	3.6185	2.3526	1.7964	1.1572	1.0931	0.9832	1.0449	1.0848
	0.1096	0.1516	0.0603	0.0576	0.0448	0.0435	0.0473	0.043	0.0396	0.0397	0.0387	0.042

16	2.8637	2.9006	2.8726	2.911	2.8629	2.6109	2.0609	1.3861	1.1599	1.0907	1.0601	1.0149
	2.9109	2.9505	2.8856	2.9157	2.8917	2.6667	2.0017	1.5937	1.159	1.1235	0.9309	1.3711
	0.1638	0.1531	0.1088	0.0473	0.0395	0.0378	0.0371	0.0387	0.0384	0.0378	0.0368	0.0367
	3.25	2.9697	2.9961	2.9608	2.9879	2.4938	1.933	1.4581	1.5877	1.0768	1.1844	1.1033
	3.086	3.0777	3.1424	3.1622	3.0817	2.4329	1.9258	1.3609	1.544	1.5503	1.0232	1.0162
	0.1787	0.191	0.1025	0.0736	0.0527	0.0399	0.0405	0.0385	0.0402	0.0377	0.0379	0.0386
	3.3009	3.4945	4	3.5444	3.0456	2.1307	1.6978	1.1154	1.5489	1.0272	0.8979	0.8557
	0.2283	0.2904	0.1019	0.0662	0.0558	0.0422	0.0422	0.0402	0.0389	0.0375	0.0379	0.0379
17	2.924	2.8789	2.911	2.8868	2.8639	2.6916	2.1146	1.6844	1.6125	1.2197	1.0772	1.0602
	2.9148	2.9584	2.9117	2.8896	2.8975	2.6849	1.9886	1.6871	1.6094	1.1921	1.0169	0.936
	0.0487	0.112	0.0842	0.0553	0.0962	0.039	0.0363	0.0438	0.0378	0.0373	0.0373	0.0395
	3.3378	3.0306	3.0507	3.0562	3.0352	2.5804	1.2994	1.1189	1.2216	0.9455	1.2345	1.0145
	3.019	3.0751	3.2055	3.0817	3.0345	2.4859	1.9283	1.6437	1.0223	1.0262	0.9957	1.0373
	0.1694	0.1081	0.0581	0.0608	0.0405	0.0398	0.0401	0.0386	0.0382	0.0377	0.037	0.0387
	3.5173	3.6892	3.9814	4	2.0837	2.2777	1.8193	1.0284	1.0637	1.0728	1.0646	1.0837
	0.1783	0.1374	0.108	0.0535	0.0509	0.0522	0.0397	0.04	0.0384	0.0399	0.0385	0.0384

19	2.8199	2.8953	2.9082	2.9423	2.9232	2.6656	2.1051	1.6781	1.0339	1.0125	1.0501	0.8895
	2.9003	2.9283	2.9346	2.9223	2.8717	2.591	1.4054	1.6716	1.5951	1.0156	1.0401	1.0025
	0.0919	0.0851	0.0794	0.0511	0.0456	0.0395	0.0357	0.0376	0.0385	0.0395	0.0379	0.0366
	2.9496	3.0545	3.136	3.1525	3.0579	2.5099	1.9909	1.1769	1.0828	1.5543	1.5179	1.0748
	2.9956	3.1118	3.1986	3.1488	3.2184	2.6357	1.3896	1.1289	1.072	1.0461	1.0078	0.8921
	0.0913	0.1114	0.0883	0.0689	0.0508	0.0488	0.0452	0.0404	0.0414	0.0396	0.0393	0.0391
	3.3245	3.7578	3.8867	3.8903	3.1237	2.1914	1.3754	1.0292	0.9613	1.0272	0.9725	0.9819
	0.1074	0.1109	0.0594	0.0557	0.0593	0.041	0.0489	0.0401	0.0395	0.0409	0.0387	0.0454
20	2.8674	2.9516	2.9223	2.9129	2.8797	2.6576	2.0281	1.1985	1.0566	1.0046	0.9903	1.1891
	2.9178	2.9352	2.9049	2.5581	2.8867	2.4598	1.9666	1.0947	1.0296	1.0423	0.9761	0.9919
	0.0981	0.0831	0.0733	0.0551	0.0402	0.0393	0.0415	0.0415	0.0388	0.0415	0.0398	0.0392
	3.0178	3.0801	3.0789	3.1485	3.0625	2.0594	1.3223	1.3488	1.1851	1.0098	1.2613	0.9902
	3.0685	3.1736	3.1665	3.2322	3.3242	2.5818	1.4259	1.1439	1.0849	1.0046	1.0193	0.9912
	0.2039	0.0789	0.06	0.0608	0.0561	0.0467	0.0455	0.0407	0.041	0.0384	0.0393	0.0403
	3.5985	3.8546	3.8246	2.6393	3.2725	2.3165	1.3817	1.6434	1.1875	1.0119	1.2671	0.9561
	0.1623	0.0898	0.077	0.0537	0.0626	0.052	0.0511	0.0414	0.0395	0.0407	0.0386	0.0456

Absorbance at 620 nm – Plates 1 through 20 from 1/29/03

1	0.964	1.1135	1.113	0.9084	1.102	1.0393	1.2458	1.2703	0.8323	0.835	0.7812	0.8049
	1.0389	1.1181	0.9631	1.1303	1.1135	1.2506	1.2904	0.758	0.7315	0.7547	0.8337	0.7934
	0.1587	0.1279	0.064	0.0647	0.0564	0.0475	0.0395	0.0384	0.0474	0.0388	0.0402	0.0362
	1.0394	1.0211	1.0725	0.9265	1.0884	1.2175	0.7972	0.8559	0.7594	0.7405	0.7595	0.7701
	0.924	1.1451	1.1314	1.1722	1.0182	1.2406	1.2751	1.1275	0.7433	0.737	0.893	0.7953
	0.1509	0.0876	0.0815	0.0697	0.0674	0.0506	0.0426	0.0403	0.0397	0.0377	0.0371	0.0386
	0.9707	1.0246	0.973	0.9023	0.8483	1.0582	1.1781	0.9235	0.7672	0.7557	0.7508	0.7526
	0.2391	0.1578	0.0823	0.0618	0.0604	0.0466	0.0572	0.0402	0.0383	0.0377	0.0379	0.039
2	1.0305	0.9402	1.0454	1.1566	1.1821	1.1673	0.8761	1.0076	0.7929	1.2459	0.8361	0.7709
	1.0461	0.9294	0.9242	1.2364	1.2434	1.2357	0.9148	0.785	0.7692	0.7925	0.8469	0.8164
	0.1537	0.1021	0.0692	0.0484	0.0379	0.0377	0.0379	0.0372	0.0371	0.0376	0.0366	0.0367
	0.9734	1.0025	1.0153	0.9961	1.0291	0.9899	1.0242	0.897	0.7648	0.9646	0.6892	0.7866
	1.0346	1.0477	1.1421	1.2561	1.1777	1.3797	0.805	0.7951	1.2467	0.6993	0.711	0.7487
	0.0967	0.1185	0.0824	0.0654	0.0482	0.0503	0.0428	0.0389	0.0386	0.0368	0.0368	0.0423
	0.9893	1.0164	0.8705	0.9961	1.1387	0.9326	0.8711	0.8263	0.8175	0.827	0.7504	0.8292
	0.1305	0.131	0.0523	0.0598	0.0468	0.0482	0.0428	0.0383	0.0367	0.0379	0.0381	0.0382

4	0.9975	1.168	1.198	1.2101	1.2138	1.1625	1.2277	0.7535	0.7587	0.7513	0.7307	0.7206
	1.1133	1.1185	1.0933	1.0945	1.2134	1.1896	0.7541	0.7021	0.776	0.8047	0.7811	0.7204
	0.098	0.1001	0.0612	0.0431	0.044	0.0366	0.0359	0.0372	0.036	0.0373	0.0375	0.0375
	0.9528	1.0821	1.1907	1.2285	0.9035	1.2436	0.8235	0.8324	0.7811	0.7511	0.7579	0.7845
	1.1633	1.1283	1.2439	0.8424	1.2722	0.993	0.8944	0.8508	0.7695	0.7811	0.757	0.7405
	0.1419	0.0992	0.0579	0.0495	0.0452	0.0467	0.0434	0.0398	0.0371	0.0397	0.0373	0.0381
	0.8589	1.2122	1.0575	0.9892	1.2314	0.8067	1.2321	0.7759	1.2234	0.7393	0.7344	0.6909
	0.1531	0.0885	0.0668	0.0618	0.0458	0.0406	0.0407	0.0385	0.0382	0.0377	0.0372	0.0375
5	1.0987	1.0317	1.2526	1.1875	1.2071	0.9442	1.2125	0.6347	0.7393	0.7603	0.7018	0.6842
	1.0603	1.0831	1.2154	1.161	1.2091	1.2844	0.8636	0.7469	0.872	1.2205	0.7326	0.7826
	0.1532	0.0992	0.0532	0.0384	0.0505	0.0383	0.0358	0.0368	0.0372	0.0361	0.0368	0.0379
	1.0142	1.0371	1.098	1.2711	1.3337	1.3018	1.2288	1.2891	0.7773	0.7433	0.7737	0.6875
	1.1407	0.9626	1.257	1.1223	1.1084	0.8783	0.8086	1.2683	0.7125	0.751	0.6788	0.6727
	0.1166	0.0798	0.0649	0.0416	0.039	0.0381	0.0381	0.0392	0.0368	0.038	0.0359	0.0378
	0.9682	0.8753	0.8869	0.8579	1.0781	1.2653	0.7967	0.779	0.7188	0.7267	0.7453	0.7294
	0.1462	0.1001	0.0736	0.0501	0.0405	0.0388	0.0436	0.0385	0.0385	0.0371	0.0381	0.0374

7	1.2817	1.2455	1.2122	1.2424	1.2542	0.8778	0.8083	0.7351	1.1573	0.783	0.7552	0.7674
	1.2477	1.2357	1.2145	1.2985	0.8289	1.2666	1.2204	0.7864	1.1968	1.2034	0.7772	0.7636
	0.1748	0.1509	0.0788	0.0484	0.0415	0.0398	0.0364	0.0374	0.0369	0.0384	0.0394	0.0388
	1.2153	1.2648	1.1922	1.2052	1.2478	1.2576	0.8263	0.8073	0.7973	0.698	0.7166	0.7885
	1.2003	1.2221	1.2175	1.2181	1.2484	1.2962	0.8309	0.7966	0.8172	0.7707	0.8122	0.7713
	0.1965	0.2174	0.102	0.0675	0.0418	0.0403	0.049	0.0395	0.0409	0.0376	0.0368	0.0391
	1.2429	1.2517	1.2415	0.9355	1.2364	0.9473	1.1723	0.7491	0.764	0.743	0.7424	0.7214
	0.1722	0.1978	0.066	0.0546	0.0502	0.0445	0.0387	0.0385	0.0394	0.0382	0.0395	0.0388
8	1.2096	1.192	1.1817	1.1755	1.2025	0.8088	1.1856	1.2309	0.7743	0.7912	0.7568	0.7637
	1.2219	1.1612	1.2157	1.2133	1.2369	1.2448	0.8703	0.8092	0.8029	0.8401	0.7513	0.7184
	0.1327	0.123	0.0852	0.0567	0.0437	0.0373	0.0356	0.0359	0.0365	0.0387	0.037	0.0375
	1.1918	1.2128	1.2369	1.2971	1.2994	1.3166	0.88	0.874	0.8467	0.7469	0.7511	0.7245
	1.1956	1.1829	1.2629	1.2426	1.2732	0.8757	0.8259	0.8752	0.7641	0.8607	0.7712	0.7437
	0.186	0.139	0.0946	0.0469	0.0507	0.0393	0.0375	0.04	0.0377	0.0397	0.0374	0.0367
	1.1857	1.2145	1.2035	1.253	0.8176	0.8645	1.2898	0.8069	0.7592	0.8618	0.7281	0.6799
	0.1928	0.1217	0.0726	0.0587	0.0512	0.0455	0.0457	0.0404	0.0385	0.0376	0.0371	0.0438

10	1.2076	1.1827	1.2164	1.1998	1.1885	0.7593	0.7778	1.1831	0.8216	0.7028	0.7379	0.6734
	1.2028	1.1527	1.1607	1.1566	1.1831	1.2285	1.1736	0.714	1.1951	0.717	0.7205	0.699
	0.1363	0.1351	0.0795	0.0603	0.0399	0.0374	0.0365	0.0359	0.0363	0.0382	0.0358	0.0372
	1.2204	1.2262	1.1933	1.3029	1.2792	0.7686	0.8078	1.2358	0.775	0.7776	0.6732	0.7294
	1.1649	1.1905	1.1672	1.2506	1.2558	1.2895	1.2384	0.7145	0.766	0.7145	0.6497	0.6751
	0.1621	0.1299	0.064	0.0465	0.0441	0.0417	0.0385	0.0402	0.0376	0.0397	0.0372	0.0379
	1.3262	1.1821	1.2187	1.2584	0.8041	1.217	0.7843	0.7347	0.7448	0.7458	0.7294	0.691
	0.1379	0.1466	0.0456	0.0429	0.0438	0.0414	0.0425	0.0385	0.0381	0.037	0.0358	0.0375
11	1.1938	1.166	1.1773	1.2391	1.2506	1.223	1.1799	1.1886	0.6327	1.1301	0.6346	0.6433
	1.1801	1.1655	1.1548	1.1559	1.2391	1.2418	1.2161	1.1965	1.1912	0.7032	0.7258	0.7867
	0.1269	0.087	0.1376	0.0429	0.0381	0.0361	0.041	0.048	0.0371	0.0356	0.0363	0.0357
	1.1864	1.2179	1.2023	1.2882	1.2368	0.7703	0.7785	0.7551	1.196	0.7825	0.775	0.7218
	1.1535	1.1916	1.179	1.2142	0.7772	1.2565	0.6992	1.1958	0.7154	0.6785	0.7021	0.7311
	0.1589	0.0986	0.0594	0.0541	0.0461	0.0445	0.0411	0.0496	0.0381	0.0374	0.0369	0.0411
	1.206	1.213	1.2535	1.1862	1.2832	0.8107	0.7617	0.677	0.7023	0.6558	0.7087	0.693
	0.136	0.0792	0.0553	0.0433	0.042	0.0405	0.0395	0.0375	0.037	0.0386	0.0383	0.0373

13	1.2332	1.2311	1.2028	1.2098	1.149	0.8203	1.1589	0.7818	0.7783	0.7159	0.6546	0.8144
	1.1971	1.2275	1.2004	1.1853	1.2659	0.8484	1.2065	1.1913	0.7059	0.7194	0.7187	0.7236
	0.099	0.0946	0.0684	0.0632	0.0484	0.0374	0.0384	0.0372	0.0378	0.0366	0.0376	0.0383
	1.1805	1.2188	1.2419	1.2644	1.2631	0.8418	0.8476	0.8399	0.7723	1.2683	0.7785	0.8073
	1.1888	1.2143	1.2091	1.3045	1.2603	1.3112	0.7786	0.7638	0.8033	0.7718	0.7514	0.6938
	0.116	0.0849	0.0583	0.0563	0.0555	0.0386	0.0405	0.0388	0.04	0.0381	0.0386	0.0386
	1.18	1.2514	1.238	1.1887	1.2517	1.2805	0.8284	0.7765	0.7498	0.8525	0.7853	0.7201
	0.1178	0.0943	0.054	0.0464	0.0525	0.039	0.041	0.0391	0.0371	0.0381	0.0456	0.0383
14	1.2363	1.2593	1.2321	1.196	0.7796	1.2004	1.1461	0.7789	0.7872	0.9028	0.9132	0.8102
	1.29	1.2169	1.2212	1.2472	1.1973	1.2214	1.2026	1.2208	1.204	0.8262	0.6472	1.1749
	0.1171	0.1098	0.0525	0.0521	0.0433	0.0384	0.0356	0.0364	0.037	0.0366	0.0359	0.036
	1.2526	1.2544	1.2265	1.2384	0.855	1.3008	0.8398	0.8404	0.7712	1.2127	0.915	0.8431
	1.2492	1.2408	1.2393	1.2304	0.8896	1.3063	0.8124	1.2274	0.7252	0.8077	0.6813	0.7685
	0.1524	0.0723	0.0559	0.0432	0.048	0.0404	0.0394	0.0431	0.0374	0.04	0.0358	0.0371
	1.2387	1.2211	1.2631	1.2978	1.3282	1.2535	1.1944	0.7918	0.7603	0.6981	0.7636	0.8101
	0.1124	0.1258	0.0565	0.062	0.042	0.0415	0.0439	0.0406	0.0379	0.0379	0.0368	0.0404

16	1.2124	1.2131	1.1996	1.21	1.2057	1.2088	1.1653	0.8783	0.7953	0.7884	0.7756	0.7469
	1.2161	1.211	1.1975	1.231	1.2319	1.2525	1.1642	1.1391	0.8053	0.8141	0.6882	1.1053
	0.1351	0.1414	0.0858	0.0431	0.037	0.0364	0.0374	0.0366	0.0367	0.0363	0.0357	0.0356
	1.5786	1.2007	1.214	1.2116	1.2648	1.2694	1.2127	0.9802	1.2167	0.7941	0.8546	0.7804
	1.2148	1.2027	1.221	1.2527	1.2713	1.2283	1.2146	0.8404	1.1829	1.2196	0.7471	0.7567
	0.1389	0.1231	0.0854	0.0574	0.0464	0.0382	0.0384	0.037	0.0382	0.0365	0.053	0.038
	1.1924	1.2663	1.2796	1.2366	1.2362	1.1957	1.1706	0.7761	1.2139	0.7519	0.6515	0.6105
	0.1676	0.2394	0.1046	0.0614	0.0508	0.0403	0.0403	0.0385	0.0375	0.037	0.0366	0.0367
17	1.2249	1.1988	1.2049	1.1983	1.19	1.2055	1.235	1.1352	1.1927	0.8645	0.7992	0.7977
	1.2384	1.2191	1.2029	1.204	1.2237	1.2315	1.1672	1.1563	1.2064	0.855	0.7379	0.6732
	0.0464	0.1001	0.0764	0.0512	0.0803	0.0376	0.0353	0.0411	0.0358	0.0358	0.0363	0.0379
	1.643	1.2252	1.2535	1.2778	1.3183	1.3048	0.7731	0.7431	0.8232	0.6807	0.9019	0.7557
	1.1916	1.1999	1.2492	1.2521	1.2393	1.2132	1.2357	1.1839	0.7154	0.7318	0.724	0.773
	0.1236	0.0886	0.0513	0.0546	0.038	0.0381	0.038	0.0371	0.0364	0.0365	0.0364	0.0379
	1.2021	1.2376	1.2515	1.318	0.8407	1.2685	1.2364	0.7071	0.7564	0.7887	0.7927	0.8155
	0.1518	0.1201	0.093	0.0501	0.0461	0.0485	0.0376	0.038	0.0372	0.0382	0.0374	0.0369

19	1.2068	1.1957	1.2042	1.2138	1.1868	1.2009	1.1853	1.1597	0.7123	0.7241	0.71	0.6435
	1.206	1.2043	1.2062	1.2244	1.2163	1.2303	0.7741	1.1825	1.2098	0.733	0.7695	0.7417
	0.081	0.0697	0.0642	0.0462	0.0413	0.0376	0.035	0.0363	0.0367	0.038	0.0367	0.0352
	1.2246	1.2346	1.2741	1.2872	1.2809	1.1971	1.2459	0.7841	0.7681	1.2199	1.2069	0.8103
	1.1958	1.2208	1.2429	1.2297	1.2734	1.2954	0.8816	0.744	0.7568	0.7703	0.7451	0.6428
	0.082	0.07	0.0741	0.0573	0.0446	0.045	0.0432	0.0388	0.0399	0.0385	0.0379	0.0381
	1.2096	1.2327	1.242	1.255	1.2213	1.1875	0.8137	0.7108	0.6827	0.7426	0.7251	0.727
	0.0912	0.0831	0.0538	0.0481	0.0505	0.0391	0.0453	0.038	0.0374	0.0388	0.0375	0.0437
20	1.2073	1.2274	1.2124	1.2136	1.1907	1.1998	1.1677	0.765	0.7521	0.7338	0.74	0.8925
	1.2174	1.1972	1.1961	0.7721	1.2181	1.2025	1.2316	0.7167	0.7284	0.7729	0.7171	0.7407
	0.076	0.0678	0.061	0.0491	0.037	0.0373	0.0397	0.0394	0.0368	0.0395	0.0378	0.0368
	1.2213	1.2344	1.2219	1.2839	1.2557	0.8776	0.7908	0.8242	0.7845	0.7278	0.946	0.7293
	1.2147	1.2167	1.2197	1.2554	1.3052	1.2999	0.7568	0.7377	0.7729	0.7312	0.7485	0.7399
	0.1017	0.0653	0.0556	0.0524	0.049	0.0426	0.043	0.0391	0.0391	0.0375	0.0377	0.0387
	1.2329	1.2271	1.2317	0.7876	1.2449	1.2473	0.7549	1.2378	0.7976	0.7256	0.9303	0.7013
	0.0822	0.069	0.0613	0.0462	0.0573	0.0458	0.0463	0.0388	0.0372	0.0387	0.0372	0.0442

Absorbance at 620 nm for Blanks – Plates 3 through 21 from 1/29/03

3	0.852	0.9823	0.8476	1.1445	0.9106	0.8944	1.055	0.7715	0.787	0.8586	0.8389	0.832
	0.1122	0.1106	0.0891	0.0596	0.0465	0.042	0.039	0.0387	0.0393	0.0385	0.0375	0.0362
	0.947	0.8619	0.9165	0.9007	0.8848	0.979	0.9615	0.8577	0.7472	0.8033	0.9141	0.7952
	0.1805	0.0954	0.1028	0.0564	0.0664	0.0421	0.0467	0.0403	0.041	0.0368	0.0383	0.039
	0.8586	0.9164	0.9851	1.0646	0.9608	0.9758	0.9109	0.8593	1.0822	0.8523	0.7832	0.7744
	0.2031	0.1231	0.0993	0.0606	0.0514	0.0643	0.0541	0.0427	0.0387	0.037	0.0381	0.0385
	0.0477	0.0469	0.0463	0.0461	0.0456	0.0461	0.0474	0.0468	0.047	0.0465	0.0468	0.0478
	0.0458	0.0458	0.047	0.0485	0.0454	0.0876	0.0601	0.046	0.0465	0.0469	0.0473	0.0463
6	0.8318	0.8646	0.7048	0.8498	0.8176	0.9628	0.7075	0.7115	0.8065	0.7947	0.7398	0.7802
	0.105	0.0727	0.0902	0.0653	0.0446	0.0441	0.041	0.0388	0.0362	0.0375	0.0383	0.0381
	0.8668	1.015	0.9006	0.8316	0.7782	0.8207	0.8642	0.8321	0.8045	0.8158	0.8155	0.7563
	0.1743	0.0866	0.0591	0.0591	0.0484	0.0384	0.0428	0.0393	0.0412	0.0372	0.0363	0.0375
	0.9592	0.8379	0.9389	0.9183	0.9791	0.8339	0.853	0.841	0.8947	0.7841	0.8336	0.7399
	0.1117	0.0875	0.0563	0.0676	0.0443	0.0697	0.0407	0.0379	0.038	0.0366	0.0361	0.4731
	0.0456	0.0468	0.0472	0.0455	0.0455	0.0463	0.0458	0.047	0.0469	0.0467	0.0468	0.047
	0.0459	0.0462	0.0466	0.0481	0.0453	0.0459	0.0454	0.0459	0.0462	0.0469	0.047	0.0455

9	1.0797	0.719	0.7441	0.7806	0.7908	0.7448	0.6982	0.7178	0.7786	0.709	0.6703	0.7634
	0.0838	0.1795	0.083	0.0542	0.0459	0.0442	0.0403	0.0374	0.038	0.0392	0.0376	0.0372
	0.7274	0.7731	0.8292	0.7578	0.7804	0.8553	0.7722	0.8439	0.782	0.8466	0.727	0.7622
	0.1517	0.1602	0.1505	0.1049	0.0595	0.0402	0.0543	0.0388	0.0377	0.0382	0.0387	0.0379
	0.8181	0.7997	0.7838	0.8167	0.854	0.8515	0.8109	0.8519	0.7887	0.8066	0.8002	0.8296
	0.3093	0.1928	0.1258	0.0839	0.066	0.0401	0.0427	0.0394	0.038	0.0399	0.0368	0.0374
	0.0479	0.0511	0.0478	0.0463	0.0491	0.0476	0.0487	0.0461	0.0463	0.0501	0.0462	0.0464
	0.0468	0.0467	0.0492	0.0505	0.0517	0.0499	0.0524	0.0472	0.0472	0.0474	0.0468	0.0473
12	1.1048	0.7284	0.724	1.1131	0.7704	0.8072	0.7187	0.7754	0.8469	0.7694	0.9056	1.1078
	0.0621	0.0981	0.0532	0.0491	0.0534	0.0491	0.0392	0.0371	0.0382	0.0384	0.0367	0.0402
	0.6695	0.6628	0.7239	0.7616	0.7747	0.7322	0.8471	0.7824	0.8421	0.7682	0.8177	0.7132
	0.1602	0.075	0.2092	0.0704	0.0395	0.0449	0.0422	0.044	0.0505	0.0377	0.0366	0.0413
	0.7989	0.7711	0.8052	0.7357	0.7982	0.8312	0.8027	0.8422	0.8247	0.8301	0.7401	0.6631
	0.1666	0.0611	0.0617	0.0486	0.0581	0.0494	0.046	0.0438	0.0383	0.0372	0.0367	0.037
	0.0471	0.0462	0.0464	0.046	0.0458	0.0475	0.0462	0.0457	0.047	0.0462	0.0466	0.0456
	0.0465	0.0467	0.0471	0.047	0.0457	0.046	0.046	0.0459	0.0485	0.0458	0.0462	0.0458

15	0.7616	0.7872	0.8225	0.8439	0.7611	0.796	0.8018	0.8118	0.8216	0.8373	0.7495	0.7146
	0.1308	0.0925	0.0672	0.0576	0.046	0.0424	0.0396	0.0377	0.0389	0.0382	0.0374	0.0378
	0.6462	1.2348	0.8157	0.7994	0.787	0.7775	0.7864	0.7977	0.7795	0.7656	0.7253	0.7722
	0.0883	0.1745	0.0636	0.054	0.0547	0.0503	0.039	0.038	0.0395	0.038	0.0376	0.0377
	1.0586	0.7332	0.8029	0.7892	0.7638	0.7623	0.7665	0.7495	0.7632	0.7848	0.6917	0.7474
	0.1119	0.1209	0.0756	0.0637	0.0773	0.0438	0.0425	0.0409	0.0428	0.0401	0.0368	0.0371
	0.0474	0.0504	0.0475	0.0463	0.0485	0.0475	0.0486	0.0459	0.0463	0.0474	0.0466	0.047
	0.0466	0.0465	0.0493	0.0466	0.0476	0.0492	0.0484	0.0469	0.0469	0.0474	0.0462	0.0469
18	1.1124	0.7231	0.7354	0.7957	0.7732	0.8592	0.7462	0.7884	0.7342	0.7899	0.7722	0.7299
	0.0667	0.1303	0.1252	0.0759	0.0552	0.0385	0.0476	0.0378	0.0399	0.0403	0.0364	0.0376
	0.6856	0.7321	0.7151	0.8223	0.7931	0.7755	0.7818	0.7253	0.7632	0.7084	0.7201	0.7415
	0.118	0.2626	0.1827	0.0858	0.045	0.0451	0.0393	0.0389	0.038	0.0368	0.0366	0.0381
	0.7351	0.7101	0.7735	0.7429	0.7778	0.8358	1.0209	0.8634	0.8164	0.7653	0.7791	0.744
	0.0444	0.1404	0.0802	0.056	0.0501	0.0421	0.0418	0.0399	0.0374	0.0385	0.0366	0.0371
	0.0496	0.0473	0.0467	0.0454	0.0457	0.0469	0.0462	0.0473	0.0468	0.0472	0.0493	0.046
	0.0464	0.046	0.0463	0.0466	0.0458	0.0461	0.046	0.0456	0.0479	0.0464	0.0464	0.0462

21	0.9477	1.1252	0.7239	1.1224	0.7535	0.7477	0.7205	0.7762	0.7747	0.7044	0.6995	0.6524
	0.065	0.1065	0.076	0.0699	0.0454	0.0405	0.0481	0.0381	0.038	0.0381	0.0384	0.0367
	0.662	1.2361	0.6933	0.6842	0.7282	0.7317	0.7459	0.796	0.7222	0.683	0.6588	0.6878
	0.0884	0.0942	0.0528	0.058	0.0581	0.0501	0.0469	0.0398	0.0387	0.0385	0.0404	0.037
	0.9556	1.1418	0.7341	0.6699	0.7687	1.1882	0.8044	0.7631	0.6895	0.7017	0.6961	0.7177
	0.0498	0.063	0.073	0.0675	0.0503	0.0457	0.0398	0.0377	0.038	0.0409	0.0404	0.0373
	0.0487	0.0499	0.0474	0.0455	0.0485	0.0474	0.0486	0.0462	0.0476	0.0483	0.0479	0.0495
	0.0463	0.0466	0.0505	0.0468	0.048	0.049	0.0477	0.0471	0.0466	0.0481	0.0461	0.0474

21 - FSONC	0.9477	1.1252	0.7239	1.1224	0.7535	0.7477	0.7205	0.7762	0.7747	0.7044	0.6995	0.6524
	0.065	0.1065	0.076	0.0699	0.0454	0.0405	0.0481	0.0381	0.038	0.0381	0.0384	0.0367
	0.662	1.2361	0.6933	0.6842	0.7282	0.7317	0.7459	0.796	0.7222	0.683	0.6588	0.6878
	0.0884	0.0942	0.0528	0.058	0.0581	0.0501	0.0469	0.0398	0.0387	0.0385	0.0404	0.037
	0.9556	1.1418	0.7341	0.6699	0.7687	1.1882	0.8044	0.7631	0.6895	0.7017	0.6961	0.7177
	0.0498	0.063	0.073	0.0675	0.0503	0.0457	0.0398	0.0377	0.038	0.0409	0.0404	0.0373
	0.0487	0.0499	0.0474	0.0455	0.0485	0.0474	0.0486	0.0462	0.0476	0.0483	0.0479	0.0495
	0.0463	0.0466	0.0505	0.0468	0.048	0.049	0.0477	0.0471	0.0466	0.0481	0.0461	0.0474

Samples from 3/27/03

Plate Number	Description
1	E2 Standard and Blanks
2	E2 Spiked into Nano
3	< 1.5 um Sample - E2 Spiked
4	< 1.5 um Sample - E2 Spiked
5	< 1.5 um Sample - Blank
6	< 0.22 um Sample - E2 Spiked
7	< 0.22 um Sample - E2 Spiked
8	< 0.22 um Sample - Blank
9	100 kD Sample - E2 Spiked
10	100 kD Sample - E2 Spiked
11	100 kD Sample - Blank
12	30 kD Sample - E2 Spiked
13	30 kD Sample - E2 Spiked
14	30 kD Sample - Blank
15	10 kD Sample - E2 Spiked
16	10 kD Sample - E2 Spiked
17	10 kD Sample - Blank
18	1 kD Sample - E2 Spiked
19	1 kD Sample - E2 Spiked
20	1 kD Sample - Blank

Samples from 3/27/03 – Summary of E2 Dose-Response Curves and Organic Carbon Parameters

Exper	COC	Repl	Min	Max	EC50	EC50 err	Slope	Slope err	Protein	Polys	Humics	Fulvics
<1.5	3.39	1	1.0733	2.81897	141.5956	8.96505	3.40979	0.74428	2.82	0.33	1.63	2.6
<1.5	4.52	2	1.04303	3.13141	153.0522	15.94438	3.12397	0.82561	3.76	0.44	2.17	3.46
<1.5	5.085	3	1.00716	3.38397	267.7964	--	51.66962	--	4.23	0.49	2.44	3.89
<1.5	3.39	1	1.07077	2.90844	158.0963	8.56017	3.14404	0.51217	2.82	0.33	1.63	2.6
<1.5	4.52	2	1.03322	3.27091	158.5781	12.43329	3.51785	0.78298	3.76	0.44	2.17	3.46
<1.5	5.085	3	1.1179	3.34161	222.9703	1.82E+24	135.2386	3.80E+24	4.23	0.49	2.44	3.89
<0.22	1.6	1	0.96104	2.82496	162.8443	5.56304	2.67149	0.21873	1.51	0.17	0.22	0.78
<0.22	2.13	2	0.95342	3.10526	158.4627	15.93633	3.21011	0.85682	2.01	0.22	0.29	1.05
<0.22	2.4	3	0.98009	3.39984	157.9594	4.57757	5.26714	1.3349	2.27	0.25	0.33	1.18
<0.22	1.6	1	0.9648	2.88642	161.1478	10.21974	3.11899	0.58157	1.51	0.17	0.22	0.78
<0.22	2.13	2	0.93827	3.11704	150.632	10.25485	2.81725	0.44986	2.01	0.22	0.29	1.05
<0.22	2.4	3	0.99099	3.38278	163.4542	6.92769	4.93801	1.31609	2.27	0.25	0.33	1.18
<100kDa	1.88	1	0.96682	2.83012	157.1451	6.16775	3.09384	0.35765	2.37	0.15	0.68	1.14
<100kDa	2.51	2	0.95418	3.16051	149.6327	14.37343	2.37166	0.46486	3.16	0.2	0.91	1.52
<100kDa	2.82	3	1.02118	3.51306	202.9351	7.54961	2.86756	0.27332	3.56	0.23	1.02	1.71
<100kDa	1.88	1	0.99341	2.67844	150.1158	26.50747	3.81896	2.9796	2.37	0.15	0.68	1.14
<100kDa	2.51	2	0.96081	3.226	160.4405	10.85504	2.23521	0.29431	3.16	0.2	0.91	1.52
<100kDa	2.82	3	1.03625	3.33783	154.2419	8.01917	3.34807	0.58779	3.56	0.23	1.02	1.71
<30kDa	0.89	1	0.95655	2.75169	144.0698	7.42543	3.46377	0.64028	0.75	0.07	0.19	0.77
<30kDa	1.18	2	1.0058	3.26208	141.7205	8.30572	3.25271	0.47978	1	0.09	0.26	1.02
<30kDa	1.3275	3	1.49058	4.01828	102.4039	10.95785	3.13768	0.82761	1.13	0.1	0.29	1.15
<30kDa	0.89	1	0.82659	2.77389	107.5802	17.05907	1.56769	0.35278	0.75	0.07	0.19	0.77
<30kDa	1.18	2	1.07949	3.17325	124.3101	18.85519	6.82154	4.06697	1	0.09	0.26	1.02
<30kDa	1.3275	3	1.15385	3.4359	111.4292	19.65385	4.14309	2.03383	1.13	0.1	0.29	1.15
<10kDa	0.86	1	0.95291	2.68506	154.4048	6.60796	3.49184	0.5434	0.56	0.02	0.13	0.69
<10kDa	1.14	2	1.0392	3.19298	135.9987	14.12517	4.03588	1.1385	0.75	0.03	0.17	0.92
<10kDa	1.2825	3	1.04705	3.38382	165.7552	6.90782	5.25759	1.40526	0.84	0.03	0.19	1.03
<10kDa	0.86	1	0.95191	2.91904	144.4023	5.64526	3.07193	0.35239	0.56	0.02	0.13	0.69
<10kDa	1.14	2	1.05601	3.41629	124.8309	3.4127	4.25049	0.36354	0.75	0.03	0.17	0.92
<10kDa	1.2825	3	1.05031	3.3467	153.8832	7.22941	4.13213	1.0125	0.84	0.03	0.19	1.03
<1kda		1	0.97815	2.88719	159.384	7.06862	2.8844	0.3395				
<1kda		2	1.006	3.36317	143.7459	7.78847	2.77904	0.34654				
<1kda		3	1.00045	3.33394	154.2552	6.71283	4.13459	0.93657				
<1kda		1	0.98589	2.90601	145.9871	8.11498	3.01678	0.47988				
<1kda		2	1.02212	3.28447	156.6207	10.34726	3.47153	0.63534				
<1kda		3	1.00574	3.4004	151.0762	6.67489	3.61634	0.62901				

Absorbance at 540 nm – Plates 1 through 20 for 3/27/03

1	3.2041	3.2816	3.3496	3.5129	3.3664	2.4343	1.737	1.5417	0.938	1.4325	0.7853	1.3469
	3.2011	3.2788	3.341	3.4511	3.3573	2.4283	1.7267	1.5473	0.8775	0.8741	0.859	1.4428
	0.0372	0.0371	0.0373	0.0358	0.0398	0.0391	0.0377	0.0382	0.0369	0.0377	0.0366	0.0358
	3.4714	3.6987	3.9271	3.8524	2.823	2.0543	1.5869	1.5137	0.9389	0.9794	0.8059	0.884
	3.6624	4	4	4	3.0036	2.0472	1.6524	1.5262	0.9476	0.9136	0.9495	0.8361
	0.0394	0.0384	0.0416	0.0404	0.0386	0.0402	0.041	0.0396	0.0395	0.0389	0.0388	0.0397
	4	4	4	4	2.5323	1.7109	1.5399	1.5161	0.9789	1.4459	1.4644	0.9014
	0.0379	0.0384	0.0438	0.0371	0.0375	0.0374	0.0387	0.0382	0.0377	0.0392	0.0389	0.0386
2	3.2041	3.2816	3.3496	3.5129	3.3664	2.4343	1.737	1.5417	0.938	1.4325	0.7853	1.3469
	3.2011	3.2788	3.341	3.4511	3.3573	2.4283	1.7267	1.5473	0.8775	0.8741	0.859	1.4428
	0.0372	0.0371	0.0373	0.0358	0.0398	0.0391	0.0377	0.0382	0.0369	0.0377	0.0366	0.0358
	3.4714	3.6987	3.9271	3.8524	2.823	2.0543	1.5869	1.5137	0.9389	0.9794	0.8059	0.884
	3.6624	4	4	4	3.0036	2.0472	1.6524	1.5262	0.9476	0.9136	0.9495	0.8361
	0.0394	0.0384	0.0416	0.0404	0.0386	0.0402	0.041	0.0396	0.0395	0.0389	0.0388	0.0397
	4	4	4	4	2.5323	1.7109	1.5399	1.5161	0.9789	1.4459	1.4644	0.9014
	0.0379	0.0384	0.0438	0.0371	0.0375	0.0374	0.0387	0.0382	0.0377	0.0392	0.0389	0.0386

3	3.2877	3.4698	3.3774	3.472	3.359	2.4408	1.7456	1.1673	1.5002	0.8801	1.1188	1.4773
	3.129	3.3216	3.3248	3.4995	3.3002	2.4751	1.7886	1.5994	0.9888	0.9561	0.9198	0.8461
	0.1603	0.1311	0.0601	0.0441	0.0549	0.0337	0.0353	0.0358	0.0356	0.0365	0.035	0.0319
	3.5486	3.4133	3.7185	3.7914	3.0651	2.1205	1.6595	1.5745	1.5064	1.5389	0.9081	0.8787
	3.4641	3.6791	4	4	2.7957	1.9866	1.6176	1.5501	1.5417	0.9312	0.9113	0.9235
	0.1876	0.1081	0.0752	0.0534	0.0723	0.0374	0.0377	0.0397	0.0369	0.0706	0.037	0.0378
	4	4	4	3.9628	0.8593	1.7652	1.5976	1.0036	0.9384	0.8961	1.5535	0.9669
	0.2048	0.1702	0.0668	0.0587	0.0462	0.0401	0.0384	0.0382	0.0388	0.0382	0.0388	0.0379
4	3.4504	3.6091	3.4371	3.533	3.4384	2.3012	1.7644	1.0275	1.5081	1.509	0.9488	0.8105
	3.2157	3.3252	3.5494	3.5633	3.1187	2.4245	1.7509	1.5784	1.0036	1.5296	0.9372	0.8729
	0.1307	0.0884	0.0616	0.0488	0.039	0.0365	0.036	0.0493	0.0373	0.0405	0.0332	0.0328
	3.4921	3.6819	3.857	4	2.9773	1.3548	1.634	0.982	0.9734	0.914	0.9475	0.9363
	3.6448	4	4	4	3.1186	2.0125	1.7197	1.5965	0.9713	0.9352	0.9407	0.8836
	0.1482	0.0936	0.0618	0.051	0.0372	0.0387	0.0384	0.0377	0.0368	0.0369	0.0387	0.0372
	4	4	4	4	2.3365	1.823	1.6184	0.9805	1.5597	0.9512	0.9102	0.8822
	0.1526	0.0935	0.072	0.0629	0.0446	0.0419	0.04	0.0447	0.0384	0.0376	0.0378	0.0381

6	3.3363	3.8215	3.3999	3.3414	3.2505	2.3947	1.7484	1.0113	0.9611	0.9863	1.4964	1.5263
	3.4418	3.2963	3.3841	3.47	3.4087	2.3285	1.134	1.5787	1.6186	0.913	0.9351	1.4602
	0.0944	0.0715	0.0534	0.0425	0.0376	0.0367	0.0379	0.0397	0.0369	0.0375	0.035	0.0335
	3.4288	3.5341	3.7695	3.8456	2.8849	2.055	1.6462	1.5959	0.9943	1.51	0.9208	0.9013
	3.685	4	4	4	2.9996	1.9758	1.6454	1.0192	0.9506	0.9832	0.8788	0.864
	0.0939	0.0694	0.0628	0.051	0.0379	0.0377	0.0397	0.0391	0.038	0.0378	0.0377	0.0388
	4	4	4	4	2.6692	1.8661	1.0254	0.9998	0.9842	0.9492	0.9172	0.9327
	0.1099	0.0653	0.0616	0.0505	0.0419	0.0404	0.0389	0.0387	0.0386	0.0383	0.0395	0.039
7	3.4795	3.7466	3.4624	4	3.2892	2.4301	1.886	1.0178	1.5909	0.9353	0.9907	0.9553
	3.3516	3.2453	3.2712	3.3933	3.268	2.1973	1.1308	0.9694	0.9507	0.961	0.9547	0.8363
	0.0922	0.0631	0.0571	0.0418	0.0362	0.0358	0.0373	0.037	0.0366	0.0353	0.0346	0.0368
	3.3268	3.8394	3.6268	4	2.9267	2.0421	1.6251	1.0167	1.5537	0.9665	0.9604	0.9718
	3.725	3.9211	4	4	3.1289	2.0842	1.6956	1.6621	0.941	0.9199	0.9668	0.9551
	0.0895	0.0649	0.0627	0.0485	0.0416	0.0407	0.0425	0.041	0.0393	0.0397	0.0407	0.0407
	4	4	4	4	2.5329	1.8282	1.6255	0.9558	0.9896	0.9854	0.8938	0.8903
	0.1043	0.0618	0.0625	0.0483	0.0442	0.0426	0.0424	0.0392	0.0412	0.0399	0.0394	0.0417

9	3.5376	3.5164	3.301	3.4473	3.4011	2.2964	1.7277	1.5705	1.5213	0.7371	0.8156	0.7808
	3.272	3.3947	3.3984	3.3926	3.3603	2.351	1.7642	1.6023	1.4779	1.5059	0.9082	0.8268
	0.3687	0.1233	0.0993	0.0852	0.0499	0.0376	0.0389	0.0368	0.0365	0.0388	0.0351	0.0359
	3.3996	4	4	3.8659	3.0832	2.1001	1.6611	1.5939	0.9176	0.9147	0.8573	0.8679
	3.7934	4	4	2.7749	3.0989	2.0613	1.6827	0.9647	0.9181	0.9258	0.915	0.8236
	0.1927	0.137	0.0674	0.0729	0.0621	0.0493	0.0547	0.0423	0.0817	0.0382	0.0391	0.0397
	1.9821	4	4	4	2.2484	4	0.9126	0.912	0.9141	0.8897	0.8981	0.8531
	0.2823	0.1757	0.0779	0.0743	0.0561	0.0442	0.0505	0.0394	0.0394	0.0444	0.0415	0.04
10	3.4571	3.5988	3.4821	3.6925	3.4482	2.4495	1.8328	1.6059	1.5258	1.5241	0.9304	0.8555
	1.8337	3.4914	3.6242	3.4154	3.2004	2.199	1.8052	1.6221	0.9062	1.5092	0.8554	0.8781
	0.1321	0.1237	0.0577	0.0451	0.0447	0.0443	0.0413	0.0399	0.0369	0.67	0.0361	0.0358
	3.8816	4	3.8528	4	3.1442	2.0499	1.7354	1.7011	1.5361	0.8656	0.8603	0.8704
	3.9565	4	4	3.1729	2.9337	2.0129	1.8009	1.6191	0.8877	0.9589	0.9085	0.9108
	0.1748	0.1	0.1216	0.0566	0.0523	0.047	0.0389	0.0398	0.037	0.0457	0.0402	0.0382
	4	4	4	4	2.5438	1.735	1.6631	0.8167	0.8806	1.5086	0.8865	0.8965
	0.3072	0.123	0.0893	0.0578	0.0536	0.0647	0.0394	0.0399	0.0383	0.0385	0.0383	0.1003

12	3.3161	3.3408	3.4882	3.4914	3.4071	2.473	1.2322	1.6098	1.5723	0.8467	0.9361	1.5079
	3.3366	3.3164	3.3248	3.5081	3.345	2.4196	1.8081	1.5358	0.9216	0.908	0.8292	0.9558
	0.1564	0.0986	0.0666	0.0446	0.0388	0.0381	0.0357	0.0354	0.0383	0.0365	0.0335	0.0334
	3.4853	3.6639	3.7578	4	3.3325	2.0899	1.69	1.0267	0.9825	0.9727	0.97	0.953
	3.883	4	4	4	3.1708	2.1277	1.214	1.0095	0.946	0.9288	0.9385	1.0091
	0.1627	0.1018	0.0659	0.0508	0.0422	0.0397	0.0392	0.0393	0.0375	0.0422	0.0382	0.0383
	4	4	4	4	3.4682	2.1045	1.7773	1.6434	1.5527	0.9652	1.6344	1.4058
	0.1134	0.1725	0.0436	0.185	0.0428	0.0397	0.0395	0.0386	0.0384	0.0385	0.0389	0.0386
13	3.3711	3.3003	3.3687	3.429	3.2287	2.2851	1.7746	1.595	0.9632	0.9078	0.9067	1.4959
	3.3564	3.3218	3.3295	3.4223	3.2673	2.506	1.8255	0.9901	0.9948	0.8555	0.9595	0.8454
	0.1186	0.0993	0.0637	0.0445	0.0373	0.0379	0.0366	0.0362	0.0378	0.0372	0.0367	0.0385
	3.3438	3.5689	3.7382	3.9389	3.0756	2.0557	1.7293	1.0877	0.9297	1.5264	1.496	0.9407
	3.6531	3.7897	4	4	3.7359	1.4649	1.1936	1.08	1.5599	0.9395	0.9241	0.9199
	0.1473	0.0894	0.0699	0.0478	0.1228	0.0386	0.0381	0.0373	0.0367	0.0476	0.0369	0.0613
	4	4	4	4	3.4682	2.1045	1.7773	1.6434	1.5527	0.9652	1.6344	1.4058
	0.1393	0.1443	0.0704	0.0533	0.0441	0.0398	0.0384	0.0381	0.0387	0.0378	0.0392	0.0405

15	3.4306	3.4291	3.4118	3.5294	3.3391	2.434	1.1331	1.6262	0.9662	1.1314	0.781	1.5744
	3.2863	3.3182	3.4335	3.4507	3.2617	2.4236	1.7992	1.6216	1.0311	1.534	0.9447	0.7786
	0.1466	0.1017	0.068	0.0473	0.0419	0.0406	0.0384	0.0377	0.0381	0.0378	0.0362	0.0345
	3.2931	3.8808	4	4	3.2594	2.1412	1.772	1.7205	0.9232	1.0101	0.9712	0.9582
	3.5603	3.9096	4	4	3.8615	2.1053	1.176	1.0907	0.9273	0.9743	0.9501	1.6126
	0.1407	0.1148	0.0777	0.0497	0.0389	0.0405	0.0403	0.0389	0.0369	0.0414	0.0371	0.0379
	4	4	4	4	2.4553	1.8777	1.0559	0.9735	0.9779	1.5665	0.9551	0.9385
	0.194	0.1121	0.0792	0.0676	0.0385	0.0421	0.0412	0.039	0.039	0.0377	0.0382	0.0388
16	3.6211	3.6248	3.6452	3.551	3.5993	2.5319	1.7542	1.0583	1.0328	0.9769	0.9474	0.9238
	3.4222	3.5273	3.5202	3.5673	3.4374	2.563	1.8774	1.0778	1.577	1.0339	0.9294	0.8208
	0.1388	0.0873	0.0655	0.0448	0.0415	0.0621	0.0372	0.0362	0.0376	0.0528	0.0376	0.0337
	3.9069	4	4	4	3.5219	2.1853	1.7947	1.1396	0.9955	1.0101	0.9654	0.9359
	4	4	4	4	4	2.1742	1.1857	1.1121	0.9713	1.709	1.0073	0.9479
	0.1627	0.0894	0.067	0.0518	0.0392	0.0396	0.0424	0.0376	0.0369	0.0378	0.0382	0.0385
	4	4	4	4	2.6318	1.9702	1.7533	0.9377	0.9775	0.9593	0.9323	0.9181
	0.1353	0.1058	0.0588	0.0508	0.0418	0.0393	0.0382	0.0387	0.0391	0.0374	0.0494	0.0389

18	3.5942	3.5123	3.5253	3.5919	3.2136	2.4116	1.8003	1.6077	0.9561	1.5019	0.8849	0.7859
	3.4933	3.5337	3.5115	3.6009	3.4015	2.4355	1.7933	1.0482	0.9533	1.0147	1.0107	1.0649
	0.1076	0.0996	0.0564	0.022	0.0384	0.0375	0.0379	0.0389	0.0378	0.0376	0.0368	0.0347
	4	4	4	4	3.0085	2.2992	1.1724	1.0828	1.063	1.0167	0.962	0.8934
	4	4	4	4	3.3893	2.2087	1.7494	1.6172	0.9841	1.02	0.998	0.9769
	0.1125	0.1218	0.0557	0.048	0.0369	0.0379	0.0378	0.0374	0.0359	0.0364	0.0367	0.0379
	4	4	4	4	2.6606	1.8466	1.6073	1.6604	1.0203	0.939	1.0203	0.9312
	0.1154	0.073	0.0556	0.0435	0.0381	0.0387	0.0384	0.0399	0.038	0.037	0.0376	0.0431
19	3.5171	3.4842	3.5636	3.6556	3.4229	2.496	1.8609	1.0176	0.9615	1.5018	1.5302	0.8903
	3.5727	3.3549	3.4727	3.5799	3.4049	2.5458	1.845	1.6246	1.024	0.9952	1.0177	0.9839
	0.1202	0.1066	0.0512	0.0412	0.0357	0.05	0.0348	0.0418	0.0376	0.0369	0.0351	0.0338
	4	3.8277	4	4	3.0474	1.3059	1.1479	1.5665	1.0065	0.9918	0.9471	0.9707
	4	4	4	4	3.248	2.1093	1.6945	1.579	1.0108	0.9699	0.9657	0.9365
	0.1256	0.0867	0.0546	0.0472	0.045	0.0369	0.0373	0.0369	0.0367	0.0362	0.0376	0.0382
	4	4	4	4	2.6986	1.8645	1.6179	0.9962	0.9582	0.9773	1.0026	0.9335
	0.107	0.0766	0.0515	0.0437	0.0426	0.0396	0.0421	0.0377	0.0375	0.0371	0.037	0.0383

Absorbance at 620 nm – Plates 1 through 20 for 3/27/03

1	1.3518	1.2967	1.2697	1.3448	1.3233	1.2616	1.2531	1.2319	0.7307	1.1983	0.6265	1.1284
	1.3375	1.3606	1.2902	1.3228	1.3463	1.2796	1.2159	1.237	0.6826	0.6615	0.6806	1.221
	0.0349	0.036	0.0363	0.0346	0.038	0.037	0.0367	0.0371	0.0342	0.0361	0.0355	0.0344
	1.3332	1.335	1.3119	1.3219	1.3001	1.2887	1.2066	1.2299	0.7455	0.7932	0.6432	0.7192
	1.3215	1.3173	1.3405	1.3374	1.3539	1.2948	1.29	1.2396	0.7516	0.7367	0.7739	0.6768
	0.037	0.0369	0.0397	0.0385	0.0366	0.0387	0.0396	0.0384	0.0388	0.0378	0.0376	0.0386
	1.3152	1.3335	1.3311	1.318	1.2955	1.2367	1.2294	1.261	0.7886	1.2062	1.2306	0.736
	0.0365	0.037	0.0417	0.0362	0.0363	0.036	0.0374	0.0367	0.0363	0.0374	0.0376	0.0377
2	1.3518	1.2967	1.2697	1.3448	1.3233	1.2616	1.2531	1.2319	0.7307	1.1983	0.6265	1.1284
	1.3375	1.3606	1.2902	1.3228	1.3463	1.2796	1.2159	1.237	0.6826	0.6615	0.6806	1.221
	0.0349	0.036	0.0363	0.0346	0.038	0.037	0.0367	0.0371	0.0342	0.0361	0.0355	0.0344
	1.3332	1.335	1.3119	1.3219	1.3001	1.2887	1.2066	1.2299	0.7455	0.7932	0.6432	0.7192
	1.3215	1.3173	1.3405	1.3374	1.3539	1.2948	1.29	1.2396	0.7516	0.7367	0.7739	0.6768
	0.037	0.0369	0.0397	0.0385	0.0366	0.0387	0.0396	0.0384	0.0388	0.0378	0.0376	0.0386
	1.3152	1.3335	1.3311	1.318	1.2955	1.2367	1.2294	1.261	0.7886	1.2062	1.2306	0.736
	0.0365	0.037	0.0417	0.0362	0.0363	0.036	0.0374	0.0367	0.0363	0.0374	0.0376	0.0377

3	1.4178	1.4509	1.4085	1.3864	1.3711	1.3247	1.2756	0.8995	1.2273	0.6957	0.9308	1.2472
	1.3762	1.3976	1.3969	1.4146	1.4139	1.3582	1.3011	1.2888	0.7757	0.7658	0.7399	0.6753
	0.1473	0.1541	0.0582	0.0425	0.0509	0.0318	0.0348	0.0346	0.0339	0.0353	0.0333	0.0311
	1.4064	1.3678	1.3813	1.4105	1.4105	1.3393	1.3023	1.3046	1.2743	1.3075	0.7368	0.7077
	1.4047	1.4039	1.3893	1.3912	1.3615	1.3238	1.294	1.2787	1.295	0.7544	0.7335	0.7535
	0.1421	0.1187	0.0755	0.0497	0.0769	0.0362	0.0364	0.0381	0.0356	0.0623	0.0361	0.0367
	1.3631	1.3936	1.3882	1.3539	0.6917	1.2908	1.3016	0.7848	0.7434	0.7177	1.3196	0.7985
	0.1735	0.1635	0.0596	0.0575	0.043	0.038	0.0368	0.0367	0.0371	0.0367	0.037	0.037
4	1.4157	1.5022	1.4077	1.3976	1.4131	1.3505	1.3053	0.7788	1.2518	1.2817	0.7707	0.6451
	1.3651	1.3756	1.4101	1.4081	1.3809	1.3644	1.274	1.2736	0.7887	1.2945	0.7635	0.7046
	0.1444	0.0961	0.0559	0.0483	0.0367	0.035	0.0358	0.0474	0.0353	0.0387	0.0324	0.0317
	1.3992	1.4013	1.4008	1.4333	1.3746	0.8135	1.2767	0.7538	0.7749	0.7266	0.7746	0.7639
	1.3574	1.3875	1.3812	1.3617	1.3704	1.3157	1.3769	1.334	0.7715	0.7546	0.7646	0.7192
	0.1477	0.1066	0.0597	0.0484	0.0351	0.0373	0.0371	0.0366	0.0358	0.036	0.0373	0.0362
	1.4779	1.3885	1.4058	1.3779	1.3124	1.3087	1.3103	0.7668	1.3061	0.7629	0.7416	0.7336
	0.133	0.0957	0.0734	0.0616	0.0421	0.0399	0.0378	0.0418	0.037	0.0362	0.0366	0.0372

6	1.3584	1.4998	1.3755	1.3399	1.4097	1.3644	1.2494	0.94	0.7571	0.7995	1.2622	1.3021
	1.4151	1.3583	1.3347	1.4149	1.7184	1.3469	0.7427	1.2845	1.3619	0.7278	0.7542	1.2438
	0.0885	0.0698	0.0506	0.0401	0.0365	0.0347	0.0366	0.0377	0.0357	0.0362	0.0346	0.0326
	1.6672	1.3815	1.3905	1.4338	1.3932	1.3917	1.2784	1.3118	0.8035	1.2726	0.7511	0.7299
	1.35	1.3598	1.4109	1.3773	1.3707	1.299	1.2799	0.7901	0.7605	0.794	0.7063	0.7006
	0.0815	0.0676	0.0594	0.0473	0.036	0.0362	0.038	0.0375	0.0363	0.0367	0.0365	0.038
	1.3599	1.3807	1.4187	1.3522	1.4462	1.4439	0.7818	0.7939	0.7925	0.762	0.74	0.7547
	0.0958	0.0649	0.0577	0.0491	0.0401	0.0389	0.0376	0.0376	0.0372	0.0371	0.0382	0.0375
7	1.3671	1.4203	1.2898	1.4405	1.3408	1.2901	1.3881	0.7675	1.3376	0.7582	0.8095	0.7887
	1.4088	1.3961	1.3048	1.3425	1.4541	1.3127	0.7448	0.7306	0.7477	0.7739	0.777	0.6725
	0.085	0.0622	0.053	0.0421	0.0343	0.035	0.0363	0.0369	0.0352	0.0339	0.0338	0.0355
	1.3422	1.4594	1.3478	1.7808	1.3694	1.3019	1.2452	0.7928	1.3099	0.7872	0.8019	0.7952
	1.3476	1.4101	1.4646	1.4149	1.4341	1.3565	1.3274	1.3797	0.7571	0.7496	0.7943	0.7878
	0.0778	0.0612	0.0585	0.046	0.0373	0.0386	0.0402	0.0395	0.0382	0.0382	0.0413	0.0398
	1.3833	1.4224	1.3992	1.4086	1.4062	1.3555	1.2957	0.7513	0.8057	0.8067	0.7312	0.7306
	0.0896	0.0568	0.0584	0.0462	0.0422	0.0405	0.0412	0.0374	0.0393	0.0389	0.0383	0.04

9	1.3441	1.401	1.0755	1.3811	1.389	1.2814	1.2782	1.2897	1.2515	0.5463	0.6405	0.6092
	1.3754	1.4034	1.3699	1.3361	1.519	1.3093	1.2862	1.3009	1.2345	1.2697	0.7235	0.6579
	0.3918	0.1209	0.0905	0.0705	0.0449	0.0355	0.0379	0.0356	0.0347	0.0369	0.0339	0.0343
	1.4432	1.6088	1.4931	1.3884	1.4057	1.3352	1.303	1.3178	0.7196	0.7338	0.6867	0.7002
	1.37	1.4065	1.3755	1.2106	1.3343	1.3384	1.3378	0.7344	0.7205	0.7407	0.7338	0.66
	0.1693	0.1439	0.0629	0.0664	0.0557	0.0458	0.0509	0.0408	0.0745	0.0374	0.038	0.0387
	1.1904	1.4495	1.4357	1.9249	1.3427	1.4466	0.6699	0.7147	0.7286	0.7021	0.7201	0.6826
	0.2539	0.1636	0.0703	0.0654	0.0417	0.0422	0.0476	0.0375	0.0365	0.0407	0.0398	0.0384
10	1.4255	1.4305	1.4012	1.4445	1.4046	1.342	1.3047	1.2596	1.2559	1.2753	0.745	0.6813
	1.2955	1.4912	1.4393	1.32	1.3978	1.27	1.3316	1.3209	0.6929	1.263	0.6792	0.7087
	0.1195	0.1208	0.053	0.0437	0.0412	0.0418	0.039	0.0376	0.0436	0.0353	0.035	0.035
	1.5784	1.4875	1.3726	1.7166	1.3647	1.3103	1.3805	1.4198	1.2922	0.6864	0.6907	0.7011
	1.557	1.472	1.4565	1.3518	1.3584	1.3431	1.4287	1.351	0.6935	0.7741	0.7289	0.74
	0.1538	0.0904	0.112	0.2	0.046	0.0435	0.0376	0.0382	0.0363	0.0435	0.0385	0.0365
	1.339	1.685	1.5248	1.6668	1.29	1.3027	1.3818	0.6243	0.6945	1.2777	0.7184	0.7385
	0.2696	0.1103	0.0883	0.0554	0.0493	0.0596	0.0379	0.0383	0.0369	0.037	0.0366	0.0898

12	1.3302	1.3701	1.3864	1.3984	1.3606	1.3223	0.7671	1.2688	1.2971	0.6384	0.7326	1.2574
	1.4073	1.3126	1.3782	1.3811	1.3624	1.3054	1.2733	1.2437	0.6965	0.7014	0.6419	0.7659
	0.1526	0.1127	0.0652	0.0422	0.0363	0.0362	0.0344	0.034	0.0358	0.0349	0.0325	0.0325
	1.3332	1.3616	1.452	1.4319	1.4345	1.3051	1.2942	0.7758	0.7625	0.7691	0.7836	0.7616
	1.3736	1.3532	1.3799	1.3415	1.3623	1.375	0.8564	0.7561	0.7313	0.728	0.7431	0.8188
	0.1471	0.106	0.0654	0.0475	0.0389	0.0385	0.0378	0.0398	0.0368	0.0404	0.0367	0.0366
	0.7794	0.812	0.778	0.8228	0.7964	0.815	0.7765	0.7824	0.7615	0.7129	0.7699	0.7901
	0.0995	0.1619	0.041	0.2001	0.0411	0.0388	0.0381	0.037	0.037	0.0372	0.0371	0.0372
13	1.3712	1.3779	1.3626	1.3616	1.3245	1.2598	1.2723	0.7285	0.6982	0.7069	1.249	1.2574
	1.3493	1.3213	1.3429	1.3485	1.3222	1.2997	0.7093	0.7629	0.6536	0.7632	0.6549	0.7659
	0.1082	0.0564	0.0408	0.0354	0.0355	0.035	0.0354	0.0365	0.0358	0.0355	0.037	0.0325
	1.32	1.3867	1.3895	1.3787	1.3026	1.3587	0.8337	0.7182	1.2783	1.26	0.7564	0.7616
	1.3276	1.347	1.3659	1.4109	0.8668	0.8525	0.8112	1.2905	0.7394	0.7288	0.7342	0.8188
	0.1166	0.068	0.0442	0.1172	0.0371	0.0368	0.036	0.0357	0.0464	0.036	0.0603	0.0366
	1.3618	1.3347	1.3852	1.3675	1.3491	1.3889	1.3518	1.2814	0.7565	1.3766	1.2021	0.7901
	0.1517	0.0668	0.0534	0.0415	0.0383	0.0373	0.0364	0.0372	0.0367	0.0371	0.0387	0.0372

15	1.4684	1.5062	1.4449	1.4673	1.4582	1.4046	0.7715	1.309	0.7397	0.8454	0.5997	1.3422
	1.4103	1.4265	1.4353	1.3971	1.4492	1.4001	1.3238	1.3145	0.8085	1.283	0.7609	0.6029
	0.1545	0.1144	0.0648	0.0448	0.0395	0.0381	0.0372	0.036	0.0367	0.0366	0.0348	0.0335
	1.5054	1.4704	1.4362	1.4225	1.4849	1.4428	1.4155	1.4292	0.7183	0.8076	0.7812	0.769
	1.4089	1.4118	1.449	1.4507	1.6947	1.3996	0.8292	0.8302	0.719	0.7755	0.7552	1.3668
	0.1556	0.1342	0.0756	0.0465	0.0364	0.0385	0.0387	0.0379	0.0357	0.0402	0.036	0.0361
	1.3914	1.4705	1.451	1.4494	1.3837	1.3987	0.7941	0.7558	0.7716	1.316	0.7706	0.762
	0.1583	0.1318	0.0677	0.0691	0.0368	0.0404	0.0398	0.0373	0.0376	0.0365	0.0371	0.0371
16	1.3575	1.3873	1.3767	1.3551	1.4069	1.3597	1.259	0.7471	0.7899	0.757	0.7308	0.7188
	1.3445	1.3712	1.4053	1.3971	1.3887	1.3294	1.283	0.7893	1.2942	0.8152	0.7194	0.6213
	0.1328	0.0922	0.0637	0.0431	0.0394	0.0574	0.0363	0.0353	0.0356	0.0495	0.0349	0.0325
	1.384	1.3732	1.4132	1.4657	1.45	1.3241	1.3385	0.8647	0.7719	0.787	0.7568	0.7294
	1.3735	1.3447	1.3777	1.3568	1.3881	1.2996	0.8069	0.845	0.7575	1.4327	0.7915	0.7534
	0.1634	0.0914	0.0665	0.0492	0.037	0.0379	0.0402	0.0363	0.0364	0.0369	0.037	0.0379
	1.4668	1.459	1.5674	1.4396	1.3755	1.442	1.4176	0.7106	0.7579	0.7534	0.732	0.7382
	0.1351	0.1092	0.0567	0.0493	0.0395	0.0382	0.0372	0.0372	0.0379	0.0363	0.0466	0.0378

18	1.4218	1.4001	1.3906	1.3925	1.3458	1.3422	1.2718	1.2694	0.7146	1.2253	0.6729	0.7004
	1.4373	1.4112	1.4185	1.4118	1.4135	1.4204	1.2881	0.7656	0.7178	0.7916	0.7974	0.7839
	0.1015	0.1027	0.0554	0.0406	0.0367	0.0357	0.0367	0.0368	0.0364	0.0364	0.036	0.0334
	1.7665	1.3769	1.3848	1.4225	1.3538	1.4326	0.8082	0.813	0.8317	0.8007	0.751	0.7682
	1.3822	1.3893	1.3941	1.3755	1.3625	1.3473	1.3114	1.2874	0.7565	0.7988	0.7816	0.7391
	0.0988	0.1464	0.0522	0.0446	0.0349	0.0368	0.0366	0.0415	0.0353	0.0357	0.8098	0.0375
	1.4413	1.4458	1.4841	1.4233	1.3887	1.3297	1.2754	1.379	0.805	0.779	0.0364	0.7324
	0.1099	0.0694	0.0523	0.0424	0.0362	0.0374	0.0367	0.0384	0.0363	0.0361	0.0361	0.0372
19	1.3836	1.3726	1.3423	1.3475	1.3383	1.3144	1.3086	0.7381	0.7357	1.2247	1.2608	0.7004
	1.3892	1.4005	1.3502	1.3858	1.4085	1.3826	1.2933	1.2688	0.7849	0.7722	0.8018	0.7839
	0.1184	0.0992	0.0466	0.0409	0.0343	0.0471	0.0338	0.0401	0.0364	0.0355	0.0334	0.0334
	2.025	1.3782	1.3614	1.3931	1.3677	0.7201	0.8001	1.2669	0.781	0.7817	0.7385	0.7682
	1.4125	1.4064	1.4523	1.4151	1.4028	1.382	1.2846	1.258	0.7888	0.7539	0.7549	0.7391
	0.1113	0.0872	0.0525	0.0419	0.0411	0.0353	0.0359	0.0358	0.0362	0.0356	0.0365	0.0375
	1.377	1.3703	1.4337	1.3907	1.3559	1.3069	1.2642	0.7518	0.7385	0.7599	0.7928	0.7324
	0.0944	0.0767	0.0495	0.0427	0.0399	0.038	0.0414	0.0365	0.0362	0.0363	0.036	0.0372

Absorbance at 620 nm for Blanks – Plates 0 through 20 from 3/27/03

0	0.6882	0.7002	0.7313	0.7135	0.7412	0.7568	0.8188	0.7751	0.7492	0.7034	0.6876	0.5937
	0.6538	0.6962	0.7055	0.7201	0.8229	0.8404	0.8541	0.8541	0.8938	0.9712	0.917	0.6347
	0.6538	0.6962	0.7055	0.7201	0.8229	0.8404	0.8541	0.8541	0.8938	0.9712	0.917	0.6347
	0.6882	0.7002	0.7313	0.7135	0.7412	0.7568	0.8188	0.7751	0.7492	0.7034	0.6876	0.5937
	0.6538	0.6962	0.7055	0.7201	0.8229	0.8404	0.8541	0.8541	0.8938	0.9712	0.917	0.6347
	0.6538	0.6962	0.7055	0.7201	0.8229	0.8404	0.8541	0.8541	0.8938	0.9712	0.917	0.6347
	0.6882	0.7002	0.7313	0.7135	0.7412	0.7568	0.8188	0.7751	0.7492	0.7034	0.6876	0.5937
	0.6882	0.7002	0.7313	0.7135	0.7412	0.7568	0.8188	0.7751	0.7492	0.7034	0.6876	0.5937
5	0.8759	0.8959	0.786	0.927	0.8302	0.8323	0.8084	0.8577	0.8356	0.8628	0.9359	0.7057
	0.3299	0.2615	0.0477	0.1194	0.038	0.0387	0.0372	0.037	0.0384	0.0368	0.0373	0.0371
	0.9384	0.8697	0.8569	0.817	0.7939	0.8139	0.8014	0.7382	0.751	0.8059	0.8123	0.7991
	0.0638	0.0401	0.039	0.0374	0.0385	0.0376	0.0367	0.0372	0.0374	0.0388	0.0406	0.0385
	0.8742	0.7526	0.7754	0.7777	0.7569	0.8351	0.7736	0.7085	0.7117	0.7255	0.7347	0.7613
	0.1504	0.0497	0.0447	0.0373	0.038	0.0392	0.0385	0.0386	0.0379	0.0391	0.0371	0.0389
	0.0476	0.0519	0.0486	0.0469	0.0494	0.0477	0.0492	0.0468	0.0465	0.0481	0.047	0.0537
	0.19	0.1228	0.0512	0.0484	0.0479	0.0496	0.0482	0.0487	0.048	0.0484	0.0463	0.0475

8	0.6617	0.6417	0.6935	0.7352	0.7768	0.7649	0.8514	0.8155	0.7982	0.7914	0.7921	0.7777
	0.0973	0.0975	0.0425	0.0399	0.0367	0.0371	0.0368	0.0354	0.0381	0.0376	0.0362	0.0475
	0.6574	0.6245	0.7716	0.759	0.7604	0.7578	0.8365	0.7268	0.7382	0.7467	0.691	0.7083
	0.0565	0.0411	0.0373	0.0378	0.0383	0.0375	0.0396	0.0369	0.0367	0.0374	0.0368	0.0378
	0.7358	0.7405	0.7458	0.7126	0.7707	0.792	0.7872	0.7923	0.7794	0.7729	0.7366	0.7424
	0.12	0.1072	0.0667	0.0368	0.0368	0.0396	0.0382	0.0388	0.0378	0.0394	0.0365	0.0391
	0.0834	0.0515	0.0477	0.0462	0.0485	0.047	0.0487	0.0459	0.0461	0.0485	0.0468	0.0466
	0.0465	0.0463	0.0507	0.0477	0.0476	0.0491	0.0476	0.0477	0.047	0.0502	0.047	0.0477
11	0.9659	0.9765	0.6863	0.8754	0.6981	0.684	0.6869	0.7043	0.6709	0.6676	0.5807	0.6893
	0.3297	0.1998	0.1279	0.0691	0.051	0.0394	0.045	0.0382	0.036	0.0367	0.0365	0.0419
	0.9849	0.9443	0.8348	0.6619	0.7049	0.7015	0.7742	0.7379	0.6985	0.7362	0.6882	0.7474
	0.1764	0.1563	0.0898	0.0747	0.0514	0.0471	0.0411	0.0404	0.0395	0.0386	0.0393	0.0378
	1.0502	1.216	0.8957	0.7867	0.8157	0.7989	0.7815	0.8511	0.7148	0.6127	0.7389	0.7228
	0.0859	0.1203	0.0858	0.0689	0.0794	0.046	0.0489	0.0404	0.0389	0.039	0.0381	0.0387
	0.0506	0.0479	0.0476	0.0468	0.0476	0.0489	0.0487	0.0485	0.0499	0.0498	0.0686	0.0469
	0.0455	0.047	0.0467	0.0462	0.0486	0.0467	0.0496	0.0505	0.0476	0.0506	0.0497	0.0477

14	0.7545	0.717	0.7457	0.8227	0.6899	0.6843	0.646	0.6786	0.7188	0.6239	0.7167	0.7221
	0.3837	0.0749	0.1876	0.0389	0.0371	0.0372	0.0372	0.0355	0.0368	0.04	0.0366	0.0377
	0.7412	0.722	0.7717	0.7179	0.8289	0.9084	0.8175	0.7728	0.7651	0.8042	0.7509	0.7805
	0.1077	0.1866	0.0381	0.0384	0.0384	0.0376	0.0372	0.0367	0.0372	0.0382	0.0367	0.0372
	0.7019	0.708	0.7757	0.7462	0.8156	0.8012	0.8161	0.8398	0.8461	0.7766	0.8284	0.8067
	0.0365	0.1932	0.0375	0.0381	0.037	0.0396	0.0389	0.0405	0.0358	0.0397	0.0443	0.0379
	0.0486	0.055	0.0485	0.0469	0.0532	0.0479	0.049	0.0473	0.0569	0.0478	0.047	0.0469
	0.0467	0.0482	0.0502	0.0489	0.0496	0.0499	0.0483	0.0486	0.0487	0.0489	0.046	0.0469
17	0.6597	0.7	0.7408	0.8371	0.6603	0.7537	0.706	0.7188	0.7025	0.6916	0.6728	0.6754
	0.2839	0.0775	0.1647	0.0379	0.0791	0.0371	0.0378	0.0355	0.0356	0.0363	0.0361	0.0438
	0.7489	0.7801	0.7478	0.8161	0.8051	0.869	0.8529	0.8431	0.8669	0.859	0.7755	0.7029
	0.1938	0.2112	0.1584	0.1131	0.0389	0.0385	0.0389	0.0398	0.0376	0.0367	0.0378	0.0384
	0.7425	0.7556	0.7911	0.7734	0.8287	0.8215	0.8189	0.8304	0.8039	0.7971	0.8445	0.7968
	0.1778	0.1054	0.0381	0.0367	0.0368	0.0396	0.0377	0.0385	0.0358	0.0467	0.0366	0.0367
	0.0477	0.0522	0.0482	0.0465	0.0493	0.0492	0.0483	0.0465	0.0465	0.0475	0.0461	0.0465
	0.0489	0.0469	0.0499	0.048	0.0484	0.0497	0.0491	0.0476	0.0473	0.0484	0.0629	0.0468

20	0.6269	0.7257	0.7639	0.798	0.7877	0.775	0.7966	0.8205	0.7402	0.6687	0.6616	0.6616
	0.5471	0.0896	0.1198	0.1049	0.053	0.0373	0.0362	0.0627	0.0366	0.0363	0.0367	0.0367
	0.8983	0.7819	0.774	0.7467	0.8047	0.7311	0.7285	0.8138	0.778	0.8268	0.693	0.693
	0.6512	0.0716	0.053	0.0641	0.0378	0.0371	0.0387	0.0366	0.0363	0.8158	0.0368	0.0368
	1.0105	0.8271	0.8371	0.8126	0.8119	0.8561	0.8179	0.7995	0.793	0.0364	0.7077	0.7077
	0.5875	0.2137	0.0592	0.0721	0.3116	0.0372	0.037	0.0363	0.0356	0.0457	0.0358	0.0358
	0.0492	0.0468	0.0459	0.0455	0.0458	0.0466	0.0459	0.048	0.051	0.0459	0.0451	0.0451
	0.0468	0.0475	0.0463	0.0471	0.0459	0.0458	0.047	0.0466	0.0496	0.0496	0.0454	0.0454

Samples from 4/03/03

Plate Number	Description
1	E2 Standard Curve
3	1 kD Sample - E2 Spiked
4	1 kD Sample - E2 Spiked
5	1 kD Sample - Blank
6	10 kD Sample - E2 Spiked
7	10 kD Sample - E2 Spiked
8	10 kD Sample - Blank
9	30 kD Sample - E2 Spiked
10	30 kD Sample - E2 Spiked
11	30 kD Sample - Blank
12	100 kD Sample - E2 Spiked
13	100 kD Sample - E2 Spiked
14	100 kD Sample - Blank
15	< 0.22 um Sample - E2 Spiked
16	< 0.22 um Sample - E2 Spiked
17	< 0.22 um Sample - Blank
18	< 1.5 um Sample - E2 Spiked
19	< 1.5 um Sample - E2 Spiked
20	< 1.5 um Sample - Blank

Samples from 4/13/03 – Summary of E2 Dose-Response Curves and Organic Carbon Parameters

Exper	COC	Repl	Min	Max	EC50	EC50 err	Slope	Slope err	Protein	Polys	Humics	Fulvics
<1.5	1.1	1	1.03293	2.7167	159.4012	11.56562	2.63905	0.45447	0.78	0.1	0.4	0.066
<1.5	1.1	1	1.02427	2.76936	156.9004	10.16921	3.09152	0.58989	0.78	0.1	0.4	0.066
<1.5	1.7	2	1.04989	3.09664	183.7752	17.12326	3.89931	1.54981	1.2	0.15	0.6	0.1
<1.5	1.7	2	1.02859	3.16856	168.1958	16.03365	3.11117	0.80831	1.2	0.15	0.6	0.1
<1.5	2	3	0.97729	3.46794	166.7454	9.16209	6.83729	3.01298	1.4	0.18	0.72	0.12
<1.5	2	3	1.00665	3.50005	166.6087	11.03997	7.17009	3.93612	1.4	0.18	0.72	0.12
<0.22	0.86	1	1.04389	2.80013	167.2859	10.50509	3.10321	0.55374	0.63	0.033	0.2	0.066
<0.22	0.86	1	1.0257	2.80268	170.8081	11.34355	2.98341	0.52877	0.63	0.033	0.2	0.066
<0.22	1.3	2	1.04035	3.19376	176.2958	16.47916	3.15407	0.85197	0.95	0.05	0.3	0.1
<0.22	1.3	2	1.01212	3.21306	179.2	15.53862	2.73961	0.58232	0.95	0.05	0.3	0.1
<0.22	1.56	3	1.04966	3.57542	167.5172	8.58877	6.04944	2.18406	1.14	0.06	0.4	0.12
<0.22	1.56	3	1.04993	3.54938	166.3862	13.12029	7.33141	4.89484	1.14	0.06	0.4	0.12
<100kDa	0.83	1	1.04746	2.78315	165.5262	11.15341	2.85954	0.498	0.66	0	0.165	0.066
<100kDa	0.83	1	1.05303	2.91574	160.1537	9.53759	3.01346	0.50442	0.66	0	0.165	0.066
<100kDa	1.25	2	1.02055	3.08904	173.6117	14.81284	3.18658	0.78445	1	0	0.25	0.1
<100kDa	1.25	2	1.20851	3.54843	153.466	28.58068	2.48097	0.98281	1	0	0.25	0.1
<100kDa	1.5	3	1.02966	3.58977	198.7285	8.01762	3.51015	0.3747	1.2	0	0.3	0.12
<100kDa	1.5	3	1.05403	3.57183	167.8632	24.8845	8.6853	10.56695	1.2	0	0.3	0.12
<30kDa	0.79	1	1.00664	2.68898	154.5668	9.98438	3.36644	0.73998	0.66	0.03	1.58	0.066
<30kDa	0.79	1	0.98162	2.72146	143.064	9.52516	3.07313	0.59901	0.66	0.03	1.58	0.066
<30kDa	1.2	2	1.02716	3.13754	157.9972	15.83703	3.13423	0.81678	1	0.05	2.4	0.1
<30kDa	1.2	2	1.01137	3.14624	170.8839	13.53054	2.94295	0.60575	1	0.05	2.4	0.1
<30kDa	1.44	3	1.05154	3.5344	166.7263	8.81823	6.19363	2.44347	1.2	0.06	2.9	0.12
<30kDa	1.44	3	1.00957	3.50414	169.4341	3.48148	3.94959	0.31215	1.2	0.06	2.9	0.12

<10kDa	0.73	1	0.98551	2.70267	133.7701	10.57195	3.03677	0.66598	0.56	0	0.1	0.066
<10kDa	0.73	1	0.97899	2.74201	134.0382	9.13757	3.09469	0.60081	0.56	0	0.1	0.066
<10kDa	1.1	2	1.02586	3.22644	142.3178	8.76767	3.09902	0.46928	0.85	0	0.15	0.1
<10kDa	1.1	2	1.02497	3.21696	151.8317	13.5215	3.4077	0.80609	0.85	0	0.15	0.1
<10kDa	1.32	3	1.04341	3.44662	166.3057	7.94682	5.70126	1.90054	1	0	0.18	0.12
<10kDa	1.32	3	1.05527	3.55414	158.9026	7.23616	4.29941	1.04591	1	0	0.18	0.12
<1kda	0.1	1	0.93144	2.68337	136.4048	9.75915	2.94337	0.56787				
<1kda	0.1	1	0.93501	2.73251	146.9186	10.14919	3.10777	0.64369				
<1kda	0.1	2	1.0314	3.32009	159.8284	13.31065	2.98582	0.62695				
<1kda	0.1	2	1.04668	3.37107	172.4879	11.39411	3.066	0.55503				
<1kda	0.1	3	1.06203	3.563	166.0505	7.86028	5.61727	1.84187				
<1kda	0.1	3	1.05169	3.55353	165.2979	8.24265	5.74046	2.10427				

Absorbance at 540 nm – Plates 1 through 20 from 4/03/03

3	3.085	3.1758	3.2996	3.308	2.9264	2.1684	1.6842	1.4892	1.393	1.4057	0.957	0.9828
	3.0275	3.0623	3.2018	3.2264	2.8433	2.1175	1.6913	1.5089	1.0109	0.9913	0.9756	0.975
	0.2141	0.0894	0.0564	0.0439	0.0456	0.0368	0.0384	0.0373	0.0363	0.0367	0.0387	0.0334
	3.0934	3.3977	3.5678	3.3819	2.6446	1.2379	1.548	0.9932	1.3954	0.957	1.0429	0.9339
	3.263	3.8148	4	3.9357	2.5409	1.7984	1.5374	1.4382	1.4045	0.9596	1.4079	0.9564
	0.1745	0.0896	0.058	0.0446	0.0376	0.0371	0.0393	0.0385	0.0369	0.0374	0.0365	0.0366
	4	4	4	4	2.2483	1.651	0.9695	0.9465	1.0047	1.0005	0.9969	0.9825
	0.1739	0.083	0.0557	0.0452	0.0398	0.0368	0.0366	0.038	0.0402	0.0382	0.0368	0.0377
4	3.1833	3.2405	3.2782	3.3549	3.1565	2.1478	1.6661	0.955	0.9518	0.8853	0.9015	0.9192
	3.1075	3.1357	3.2818	3.2778	2.9569	2.1691	1.6364	1.035	1.413	1.395	0.893	0.9349
	0.17	0.0836	0.0577	0.0417	0.041	0.038	0.0363	0.0411	0.0371	0.0367	0.0367	0.0367
	3.2477	3.4667	3.6199	3.5207	2.6609	1.855	1.5556	1.4809	0.9161	0.9208	0.8806	0.8897
	3.4785	3.6872	4	3.8163	2.7765	1.8986	1.5419	1.007	1.4399	0.9373	0.9021	0.9075
	0.1263	0.0825	0.0858	0.0443	0.0418	0.0412	0.0437	0.0392	0.0391	0.0379	0.0372	0.0391
	4	4	4	4	2.2129	1.6048	1.4957	1.4464	0.9372	1.4104	0.9577	0.9581
	0.0958	0.0767	0.0519	0.0407	0.0387	0.0381	0.0388	0.038	0.0378	0.0394	0.039	0.0394

6	3.1294	3.0828	3.3188	3.3741	3.057	2.1588	1.6423	1.4776	1.0124	1.0021	0.9523	0.9739
	3.1815	3.2717	3.2704	3.3292	2.9796	2.0077	1.5944	1.4852	1.427	0.9339	1.3699	0.9237
	0.1218	0.1231	0.068	0.049	0.0374	0.0373	0.0367	0.0382	0.0377	0.0363	0.04	0.0323
	3.217	3.4873	3.6943	3.4966	2.6034	1.8248	1.5172	1.4305	1.0008	1.4182	0.9465	0.8003
	3.5092	3.7097	4	3.7773	2.5976	1.7794	1.5055	1.4591	0.9436	0.9748	1.4324	0.9434
	0.1655	0.0912	0.0718	0.0525	0.0384	0.0388	0.0404	0.038	0.0368	0.0376	0.0366	0.0375
	4	4	4	4	2.2847	1.6392	1.5003	1.4215	0.9637	0.9947	0.9186	0.9071
	0.1618	0.0804	0.0626	0.0443	0.0426	0.0375	0.0368	0.0383	0.0365	0.0397	0.0376	0.0398
7	3.1427	3.2385	3.4019	3.3323	3.0003	2.1074	1.6111	1.4739	0.9823	0.9707	0.9576	1.0014
	3.1109	3.2346	3.2406	3.3944	2.9426	2.0992	1.1132	1.4983	0.9495	1.4126	0.9382	0.9523
	0.1476	0.0953	0.0563	0.0445	0.0388	0.0397	0.0375	0.0374	0.0379	0.0363	0.0397	0.0358
	3.3046	3.4949	3.5654	3.3891	2.6279	1.8266	1.5261	0.9994	0.9668	0.9745	0.9388	0.8348
	3.5201	4	4	3.6405	2.509	1.7761	1.5404	0.9877	0.9245	0.9393	0.9333	0.8982
	0.0963	0.0874	0.0529	0.0432	0.0385	0.0369	0.0394	0.0388	0.0384	0.038	0.0374	0.036
	4	4	4	4	2.2274	1.6587	0.9938	1.437	1.3926	0.9422	0.8922	0.9431
	0.1152	0.0791	0.0571	0.0469	0.04	0.0368	0.0365	0.0384	0.0357	0.0379	0.0359	0.0486

9	3.012	3.1397	3.2599	3.272	2.9858	2.058	1.6674	1.4802	0.9191	0.9476	0.9446	0.965
	3.0402	3.0928	3.1402	3.2522	2.8284	2.0637	1.6121	1.1018	1.4069	0.9669	1.2935	0.9552
	0.1178	0.1143	0.0587	0.0427	0.0374	0.0375	0.0368	0.0376	0.0372	0.0352	0.039	0.043
	3.2156	3.2983	3.5903	3.4748	2.622	1.808	1.4322	1.919	1.3658	1.4394	0.9511	0.9462
	3.346	3.7056	3.7734	3.6858	2.5622	1.6862	1.5155	1.4976	0.991	0.9678	0.9696	0.968
	0.1773	0.0912	0.0573	0.0442	0.0377	0.0378	0.0393	0.041	0.0369	0.0374	0.0381	0.0357
	4	4	4	3.5096	2.0128	1.6601	1.4539	0.9837	0.9042	1.4855	0.9371	0.988
	0.1242	0.0753	0.0584	0.0439	0.0395	0.0362	0.0355	0.0385	0.0366	0.0385	0.0352	0.0354
10	3.2138	3.2381	3.3722	3.4274	3.1953	2.256	1.6886	1.0321	1.3545	1.0716	1.0755	1.3097
	3.2273	3.2237	3.404	3.4063	3.131	2.1291	1.6054	1.4491	0.9946	0.9797	0.9905	1.0646
	0.1143	0.0892	0.0502	0.0427	0.0424	0.0379	0.0391	0.0376	0.0369	0.036	0.0387	0.0342
	3.2156	3.2983	3.5903	3.4748	2.622	1.808	1.4322	1.919	1.3658	1.4394	0.9511	0.9462
	3.346	3.7056	3.7734	3.6858	2.5622	1.6862	1.5155	1.4976	0.991	0.9678	0.9696	0.968
	0.1196	0.0785	0.0552	0.0445	0.042	0.0382	0.0482	0.0384	0.0372	0.0385	0.0365	0.0368
	4	4	4	4	1.991	1.6807	1.536	0.9917	0.9932	0.9393	0.9692	1.0117
	0.1021	0.0742	0.0599	0.0454	0.041	0.0367	0.0362	0.0376	0.0359	0.0374	0.0364	0.0368

12	3.1115	3.1266	3.2524	3.3424	3.0972	2.228	1.6709	1.4996	1.0455	1.4154	1.0326	0.9479
	3.0779	3.1558	3.1689	3.3158	3.0746	2.1769	1.6441	1.4928	1.4458	1.4136	0.9654	1.3661
	0.1376	0.0926	0.0568	0.0443	0.039	0.0393	0.0381	0.0385	0.0382	0.0372	0.04	0.0314
	3.4491	3.355	3.6798	3.6468	2.7534	1.8988	1.5431	1.4701	1.4534	1.0149	0.9898	0.9281
	3.4989	3.6133	4	3.8222	2.835	1.8898	1.5766	1.4204	0.9915	0.9868	0.8814	1.3291
	0.1635	0.1028	0.056	0.0467	0.0389	0.0375	0.039	0.0379	0.0432	0.0446	0.0369	0.0366
	4	4	4	4	2.2922	1.6736	1.5033	0.9653	0.9823	0.9947	0.9943	0.9365
	0.1634	0.0953	0.0596	0.0474	0.0419	0.0431	0.038	0.0383	0.0369	0.0388	0.037	0.037
13	3.1016	3.1302	3.2609	3.2884	3.0607	2.3052	1.6674	1.466	1.0338	0.9897	0.9222	1.0046
	3.1334	3.3474	3.29	3.3229	3.1958	2.2656	1.6996	1.5178	1.0496	1.4101	0.9686	1.0189
	0.1377	0.095	0.0515	0.0481	0.0406	0.0381	0.0381	0.0377	0.0382	0.0359	0.0407	0.033
	3.3036	3.4421	3.7134	3.3477	2.6491	1.7893	1.5788	1.505	0.9809	0.9802	0.9567	0.8584
	3.6442	3.5601	4	3.6609	2.7092	1.7891	1.5403	1.0974	0.9993	0.9275	1.4273	0.9408
	0.1218	0.0915	0.0557	0.0449	0.037	0.0373	0.0387	0.0419	0.0372	0.0397	0.0375	0.0365
	4	4	4	3.7126	2.3355	1.1186	1.0084	0.9437	1.0099	0.9955	0.9159	0.9221
	0.1642	0.0838	0.0552	0.0462	0.0379	0.041	0.0364	0.0389	0.0363	0.0375	0.0366	0.0368

15	3.165	3.2085	3.2001	3.3361	3.1332	2.4216	1.6911	1.4902	1.4432	0.985	1.4416	1.4177
	3.0279	3.268	3.2264	3.4064	3.2774	2.3425	1.73	1.5174	1.4553	1.0346	0.9989	1.0032
	0.1496	0.1172	0.0682	0.0453	0.0441	0.041	0.0389	0.0372	0.0379	0.0365	0.039	0.0366
	3.3413	3.5932	3.7621	3.4746	3.1445	1.9228	1.5994	1.494	1.471	1.0024	1.0679	1.011
	3.7002	3.8226	3.8738	4	3.0437	2.0129	1.5937	1.0486	0.994	0.9983	1.0027	0.9882
	0.1688	0.1092	0.0537	0.045	0.0426	0.037	0.0385	0.0379	0.0367	0.0368	0.0362	0.0359
	4	4	4	4	2.4045	1.7699	1.0415	1.5069	1.4976	0.9871	0.9735	0.9545
	0.2027	0.1064	0.0641	0.0482	0.0449	0.0366	0.0369	0.038	0.036	0.0372	0.0362	0.037
16	3.1258	3.2041	3.3029	3.368	3.2226	2.4259	1.7196	1.4993	1.397	1.3894	0.9585	1.4048
	3.2061	3.2011	3.3105	3.3409	3.2835	2.4043	1.6885	1.532	1.0086	0.9979	0.9539	1.0108
	0.1571	0.0883	0.0522	0.0433	0.0379	0.0381	0.0368	0.0362	0.0367	0.0368	0.0385	0.0309
	3.4459	3.458	3.7689	3.7149	2.9557	1.9336	1.0938	1.0504	0.9653	0.9807	0.9523	1.0447
	3.4524	3.7344	4	4	2.9646	1.8565	1.5774	1.5101	0.9844	0.977	0.9759	0.9404
	0.1547	0.0887	0.0553	0.0458	0.0397	0.0377	0.0377	0.0377	0.0366	0.0372	0.0376	0.037
	4	4	4	4	2.4978	1.7619	1.5869	1.4691	1.472	1.0102	0.9937	1.0053
	0.1582	0.0996	0.0616	0.0494	0.0422	0.0444	0.0373	0.0381	0.0365	0.0379	0.0364	0.0377

18	3.1079	3.1238	3.2657	3.3026	3.1993	2.3458	1.738	1.5369	0.9895	0.9745	0.9473	0.9739
	3.1468	3.2092	3.2898	3.339	3.1779	2.3612	1.6965	1.523	1.483	1.4372	1.0043	0.9343
	0.1168	0.0925	0.0528	0.0429	0.0386	0.0386	0.0371	0.0384	0.0923	0.0364	0.0399	0.0328
	3.4247	3.404	3.8628	3.6665	2.8523	1.9322	1.1159	1.0911	1.4713	0.999	0.9313	0.9282
	3.6793	4	4	4	2.8546	1.8916	1.5876	0.9768	0.9381	0.9074	0.9686	0.9672
	0.1353	0.0919	0.0757	0.0556	0.0428	0.0407	0.0405	0.0437	0.0409	0.0387	0.0362	0.0367
	4	4	4	4	2.3248	1.735	1.5214	1.4442	1.4293	0.9229	0.96	1.0087
	0.141	0.0851	0.053	0.0447	0.0407	0.0387	0.0365	0.0383	0.0365	0.0378	0.0364	0.0372
19	3.0959	3.1856	3.3079	3.3279	3.1515	2.246	1.752	1.5245	1.4362	0.9252	0.8859	0.9395
	3.1855	3.2354	3.4382	3.4541	3.207	2.3237	1.1851	1.5405	1.0093	1.4307	0.939	0.9125
	0.1455	0.0862	0.052	0.0431	0.0377	0.0381	0.0373	0.0447	0.0421	0.042	0.0392	0.0364
	3.4269	3.5124	3.8348	3.6048	2.7769	1.8841	1.5866	0.9891	1.459	0.9375	0.9663	0.9107
	3.8367	4	4	4	2.7707	1.8121	1.5352	1.471	1.4337	0.8908	0.9195	0.8847
	0.147	0.0817	0.0529	0.044	0.0389	0.0384	0.039	0.0384	0.0402	0.0401	0.0362	0.0484
	4	4	4	4	2.326	1.7044	1.5608	1.3864	0.9305	0.9821	0.9046	0.9069
	0.128	0.081	0.0548	0.0432	0.0365	0.0364	0.0357	0.0374	0.0353	0.0413	0.0354	0.0366

Absorbance at 620 nm – Plates 1 through 20 from 4/03/03

3	1.273	1.2751	1.2702	1.2685	1.2327	1.1929	1.1791	1.1482	1.1261	1.1584	0.7527	0.7916
	1.2528	1.24	1.2605	1.2635	1.2334	1.1759	1.2144	1.1736	0.7908	0.7857	0.776	0.7821
	0.2242	0.0807	0.0515	0.0413	0.0378	0.0353	0.0372	0.0362	0.0353	0.0354	0.0375	0.0327
	1.2021	1.2752	1.2799	1.2761	1.2383	0.7949	1.1688	0.7619	1.1408	0.7679	0.8481	0.7507
	1.2702	1.279	1.2915	1.2815	1.2303	1.1825	1.1677	1.1541	1.1477	0.7687	1.1661	0.7719
	0.1719	0.0884	0.0538	0.0416	0.0359	0.0354	0.0401	0.0369	0.0358	0.0362	0.0356	0.035
	1.278	1.292	1.2879	1.2721	1.2294	1.1915	0.7315	0.7373	0.806	0.8065	0.8048	0.794
	0.1762	0.0768	0.0521	0.0419	0.0378	0.0355	0.0357	0.036	0.0392	0.0369	0.036	0.0361
4	1.3514	1.3188	1.2715	1.2658	1.2565	1.1883	1.1585	0.7024	0.7381	0.6847	0.7141	0.7369
	1.2473	1.2348	1.2546	1.2271	1.2339	1.195	1.1556	0.7695	1.1513	1.1456	0.7069	0.7436
	0.1795	0.0787	0.0549	0.0403	0.0391	0.0366	0.0355	0.0395	0.0356	0.0355	0.0356	0.0361
	1.2617	1.2638	1.251	1.2485	1.2469	1.2062	1.1791	1.1814	0.7125	0.7257	0.6961	0.7038
	1.2737	1.2339	1.2377	1.2479	1.259	1.2364	1.1769	0.7667	1.2006	0.7416	0.7074	0.7224
	0.1328	0.0771	0.0813	0.0418	0.0398	0.0397	0.0422	0.0382	0.0375	0.0372	0.0365	0.0382
	1.3048	1.2543	1.1956	1.2393	1.1785	1.1363	1.1797	1.1756	0.7372	1.1579	0.7706	0.7712
	0.0876	0.0706	0.0489	0.041	0.0549	0.0368	0.0376	0.0364	0.0363	0.0376	0.0375	0.038

6	1.2517	1.2384	1.2669	1.2657	1.2543	1.1615	1.1687	1.1535	0.7916	0.7992	0.7572	0.7728
	1.2508	1.2521	1.231	1.2501	1.2212	1.171	1.1118	1.1508	1.1479	0.7295	1.129	0.7357
	0.1182	0.1181	0.0644	0.0459	0.0371	0.0358	0.0361	0.0372	0.0359	0.0352	0.0386	0.0312
	1.2434	1.2488	1.2695	1.2212	1.1948	1.1818	1.1376	1.1385	0.7934	1.1675	0.7474	0.6531
	1.2386	1.2319	1.2491	1.2205	1.1876	1.1416	1.1348	1.1705	0.7453	0.7828	1.2079	0.7537
	0.1642	0.0919	0.0661	0.0488	0.036	0.0368	0.0381	0.0362	0.0359	0.0364	0.0358	0.0362
	1.2147	1.233	1.2362	1.2332	1.2022	1.1475	1.1563	1.1403	0.7714	0.8022	0.7352	0.7246
	0.1467	0.0749	0.059	0.0419	0.0399	0.0362	0.0357	0.0364	0.0349	0.0385	0.0367	0.0382
7	1.2729	1.2817	1.2737	1.2745	1.2239	1.1961	1.1304	1.1429	0.7757	0.7759	0.7673	0.8127
	1.2556	1.2495	1.2407	1.2428	1.3003	1.1757	0.7708	1.1544	0.7407	1.1633	0.7471	0.7666
	0.1305	0.0916	0.0532	0.0426	0.0363	0.0374	0.0368	0.0365	0.0362	0.035	0.0384	0.0347
	1.2591	1.2523	1.2367	1.2238	1.2199	1.1612	1.1533	0.7787	0.759	0.783	0.7474	0.6588
	1.2758	1.2788	1.2482	1.2186	1.1752	1.1439	1.1735	0.765	0.7265	0.7469	0.7465	0.7157
	0.0905	0.0824	0.0497	0.0404	0.036	0.0353	0.0378	0.0369	0.0373	0.0367	0.0365	0.0348
	1.2229	1.2788	1.2563	1.2242	1.2002	1.146	0.7491	1.1516	1.1358	0.7526	0.7117	0.7598
	0.0999	0.0713	0.0534	0.0435	0.0378	0.0355	0.0358	0.0366	0.034	0.0367	0.0354	0.0473

9	1.1808	1.2055	1.2006	1.205	1.2407	1.1	1.2121	1.1657	0.7043	0.7547	0.7598	0.776
	1.2199	1.1898	1.184	1.1994	1.1856	1.1839	1.1356	0.8383	1.1527	0.7709	1.064	0.7719
	0.1099	0.1283	0.0563	0.0407	0.0348	0.0358	0.0364	0.0363	0.0357	0.0345	0.0382	0.0424
	1.2223	1.2281	1.2523	1.2504	1.2209	1.1795	1.0661	1.6686	1.099	1.2554	0.7618	0.7648
	1.2033	1.2599	1.2083	1.2244	1.1971	1.0866	1.1544	1.2385	0.7948	0.7725	0.7757	0.7811
	0.16	0.0905	0.0551	0.0418	0.0363	0.0359	0.037	0.0389	0.0358	0.0363	0.0391	0.0341
	1.2342	1.2461	1.2054	1.1922	1.1445	1.1801	1.1516	0.784	0.7198	1.2548	0.7586	0.8075
	0.1076	0.0713	0.0537	0.0402	0.0376	0.0342	0.0358	0.0367	0.0349	0.0372	0.0358	0.035
10	1.2419	1.2021	1.211	1.221	1.2608	1.2319	1.1817	0.7828	1.1021	0.8627	0.881	1.076
	1.2524	1.2234	1.2405	1.2503	1.3183	1.1332	1.1364	1.1336	0.7823	0.7789	0.7935	0.8693
	0.1148	0.0873	0.049	0.0411	0.0394	0.0366	0.0379	0.0365	0.0357	0.0348	0.0377	0.0327
	0.8575	0.8052	0.7503	0.7698	0.8162	0.7647	0.7994	0.7836	0.8093	0.8028	0.8103	0.8608
	0.8251	0.7265	0.7844	0.7369	0.7554	0.7388	0.7596	0.7791	0.7329	0.7644	0.7731	0.7674
	0.1058	0.071	0.0504	0.0416	0.0394	0.0364	0.0451	0.0368	0.0361	0.0372	0.0355	0.0352
	1.1981	1.2484	1.256	1.2109	1.0753	1.1722	1.1935	0.7774	0.7996	0.7552	0.7829	0.8314
	0.089	0.0703	0.0558	0.0426	0.0388	0.0358	0.0357	0.0356	0.0346	0.0364	0.0356	0.0357

12	1.2783	1.2447	1.2546	1.2612	1.2521	1.2273	1.1665	1.16	0.8118	1.1665	0.8244	0.7457
	1.2549	1.2701	1.2531	1.2577	1.2436	1.1816	1.1612	1.1567	1.1736	1.1494	0.7688	1.1223
	0.1295	0.0884	0.0541	0.0427	0.0366	0.0384	0.0378	0.0375	0.0366	0.0359	0.0389	0.0311
	1.3562	1.3164	1.2879	1.272	1.2621	1.1917	1.1621	1.1735	1.1783	0.8089	0.7924	0.735
	1.283	1.2679	1.2714	1.2363	1.2196	1.1845	1.1768	1.1326	0.787	0.7855	0.6908	1.0886
	0.1674	0.099	0.0543	0.0446	0.0362	0.0357	0.0372	0.0364	0.0418	0.0418	0.036	0.0351
	1.2621	1.2739	1.2934	1.2676	1.2208	1.1768	1.1689	0.7556	0.785	0.7943	0.7925	0.7418
	0.1691	0.086	0.0562	0.0445	0.0398	0.0414	0.0371	0.0366	0.0355	0.0378	0.0361	0.0385
13	1.2595	1.2718	1.235	1.2575	1.2055	1.2026	1.1442	1.1276	0.8116	0.7774	0.7278	0.8035
	1.2647	1.4048	1.2612	1.225	1.2535	1.1798	1.1855	1.1773	0.8289	1.1551	0.7725	0.8227
	0.1415	0.0873	0.0489	0.0449	0.038	0.0361	0.0365	0.0367	0.0365	0.0347	0.0395	0.0333
	1.2411	1.2577	1.2722	1.2118	1.2103	1.1679	1.1821	1.1844	0.7792	0.7797	0.7542	0.671
	1.2813	1.2553	1.2711	1.2375	1.2177	1.1565	1.1693	0.9114	0.7987	0.7281	1.1758	0.746
	0.1255	0.0888	0.053	0.0423	0.0355	0.0356	0.0371	0.0393	0.0364	0.0383	0.0366	0.035
	1.3633	1.284	1.2403	1.2377	1.1912	0.7774	0.7375	0.7229	0.804	0.7942	0.727	0.7307
	0.1788	0.0791	0.0512	0.043	0.0362	0.0388	0.0356	0.037	0.0344	0.0364	0.0356	0.0356

15	1.2646	1.2518	1.2192	1.2383	1.2111	1.1905	1.1045	1.1121	1.1314	0.7432	1.1634	1.1437
	1.2642	1.2703	1.2331	1.2579	1.2411	1.1736	1.1109	1.1423	1.1481	0.7962	0.7707	0.7841
	0.1474	0.1124	0.0614	0.0427	0.0384	0.0377	0.0405	0.0361	0.036	0.0351	0.0377	0.0349
	1.249	1.2866	1.2631	1.2349	1.274	1.1642	1.1519	1.1764	1.1935	0.7793	0.846	0.7959
	1.2637	1.2794	1.2648	1.2593	1.2577	1.2064	1.1863	0.8025	0.7718	0.7811	0.7938	0.7834
	0.1749	0.1104	0.0509	0.0422	0.0401	0.0352	0.0368	0.0366	0.0357	0.0355	0.0356	0.0347
	1.3907	1.3283	1.2996	1.3403	1.3095	1.2502	0.7635	1.2171	1.2258	0.7794	0.7676	0.7543
	0.1692	0.0944	0.0597	0.0451	0.0416	0.0353	0.0359	0.0363	0.0347	0.0364	0.0365	0.0357
16	1.2217	1.2307	1.2239	1.2477	1.2001	1.1956	1.1103	1.11	1.0872	1.0997	0.7317	1.1279
	1.2653	1.2678	1.2305	1.2349	1.2507	1.1848	1.1223	1.1525	0.7574	0.7726	0.7294	0.7942
	0.1537	0.0857	0.0497	0.0419	0.0354	0.0363	0.0362	0.0355	0.0356	0.0353	0.038	0.0304
	1.2691	1.2821	1.2787	1.2601	1.2264	1.1856	0.7601	0.7933	0.7387	0.7629	0.7369	0.8338
	1.2376	1.2607	1.2702	1.2509	1.239	1.2027	1.1512	1.1961	0.7643	0.7593	0.7619	0.7325
	0.1735	0.0892	0.0538	0.043	0.0371	0.0357	0.0367	0.0361	0.0356	0.0358	0.0369	0.0356
	1.2351	1.266	1.2523	1.2354	1.1935	1.1736	1.2185	1.179	1.1971	0.7959	0.7869	0.7984
	0.1676	0.0919	0.0582	0.0461	0.0398	0.0417	0.0361	0.036	0.0349	0.0367	0.0359	0.0363

18	1.2359	1.2376	1.2205	1.2659	1.2209	1.1863	1.1289	1.1917	0.7546	0.7475	0.7266	0.7619
	1.2413	1.2529	1.2531	1.2089	1.2295	1.2098	1.1426	1.1531	1.2082	1.18	0.7934	0.7308
	0.1225	0.0852	0.0477	0.0404	0.0364	0.0365	0.0367	0.0373	0.0777	0.0352	0.0388	0.0318
	1.2917	1.229	1.2631	1.2618	1.2493	1.1883	0.8054	0.8451	1.2039	0.7878	0.7251	0.7292
	1.2589	1.2498	1.2293	1.2692	1.2116	1.1502	1.1872	0.7398	0.7251	0.7053	0.7635	0.7688
	0.1352	0.0911	0.0702	0.0515	0.0402	0.0386	0.0388	0.0414	0.0386	0.0374	0.0355	0.0353
	1.2457	1.2367	1.2321	1.2394	1.1822	1.2069	1.1741	1.1568	1.1674	0.7263	0.7643	0.8131
	0.1258	0.0785	0.05	0.0414	0.0387	0.0371	0.0357	0.0365	0.0345	0.0366	0.0356	0.0358
19	1.2357	1.2326	1.2353	1.2625	1.1994	1.1603	1.1267	1.157	1.1425	0.7113	0.6781	0.731
	1.2239	1.2284	1.2425	1.261	1.252	1.1891	0.7608	1.1611	0.774	1.1842	0.7355	0.7143
	0.1385	0.0807	0.0487	0.0415	0.0356	0.0365	0.0367	0.0436	0.0374	0.0401	0.0382	0.0352
	1.2398	1.2133	1.2506	1.2355	1.2213	1.2053	1.1894	0.7516	1.1951	0.7386	0.763	0.7126
	1.2468	1.233	1.2504	1.2339	1.2093	1.147	1.1392	1.1707	1.1771	0.699	0.7184	0.6917
	0.1472	0.0766	0.0499	0.0416	0.037	0.0363	0.0375	0.0368	0.0395	0.0405	0.0369	0.04
	1.2344	1.2757	1.2359	1.2376	1.1852	1.1824	1.2033	1.1129	0.7279	0.7833	0.7111	0.7172
	0.1148	0.0741	0.0507	0.0405	0.035	0.0354	0.0352	0.0355	0.034	0.0412	0.0351	0.0356

Absorbance at 620 nm for Blanks – Plates 0 through 20 from 4/03/03

5	0.7883	0.8101	0.7624	0.7929	0.7635	0.8372	0.7738	0.7522	0.8155	0.7702	0.8101	0.8671
	0.1099	0.087	0.0393	0.0361	0.0379	0.0403	0.0377	0.0377	0.0351	0.0352	0.0361	0.0357
	0.85	0.8138	0.7653	0.7779	0.7566	0.854	0.6834	0.768	0.8391	0.7989	0.7839	0.8265
	0.1443	0.0507	0.0628	0.0686	0.0387	0.0387	0.0374	0.037	0.0377	0.0379	0.0394	0.0405
	0.6453	0.8168	0.7867	0.7453	0.7559	0.716	0.7275	0.7575	0.7439	0.7236	0.6808	0.7736
	0.0901	0.0645	0.088	0.0364	0.0364	0.0365	0.0372	0.0368	0.0363	0.0372	0.0352	0.0348
	0.0466	0.0464	0.0451	0.0446	0.0455	0.0531	0.0454	0.0473	0.0455	0.0463	0.0479	0.0464
	0.0452	0.0461	0.048	0.0463	0.0452	0.0459	0.0451	0.0484	0.044	0.0473	0.045	0.0455
8	0.8018	0.8013	0.8278	0.8236	0.7736	0.7411	0.8228	0.767	0.7591	0.7937	0.7506	0.8798
	0.042	0.0403	0.0395	0.0348	0.0408	0.0397	0.0376	0.0377	0.0347	0.0352	0.0363	0.0358
	0.8722	0.804	0.8614	0.8675	0.8411	0.7715	0.722	0.7623	0.693	0.7494	0.7331	0.8579
	0.0839	0.0438	0.0364	0.0365	0.0387	0.0385	0.0376	0.0368	0.0364	0.0374	0.0394	0.0368
	0.7812	0.8967	0.7623	0.7881	0.8048	0.8293	0.7402	0.7991	0.7803	0.821	0.7075	0.7305
	0.1117	0.0806	0.0354	0.0417	0.0394	0.0372	0.0422	0.0374	0.036	0.0374	0.0367	0.035
	0.0465	0.0463	0.045	0.0448	0.0463	0.0532	0.0456	0.0473	0.0456	0.0474	0.049	0.0457
	0.0457	0.0461	0.0491	0.0462	0.0455	0.0454	0.0453	0.0467	0.0441	0.0473	0.0452	0.0447

11	0.8247	0.8417	0.8164	0.889	0.8783	0.8003	0.8183	0.8674	0.7606	0.7696	0.8228	0.7576
	0.1263	0.0564	0.0366	0.0371	0.0365	0.0376	0.0372	0.0374	0.0347	0.0352	0.0367	0.0357
	0.7866	0.7993	0.7576	0.7387	0.8178	0.7176	0.7125	0.7679	0.8045	0.8369	0.8453	0.8335
	0.0864	0.0427	0.037	0.0371	0.0387	0.0384	0.0379	0.0368	0.0362	0.0388	0.0387	0.0381
	0.7767	0.8098	0.7903	0.8418	0.8068	0.8081	0.8333	0.8158	0.7862	0.7669	0.736	0.7807
	0.1364	0.0365	0.0358	0.0358	0.0365	0.0452	0.0377	0.0362	0.0366	0.0376	0.0367	0.0352
	0.0495	0.046	0.0452	0.045	0.0464	0.0538	0.0472	0.0486	0.0455	0.0464	0.049	0.0462
	0.0452	0.0457	0.0482	0.0462	0.0456	0.0456	0.0454	0.0466	0.0441	0.048	0.0449	0.0449
14	0.6874	0.724	0.7516	0.7871	0.6956	0.7178	0.7257	0.6671	0.7135	0.8433	0.7417	0.8442
	0.1283	0.045	0.038	0.0383	0.0359	0.0378	0.0375	0.0373	0.0345	0.0349	0.0359	0.0356
	0.7442	0.7587	0.7458	0.8818	0.8122	0.794	0.7481	0.8902	0.6881	0.8799	0.7038	0.6836
	0.1741	0.038	0.0773	0.0365	0.0413	0.038	0.0455	0.0361	0.0361	0.037	0.0384	0.0364
	0.7688	0.8019	0.7692	0.8432	0.818	0.7822	0.8054	0.8002	0.7523	0.7678	0.8141	0.7924
	0.1037	0.0356	0.0784	0.0683	0.0354	0.0357	0.0378	0.0364	0.0362	0.0369	0.037	0.0346
	0.0465	0.0457	0.0452	0.0444	0.0464	0.0531	0.0457	0.0479	0.0456	0.0465	0.0481	0.0458
	0.0469	0.046	0.0483	0.046	0.0456	0.0452	0.0451	0.0475	0.0445	0.0474	0.0449	0.0448

17	0.6805	0.6827	0.7159	0.6902	0.7587	0.6597	0.6857	0.6112	0.7368	0.7458	0.7103	0.7285
	0.0985	0.0855	0.0618	0.0348	0.0362	0.0378	0.0379	0.0373	0.0356	0.0353	0.0365	0.0354
	0.671	0.7277	0.7469	0.8439	0.7996	0.7856	0.7147	0.784	0.8166	0.7908	0.8225	0.8126
	0.1528	0.1674	0.0372	0.0362	0.039	0.0381	0.0357	0.0371	0.0359	0.0371	0.0391	0.0361
	0.7744	0.7366	0.7748	0.7551	0.7307	0.755	0.8093	0.7641	0.7155	0.7664	0.7784	0.8402
	0.0563	0.0387	0.0355	0.0391	0.0361	0.0379	0.0376	0.0353	0.0361	0.0375	0.0367	0.0353
	0.0464	0.0456	0.0451	0.0445	0.0456	0.0533	0.0493	0.048	0.0455	0.0466	0.0479	0.0459
	0.0458	0.0461	0.0493	0.046	0.0457	0.0456	0.0465	0.0471	0.0462	0.048	0.0452	0.0457
20	0.6594	0.6239	0.6877	0.6735	0.6467	0.6515	0.6702	0.6515	0.6497	0.678	0.7621	0.7648
	0.2314	0.0372	0.0386	0.0353	0.037	0.0377	0.0375	0.0373	0.0348	0.0356	0.0363	0.0354
	0.7635	0.7303	0.6817	0.8262	0.8621	0.8528	0.8301	0.7915	0.8188	0.7945	0.8061	0.8237
	0.2668	0.058	0.0376	0.0374	0.04	0.0384	0.037	0.0359	0.0364	0.0371	0.0396	0.0377
	0.7812	0.7131	0.7955	0.7816	0.6887	0.7396	0.8828	0.7656	0.8831	0.7891	0.7934	0.7493
	0.2767	0.0353	0.0396	0.0359	0.0366	0.0358	0.0395	0.0366	0.0356	0.0366	0.0368	0.0353
	0.0467	0.0466	0.0456	0.045	0.0476	0.0538	0.0466	0.0485	0.0468	0.0491	0.0484	0.046
	0.0458	0.0496	0.0489	0.0466	0.0463	0.0464	0.0454	0.0482	0.0446	0.0477	0.0457	0.0472

Samples from 5/12/03

Plate Number	Description
1	Whole Sample - E2 Spiked
2	Whole Sample - E2 Spiked
3	Whole Sample - Blank
4	< 1.5 um Sample - E2 Spiked
5	< 1.5 um Sample - E2 Spiked
6	< 1.5 um Sample - Blank
7	< 0.22 um Sample - E2 Spiked
8	< 0.22 um Sample - E2 Spiked
9	< 0.22 um Sample - Blank
10	100 kD Sample - E2 Spiked
11	100 kD Sample - E2 Spiked
12	100 kD Sample - Blank
13	30 kD Sample - E2 Spiked
14	30 kD Sample - E2 Spiked
15	30 kD Sample - Blank
16	10 kD Sample - E2 Spiked
17	10 kD Sample - E2 Spiked
18	10 kD Sample - Blank
19	1 kD Sample - E2 Spiked
20	1 kD Sample - E2 Spiked
21	1 kD Sample - Blank

Samples from 5/12/03 – Summary of E2 Dose-Response Curves and Organic Carbon Parameters

Exper	COC	Repl	Min	Max	EC50	EC50 err	Slope	Slope err	Protein	Polys	Humics	Fulvics
whole	1.730889	1	0.80272	2.88449	104.8351	9.43623	2.20488	0.379	1.27	0.33	1.49	1.94
whole	2.596333	1	0.84029	3.28014	143.6628	5.54862	2.90359	0.26568	1.695	0.44	1.98	2.59
whole	3.1156	1	0.85699	3.39534	156.0622	6.31841	4.49723	1.13573	1.905	0.49	2.23	2.91
whole	1.730889	2	0.82025	2.8389	146.2548	8.18831	3.47592	0.70718	1.27	0.33	1.49	1.94
whole	2.596333	2	0.82382	3.36988	163.4495	1.87152	3.08146	0.09295	1.695	0.44	1.98	2.59
whole	3.1156	2	0.86979	3.40344	182.7706	14.19599	7.45475	2.63216	1.905	0.49	2.23	2.91
<1.5	0.969111	1	0.82354	2.7383	145.5461	9.13824	4.07584	1.30388	0.885	0.26	0.38	2.07
<1.5	1.453667	2	0.90667	3.20707	111.3879	6.23665	5.31935	1.80798	1.18	0.35	0.51	2.76
<1.5	1.7444	3	0.91887	3.42784	117.9618	5.61515	2.8045	0.31578	1.33	0.4	0.57	3.1
<1.5	0.969111	1	0.79893	2.71175	80.87976	4.0295	3.95685	0.84123	0.885	0.26	0.38	2.07
<1.5	1.453667	2	0.85774	3.08362	131.9929	9.29467	2.84171	0.47026	1.18	0.35	0.51	2.76
<1.5	1.7444	3	0.95114	3.45539	168.2371	5.85204	5.01901	0.96403	1.33	0.4	0.57	3.1
<0.22	1.160222	1	0.90464	2.78032	102.2019	4.59677	3.80442	0.45478	0.645	0.26	0.2	1.22
<0.22	1.740333	2	0.88814	3.15737	124.4805	4.20725	4.16611	0.43853	0.855	0.34	0.26	1.63
<0.22	2.0884	3	0.88161	3.47426	135.1792	1.86203	3.96126	0.22884	0.965	0.38	0.3	1.83
<0.22	1.160222	1	0.89139	2.78392	115.8774	5.61039	2.44117	0.24793	0.645	0.26	0.2	1.22
<0.22	1.740333	2	0.89903	3.18343	122.1438	7.05825	4.03273	0.74214	0.855	0.34	0.26	1.63
<0.22	2.0884	3	0.97325	3.41601	177.4914	17.45391	5.6451	2.57762	0.965	0.38	0.3	1.83
<100kDa	0.782889	1	0.94504	2.84899	85.56249	5.1911	4.31875	1.0805	0.57	0.25	0.14	1.28
<100kDa	1.174333	2	0.98411	3.23762	97.22425	5.04997	4.74819	1.85454	0.76	0.33	0.19	1.7
<100kDa	1.4092	3	0.97267	3.22745	114.0253	15.89188	5.58259	2.44839	0.85	0.37	0.21	1.91
<100kDa	0.782889	1	0.95513	2.85025	143.9189	14.77484	3.45621	1.26672	0.57	0.25	0.14	1.28
<100kDa	1.174333	2	0.96316	3.44274	115.9021	2.7066	5.28886	0.58933	0.76	0.33	0.19	1.7
<100kDa	1.4092	3	0.91751	3.42424	98.44606	1.39302	4.44079	0.17962	0.85	0.37	0.21	1.91
<30kDa	0.784667	1	0.86007	2.86184	91.85836	8.31939	2.55048	0.50664	0.57	0.25	0.17	2.26
<30kDa	1.177	2	0.966	3.33657	103.0569	3.58954	5.49864	2.06904	0.76	0.33	0.22	3.01
<30kDa	1.4124	3	0.93557	3.35045	165.0683	8.1939	6.35903	2.61634	0.86	0.37	0.25	3.39
<30kDa	0.784667	1	0.88727	2.84118	100.1131	3.20611	4.97415	0.45365	0.57	0.25	0.17	2.26
<30kDa	1.177	2	0.93783	3.30034	107.334	4.27349	6.98052	3.08412	0.76	0.33	0.22	3.01
<30kDa	1.4124	3	0.92354	3.38942	162.0743	12.95214	8.03145	7.3239	0.86	0.37	0.25	3.39
<10kDa	0.747889	1	0.89171	2.91119	134.4819	8.15765	3.09421	0.53577	0.555	0.18	0.15	1.32
<10kDa	1.121833	2	0.91758	3.33934	119.6564	2.29583	4.11479	0.26853	0.74	0.24	0.21	1.75
<10kDa	1.3462	3	0.961	3.38714	176.2742	14.75562	6.37726	2.7831	0.835	0.28	0.23	1.97
<10kDa	0.747889	1	0.87502	2.80303	150.7053	5.16256	2.46714	0.18624	0.555	0.18	0.15	1.32
<10kDa	1.121833	2	0.80302	3.29843	110.1094	9.05947	3.97129	1.32948	0.74	0.24	0.21	1.75
<10kDa	1.3462	3	1.05085	3.38097	166.487	20.56877	8.37607	9.06795	0.835	0.28	0.23	1.97
<1kda		1	0.8731	2.80903	104.5957	5.77606	4.88044	0.73885				
<1kda		2	0.90615	3.29889	101.1628		39.43406					
<1kda		3	0.92455	3.38244	80.04141	35.81995	11.32696	64.37781				
<1kda		1	0.85957	2.88925	101.2808	4.67368	4.19171	0.52843				

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Exper	COC	Repl	Min	Max	EC50	EC50 err	Slope	Slope err	Protein	Polys	Humics	Fulvics
<1kda		2	0.93167	3.25278	101.7056	61509038	49.22497	1.1E+09				
<1kda		3	0.93955	3.39447	166.4549	7.34293	5.68721	1.71304				

Absorbance at 540 nm – Plates 1 through 20 for 5/12/03

1	3.3546	3.5441	3.6109	3.548	3.6162	2.933	1.9777	1.5904	0.8343	0.6605	0.7657	0.7249
	3.4243	3.5085	3.5693	3.581	3.4356	1.7067	1.9937	0.7641	1.3471	0.7317	0.7885	0.7929
	0.2432	0.2251	0.2735	0.3034	0.2668	0.2153	0.2305	0.2284	0.0387	0.2323	0.1917	0.1383
	3.7499	3.7304	3.8648	3.5956	3.1167	1.8886	1.4671	0.937	0.8901	1.4093	0.6623	0.9099
	3.8793	4	4	3.9663	3.1624	1.9589	1.4618	0.9045	0.7722	0.9373	0.8396	0.8408
	0.2517	0.2289	0.2093	0.2723	0.2184	0.2758	0.3245	0.0388	0.328	0.0386	0.036	0.0353
	4	4	4	4	2.4045	1.6108	1.0146	0.9522	0.9987	0.8049	0.8692	0.6284
	0.3013	0.2628	0.2792	0.2463	0.304	0.3445	0.3129	0.3137	0.3148	0.2786	0.0363	0.1577
2	3.296	3.5037	3.6296	3.6597	3.5998	1.5007	1.0163	0.7986	0.6492	0.6976	0.6274	0.6897
	3.3995	3.3523	3.4805	3.5382	3.391	2.5028	1.869	1.4271	0.8197	0.852	0.8438	0.5344
	0.1815	0.1757	0.2199	0.1906	0.2244	0.1728	0.2116	0.2388	0.1831	0.1961	0.0384	0.0328
	3.9434	4	4	3.6679	3.092	2.0803	0.9747	0.9367	0.925	0.8841	0.7118	0.7967
	4	4	4	4	2.8979	1.1204	0.9371	0.8881	1.3549	0.8407	0.7063	0.5633
	0.1885	0.1707	0.2126	0.1977	0.2558	0.2192	0.2703	0.2579	0.1866	0.0378	0.0359	0.0346
	4	4	4	4	1.3602	1.7213	0.9417	0.9048	0.8928	0.8289	0.9198	0.8592
	0.1501	0.1799	0.1979	0.1809	0.2243	0.1967	0.2093	0.2558	0.0352	0.0368	0.1647	0.0355

4	3.2656	3.4642	3.4236	3.5321	3.5332	1.9376	1.067	0.7945	0.7193	0.7832	1.3598	0.6205
	3.3336	3.2795	3.3989	3.5966	3.5122	1.9945	1.2853	0.9552	0.8812	0.8688	1.0019	0.8341
	0.2812	0.2492	0.251	0.277	0.2259	0.175	0.1963	0.0379	0.182	0.1841	0.2247	0.1291
	3.3485	3.6766	3.7793	3.8018	3.7996	1.5665	1.797	0.9654	1.4605	1.4045	0.9821	0.9833
	4	4	3.9934	4	4	2.6117	1.7207	1.0241	0.9756	0.9508	0.8693	1.0152
	0.2822	0.2733	0.2559	0.2521	0.2717	0.2325	0.201	0.1961	0.1478	0.0372	0.1726	0.0356
	4	4	4	4	2.9061	2.0496	0.9719	1.0251	1.3879	0.6927	0.8254	0.9177
	0.3029	0.2906	0.2657	0.2401	0.2582	0.205	0.1957	0.2918	0.2105	0.0372	0.0357	0.1636
5	3.2801	3.3281	3.4397	3.5165	3.55	3.3318	2.2602	0.9989	0.8329	0.7572	0.918	0.8894
	3.1556	3.2447	3.4931	3.4268	3.5334	3.1963	2.1836	1.5664	1.5312	0.8987	0.8935	0.7842
	0.2479	0.2908	0.2359	0.2599	0.2175	0.1988	0.171	0.2011	0.0354	0.1526	0.1841	0.1211
	3.5046	3.5581	3.685	3.7499	3.6481	2.6316	1.0578	0.9871	0.909	1.4797	1.2612	0.8604
	3.5909	3.9042	4	3.8451	1.9895	1.357	1.0318	0.846	0.8858	0.9453	0.8252	0.9698
	0.2449	0.2519	0.2315	0.2392	0.2012	0.2303	0.2163	0.1897	0.0361	0.0399	0.0355	0.0348
	4	4	4	4	1.8227	1.1659	0.9774	1.0302	0.9063	0.8195	0.9735	1.085
	0.2338	0.2061	0.2272	0.2045	0.2269	0.2229	0.2153	0.1856	0.2884	0.0366	0.0355	0.0356

7	3.1656	3.331	3.3347	3.5103	3.5692	2.9693	1.8905	0.7628	1.0017	0.789	0.8062	0.8568
	3.2302	3.3324	3.3288	3.4798	3.3294	2.9544	1.0907	1.5406	0.9564	0.9291	0.8383	0.8029
	0.2483	0.257	0.2328	0.2389	0.2415	0.2291	0.2068	0.2141	0.2243	0.0357	0.171	0.0363
	3.4911	3.5363	3.7677	3.7595	3.5335	1.3556	0.9936	0.9663	0.9182	0.9481	0.9312	0.869
	3.8083	3.8584	4	4	3.3063	2.3009	1.0221	0.9266	0.9194	0.9248	0.9221	0.7227
	0.2883	0.2655	0.2722	0.3178	0.2464	0.2445	0.2022	0.1826	0.0353	0.0344	0.0352	0.0358
	4	4	4	4	3.0099	1.0858	0.9055	0.8655	0.8915	0.7909	0.8812	0.8554
	0.1991	0.1882	0.2016	0.2236	0.2236	0.2233	0.1708	0.18	0.1779	0.0354	0.0362	0.0743
8	3.3001	3.4497	3.5808	3.4184	2.794	1.9347	1.17	1.4899	0.9338	0.8755	1.0147	1.0434
	3.2404	3.3606	3.4194	3.3959	2.7483	3.0871	2.122	1.0086	1.0217	0.9404	0.8768	1.0083
	0.2596	0.2698	0.2308	0.2348	0.2248	0.2265	0.2224	0.1987	0.1861	0.0367	0.0387	0.1464
	3.5158	3.7976	3.9162	3.569	3.5935	1.5258	1.2202	0.968	1.0391	0.8027	1.24	0.9367
	3.6088	4	4	3.9443	3.4437	2.272	1.5353	1.184	1.0853	0.9594	0.9132	0.6572
	0.2441	0.2467	0.2617	0.2187	0.2286	0.222	0.2434	0.0388	0.2071	0.2504	0.043	0.0551
	4	4	4	4	1.6282	2.0329	1.0218	1.4139	1.0694	0.9392	0.907	0.9821
	0.2159	0.1348	0.1952	0.2141	0.2399	0.2201	0.2011	0.2002	0.2366	0.0384	0.0411	0.0362

10	3.1617	3.4248	3.413	3.6947	3.5871	3.2769	1.3035	0.959	0.8858	1.0856	1.0176	0.9959
	3.2285	3.3003	3.4491	3.575	3.1933	3.2321	2.2801	1.0684	1.5183	1.0252	1.0597	1.065
	0.2248	0.3027	0.2644	0.1445	0.1421	0.2724	0.2988	0.299	0.2217	0.2265	0.0387	0.1358
	3.279	3.6045	3.1529	4	3.8158	2.6859	1.0756	1.1332	1.0387	1.0097	0.9809	1.0158
	3.7739	4	4	4	4	2.6512	1.8229	1.5666	1.0514	1.4699	0.9937	0.9824
	0.1654	0.1608	0.1477	0.2433	0.2602	0.2579	0.2873	0.2572	0.1985	0.0377	0.2074	0.0359
	3.8305	4	3.0374	4	3.3712	1.2132	1.6545	1.521	1.0794	0.909	1.0928	1.1631
	0.2267	0.2447	0.3256	0.2419	0.4563	0.4276	0.2324	0.2879	0.2471	0.0372	0.0359	0.0361
11	3.1798	3.3683	3.4613	3.5834	3.5835	1.8958	2.0655	0.9359	0.9319	0.7923	0.8432	0.8586
	3.254	3.2788	3.3193	3.4393	3.5495	1.7991	1.2031	1.0909	0.9363	0.9451	0.8749	0.9999
	0.1951	0.2041	0.2274	0.2458	0.2591	0.2294	0.2235	0.2173	0.1493	0.0361	0.0387	0.0353
	3.4852	3.4862	4	4	3.8293	1.4646	1.0204	1.0168	0.973	0.9652	0.9189	0.8708
	3.7299	3.249	4	4	4	2.5327	1.0768	0.939	0.9198	0.9203	0.9553	0.9871
	0.2809	0.195	0.2139	0.1179	0.2583	0.2286	0.3302	0.2761	0.2097	0.1902	0.1828	0.0357
	4	4	4	4	3.6903	1.9795	1.0069	0.8947	0.9076	0.8841	0.8493	1.0231
	0.1366	0.1914	0.2104	0.1846	0.2327	0.0954	0.2391	0.2597	0.1915	0.0368	0.0359	0.0353

13	3.2934	3.5111	3.4272	3.4239	3.436	2.1859	2.1679	1.0532	0.9171	0.9599	0.8965	1.0275
	3.3144	3.4531	3.5335	3.6065	3.5576	3.1623	2.1805	1.0541	1.0351	0.8328	1.0156	1.1277
	0.1923	0.2198	0.2232	0.2004	0.2163	0.179	0.2103	0.1977	0.2927	0.0353	0.0401	0.0341
	3.554	3.8413	3.9308	3.8895	4	2.5837	1.2915	1.0069	1.1024	1.0523	1.4166	1.0289
	4	4	4	4	4	2.4979	1.7109	1.4846	0.9556	1.0002	1.0766	1.085
	0.204	0.2146	0.2098	0.2106	0.204	0.2171	0.2244	0.0375	0.0364	0.0377	0.0407	0.0839
	4	4	4	4	1.8333	1.338	1.0813	0.9284	0.9771	1.0129	0.9102	1.0534
	0.1315	0.1933	0.19	0.1683	0.1939	0.2146	0.0354	0.037	0.1851	0.0375	0.0395	0.0362
14	3.328	3.463	3.4457	3.4387	3.5274	3.1139	1.3152	0.971	0.9455	0.8931	0.8088	1.0044
	3.4175	3.313	3.444	3.5337	3.4651	3.0311	1.2719	1.0157	0.9627	0.9107	0.8771	0.8474
	0.1769	0.1904	0.1761	0.202	0.1569	0.1597	0.1521	0.1565	0.1342	0.036	0.1456	0.1339
	3.619	3.7194	4	3.7292	3.8455	2.0396	1.0776	0.9716	0.9377	0.9099	0.9097	0.9004
	4	4	4	4	3.8106	2.4568	1.0678	1.4818	0.8665	0.8905	0.8658	0.8696
	0.1799	0.1878	0.1699	0.1772	0.1678	0.1618	0.1738	0.0377	0.2072	0.0368	0.0356	0.1391
	4	4	4	4	1.8333	1.121	0.9801	0.9404	0.9311	0.8657	0.8535	0.937
	0.134	0.1372	0.1701	0.1534	0.1474	0.1618	0.1526	0.0371	0.0355	0.1472	0.0357	0.0376

16	3.5076	3.61	3.5114	3.5469	3.5261	2.8179	1.9582	1.5754	0.9124	0.847	0.8853	0.8863
	3.4229	3.4416	3.5021	3.5305	3.5127	1.6965	1.1783	1.0102	0.9668	0.8788	0.9317	0.8118
	0.2459	0.279	0.2555	0.2532	0.2033	0.1793	0.1984	0.2411	0.1492	0.2112	0.1974	0.1708
	3.755	3.8339	3.9953	3.9285	3.2731	1.4132	1.075	0.9984	1.4951	0.9501	0.947	0.849
	4	4	4	4	4	2.505	1.0327	0.9277	0.9351	0.9281	0.9155	0.9208
	0.2042	0.2365	0.2274	0.2628	0.2339	0.2361	0.247	0.2187	0.0359	0.0374	0.0358	0.0348
	4	4	4	4	1.6426	2.0078	0.9227	0.9459	0.8829	0.8824	0.9271	0.8774
	0.1473	0.152	0.177	0.15	0.1974	0.1853	0.1518	0.18	0.0357	0.0365	0.0356	0.1604
17	3.4346	3.46	3.5289	3.5769	2.7737	3.0647	1.2114	0.939	0.9061	0.8787	0.9568	0.7837
	3.3699	3.4138	3.4076	3.2367	2.6864	1.1745	1.2016	0.9782	0.9374	0.9016	0.921	0.9551
	0.2994	0.2635	0.2304	0.2343	0.216	0.162	0.159	0.1607	0.1607	0.2005	0.1578	0.1427
	3.5192	3.9708	4	3.9485	3.6388	1.5477	1.119	1.0397	1.0008	1.4923	0.8976	0.9295
	3.89	4	4	4	4	2.6505	1.1832	1.1109	0.9666	1.0256	0.7903	0.8468
	0.3241	0.2552	0.3175	0.2609	0.212	0.2185	0.2147	0.2423	0.1815	0.1694	0.0362	0.0358
	4	4	4	4	1.8333	1.2545	1.0038	1.0692	0.9628	0.9845	1.0273	1.0587
	0.2622	0.1992	0.2098	0.2002	0.2661	0.1607	0.1942	0.0566	0.0372	0.0369	0.0355	0.0356

19	3.2612	3.4875	3.4288	3.4749	3.3785	3.0356	1.2545	1.0125	0.9499	0.9596	0.8923	0.9001
	3.3227	3.3202	3.4807	3.5967	3.5968	3.1535	1.2479	1.6658	0.9566	0.9005	0.9306	0.9493
	0.1862	0.2097	0.1865	0.1769	0.164	0.1393	0.1381	0.1788	0.123	0.1699	0.0379	0.0344
	3.6084	4	4	4	3.97	2.4292	1.1057	0.8825	0.9604	0.9547	0.8716	0.9215
	3.8569	4	4	4	4	1.4082	1.0735	1.0005	0.9403	0.9431	0.894	0.9597
	0.222	0.2107	0.1938	0.1935	0.187	0.1697	0.1759	0.1443	0.1354	0.0368	0.036	0.0352
	4	4	4	4	4	2.2033	1.6136	1.0036	0.9058	0.8812	0.8602	0.9205
	0.1918	0.1951	0.1657	0.1725	0.1795	0.1899	0.0433	0.0367	0.0351	0.038	0.0376	0.0411
20	3.4956	3.6544	3.7534	3.065	3.6607	3.2538	1.1845	1.0041	0.878	0.7963	0.91	0.9811
	3.3419	3.4262	3.5867	3.6419	3.6601	3.1753	1.9756	1.0577	1.0752	1.0545	1.0141	1.0491
	0.2086	0.1889	0.1716	0.1582	0.1848	0.1519	0.1538	0.1591	0.0357	0.0357	0.1322	0.0354
	3.4887	4	4	4	4	1.4314	1.798	1.0627	0.9282	0.9175	0.9011	0.929
	3.7994	4	4	4	4	1.4345	1.0361	0.979	1.4231	0.9612	0.9904	0.9531
	0.1998	0.1993	0.1783	0.1749	0.2071	0.2066	0.1772	0.1757	0.0362	0.1326	0.0357	0.0358
	4	4	4	4	4	1.8849	1.2014	1.0507	0.9553	0.8579	0.9008	0.94
	0.2236	0.2498	0.2251	0.2279	0.1872	0.1744	0.1644	0.1675	0.037	0.1566	0.0373	0.0371

Absorbance at 620 nm – Plates 1 through 20 for 5/12/03

1	1.2554	1.3592	1.3226	1.2693	1.3247	1.1321	1.0193	1.1561	0.6477	0.5199	0.625	0.5866
	1.3612	1.2988	1.3452	1.3532	1.2589	0.6789	1.1696	0.5148	1.0977	0.5854	0.6456	0.6563
	0.2152	0.208	0.2104	0.2455	0.1696	0.1508	0.1502	0.163	0.0363	0.1712	0.1443	0.1174
	1.3293	1.3029	1.2628	1.158	1.205	1.1038	1.1117	0.7716	0.7384	1.1976	0.5332	0.7676
	1.2722	1.3417	1.2549	1.2688	1.3108	1.1963	1.1135	0.7364	0.6321	0.7927	0.6969	0.7024
	0.1931	0.1911	0.1953	0.2499	0.1955	0.2504	0.2788	0.0367	0.2606	0.0372	0.0353	0.0341
	1.3182	1.3703	1.2877	1.2703	1.146	1.1318	0.835	0.795	0.8511	0.6655	0.7269	0.5065
	0.2195	0.2109	0.2432	0.2154	0.2664	0.3032	0.2742	0.2632	0.2726	0.2185	0.0358	0.1319
2	1.2419	1.3776	1.3284	1.3824	1.39	0.5611	0.6069	0.5993	0.5024	0.5589	0.4992	0.5577
	1.3503	1.2482	1.2885	1.3516	1.2877	1.1747	1.359	1.1652	0.6642	0.7091	0.7036	0.4187
	0.1843	0.197	0.2285	0.2137	0.2134	0.1381	0.1629	0.2175	0.1336	0.15	0.0371	0.0315
	1.3522	1.3257	1.2825	1.2241	1.242	1.2744	0.7541	0.7718	0.7755	0.7431	0.5811	0.6601
	1.2783	1.2796	1.3488	1.3138	1.2053	0.7175	0.7271	0.7292	1.1616	0.7008	0.5735	0.4433
	0.1825	0.1942	0.249	0.2314	0.2844	0.1797	0.237	0.221	0.1745	0.0364	0.0353	0.0336
	1.3359	1.2786	1.223	1.3289	0.7197	1.2809	0.7584	0.748	0.7478	0.6915	0.7785	0.7215
	0.1321	0.1715	0.2045	0.2037	0.2348	0.2073	0.2157	0.2468	0.0338	0.0356	0.1435	0.0345

4	1.2991	1.3545	1.2847	1.2984	1.3054	0.8186	0.5503	0.5413	0.5433	0.6221	1.1444	0.4798
	1.3744	1.2338	1.298	1.4485	1.316	0.8319	0.8297	0.6941	0.6899	0.7037	0.84	0.6847
	0.2195	0.2643	0.2383	0.2399	0.1583	0.1349	0.1334	0.0367	0.1428	0.1401	0.1901	0.1076
	1.2704	1.3447	1.3152	1.3424	1.415	0.8748	1.3051	0.7629	1.2366	1.188	0.8185	0.8266
	1.3218	1.3963	1.2552	1.2688	1.4008	1.3048	1.2337	0.819	0.8021	0.7831	0.7073	0.8556
	0.2715	0.2789	0.2654	0.283	0.2211	0.179	0.161	0.1401	0.1151	0.0358	0.1489	0.0343
	1.3555	1.3639	1.3847	1.3162	1.1816	1.2708	0.737	0.8359	1.167	0.5448	0.6688	0.7638
	0.3085	0.3183	0.2926	0.2062	0.2778	0.1596	0.1651	0.2492	0.1697	0.0361	0.0351	0.1388
5	1.2975	1.2532	1.3006	1.259	1.3096	1.3168	1.2302	0.7221	0.6427	0.5943	0.7526	0.7244
	1.2782	1.2691	1.3213	1.3265	1.4085	1.2852	1.2998	1.1802	1.2897	0.7345	0.7312	0.6307
	0.2958	0.3145	0.255	0.2554	0.1931	0.17	0.1352	0.1562	0.034	0.1156	0.1446	0.1013
	1.3876	1.3283	1.3283	1.3621	1.3489	1.3967	0.7723	0.7886	0.7409	1.276	0.9877	0.7046
	1.3081	1.3109	1.431	1.2424	0.7486	0.7646	0.7544	0.6681	0.7204	0.7853	0.671	0.8073
	0.2887	0.2976	0.2691	0.2632	0.2228	0.189	0.1491	0.1409	0.0355	0.0365	0.0347	0.0336
	1.28	1.3378	1.3107	1.3691	0.7618	0.7445	0.7364	0.8467	0.7454	0.666	0.8118	0.9228
	0.2691	0.2406	0.2643	0.2269	0.2565	0.2239	0.1757	0.1445	0.234	0.0355	0.035	0.0346

7	1.2147	1.3129	1.2985	1.3438	1.4195	1.2533	1.0728	0.5377	0.8216	0.6361	0.6574	0.7009
	1.3098	1.3474	1.304	1.3739	1.2642	1.3275	0.7242	1.2302	0.7837	0.7708	0.6868	0.6566
	0.252	0.2579	0.2438	0.2458	0.2237	0.1948	0.1869	0.1622	0.1598	0.0345	0.1408	0.0349
	1.3291	1.3081	1.3501	1.3559	1.348	0.7703	0.7352	0.7826	0.7596	0.7918	0.7731	0.7202
	1.3085	1.3208	1.4296	1.3464	1.2076	1.2983	0.7622	0.7422	0.7555	0.7679	0.7676	0.5814
	0.2824	0.2693	0.2623	0.3178	0.2449	0.2313	0.1771	0.171	0.0346	0.0339	0.0347	0.0348
	1.3139	1.1991	1.2423	1.3649	1.3081	0.7172	0.7022	0.7037	0.7394	0.6433	0.7328	0.7072
	0.2129	0.1979	0.2158	0.2401	0.2481	0.182	0.129	0.119	0.1973	0.0339	0.035	0.0658
8	1.3062	1.4037	1.3987	1.2815	0.9306	0.8858	0.7385	1.1237	0.7597	0.721	0.8601	0.8847
	1.3127	1.3798	1.3669	1.3205	0.9577	1.3259	1.3293	0.7734	0.85	0.7818	0.7257	0.8573
	0.2457	0.2585	0.2228	0.2547	0.2508	0.209	0.1863	0.1532	0.1463	0.0352	0.0376	0.1241
	1.3568	1.3999	1.336	1.3027	1.3622	0.941	0.9472	0.7832	0.8762	0.6541	1.0464	0.787
	1.2898	1.3845	1.2992	1.3504	1.2765	1.1743	1.1432	0.9909	0.9202	0.806	0.7612	0.5233
	0.2578	0.2552	0.2843	0.23	0.2324	0.2304	0.2643	0.0369	0.1663	0.1973	0.0409	0.0497
	1.2719	1.4082	1.3082	1.3656	0.7501	1.3066	0.8002	1.1859	0.9133	0.7882	0.7587	0.834
	0.2316	0.1718	0.2269	0.2441	0.2671	0.2341	0.1537	0.185	0.1898	0.0372	0.0371	0.0353

10	1.235	1.3234	1.3013	1.334	1.2986	1.2583	0.7407	0.663	0.6902	0.9007	0.8389	0.8216
	1.334	1.307	1.3303	1.3668	1.1618	1.3318	1.1354	0.7404	1.252	0.8439	0.8833	0.8955
	0.3081	0.3244	0.305	0.1585	0.1262	0.2116	0.3188	0.2409	0.1552	0.175	0.0375	0.1173
	1.1545	1.3493	1.1039	1.3429	1.3471	1.3157	0.7376	0.9139	0.8536	0.8341	0.8058	0.8465
	1.359	1.3348	1.3533	1.3483	1.3913	1.3285	1.2909	1.2745	0.8581	1.3184	0.8184	0.8149
	0.1812	0.1604	0.1362	0.2969	0.3034	0.2564	0.2409	0.1873	0.1544	0.0363	0.1573	0.0346
	1.1602	1.3143	1.1189	1.3416	1.3085	0.7981	1.2322	1.257	0.8985	0.7371	0.9175	0.9962
	0.2106	0.282	0.3186	0.2037	0.4181	0.3574	0.2518	0.2452	0.199	0.0358	0.0352	0.0348
11	1.2836	1.3234	1.3404	1.3359	1.3143	0.8201	1.2215	0.6956	0.7434	0.633	0.6809	0.6919
	1.3036	1.2962	1.2198	1.3241	1.3436	0.7546	0.7997	0.8494	0.7519	0.7773	0.7131	0.8303
	0.2247	0.2487	0.2493	0.3017	0.2836	0.1694	0.1763	0.1611	0.1183	0.0349	0.0376	0.0339
	1.2858	1.1871	1.3578	1.351	1.3774	0.8111	0.7315	0.8075	0.794	0.7933	0.7495	0.7125
	1.2733	1.0059	1.3198	1.3429	1.3539	1.3179	0.7834	0.7365	0.7435	0.75	0.7828	0.8171
	0.2927	0.2226	0.2155	0.152	0.2739	0.2298	0.3359	0.225	0.1586	0.1624	0.1408	0.0341
	1.2544	1.3431	1.3417	1.3603	1.3471	1.2593	0.7762	0.7123	0.7363	0.7172	0.6857	0.8553
	0.1141	0.2	0.2141	0.1879	0.2709	0.0964	0.2194	0.2468	0.2037	0.0359	0.0354	0.0346

13	1.324	1.3547	1.2191	1.2031	1.1936	0.9274	1.167	0.7837	0.7183	0.7762	0.7225	0.8544
	1.3349	1.3355	1.3548	1.3394	1.2932	1.2743	1.2078	0.7814	0.8341	0.657	0.8393	0.96
	0.1999	0.2462	0.2304	0.219	0.2021	0.1488	0.1716	0.1819	0.2545	0.034	0.0379	0.0329
	1.3694	1.3319	1.326	1.3423	1.3995	1.3535	0.9309	0.7918	0.9124	0.876	1.1828	0.8846
	1.3041	1.3467	1.3293	1.2918	1.3189	1.2127	1.2136	1.1877	0.77	0.8222	0.9004	0.9126
	0.2004	0.2429	0.2388	0.2385	0.2259	0.1985	0.2387	0.0355	0.0353	0.0363	0.039	0.0727
	1.4016	1.3597	1.3829	1.3606	0.7951	0.8919	0.8226	0.7428	0.7998	0.8405	0.7418	0.8864
	0.1327	0.2055	0.2101	0.1992	0.2094	0.1843	0.0348	0.035	0.1807	0.0363	0.0378	0.0349
14	1.2877	1.3335	1.283	1.259	1.3613	1.1825	0.7272	0.7011	0.7519	0.7138	0.6407	0.8319
	1.3443	1.2327	1.2759	1.3105	1.2583	1.1491	0.7758	0.7561	0.7644	0.7363	0.711	0.6869
	0.1984	0.2067	0.1992	0.2165	0.1569	0.1433	0.1355	0.1448	0.1227	0.0345	0.1588	0.1146
	1.3334	1.3344	1.3288	1.3236	1.3276	1.1184	0.7695	0.7628	0.7592	0.7417	0.742	0.7367
	1.3252	1.3535	1.338	1.3088	1.2101	1.2766	0.7797	1.2254	0.6934	0.7224	0.7048	0.7102
	0.1975	0.2253	0.1985	0.2053	0.1623	0.1745	0.1833	0.0358	0.208	0.0356	0.035	0.1184
	1.3517	1.3461	1.3244	1.2921	0.7951	0.7051	0.7467	0.7583	0.7641	0.705	0.6963	0.7774
	0.137	0.1297	0.2066	0.182	0.1762	0.1846	0.1556	0.0351	0.0338	0.1324	0.0349	0.0362

16	1.3336	1.383	1.3313	1.2648	1.2783	1.209	1.2636	1.1931	0.7291	0.6849	0.7271	0.7256
	1.3401	1.3411	1.3399	1.3125	1.3226	0.7834	0.7653	0.7772	0.7915	0.7237	0.7791	0.6653
	0.2533	0.2957	0.2575	0.2563	0.1917	0.1478	0.1524	0.1904	0.1278	0.1881	0.166	0.1342
	1.3457	1.3549	1.3641	1.3535	1.2549	0.7738	0.7782	0.8073	1.2796	0.7951	0.7907	0.7004
	1.3356	1.3357	1.337	1.3537	1.3481	1.2927	0.7476	0.7402	0.7715	0.7728	0.7608	0.7686
	0.2268	0.2721	0.2612	0.2837	0.2348	0.2045	0.251	0.2046	0.0349	0.0358	0.035	0.0338
	1.3297	1.3963	1.3708	1.3933	0.8168	1.435	0.7093	0.772	0.7249	0.7285	0.7741	0.7334
	0.1628	0.1688	0.1977	0.1771	0.2271	0.1697	0.1404	0.1579	0.0343	0.0356	0.0351	0.1296
17	1.3279	1.3425	1.3534	1.3146	0.8565	1.3295	0.7389	0.6992	0.7183	0.7088	0.7088	0.7088
	1.3686	1.3712	1.3326	1.2785	1.1012	0.6119	0.784	0.7443	0.7555	0.7359	0.7359	0.7359
	0.2918	0.2822	0.2381	0.2554	0.2064	0.1351	0.1266	0.1282	0.1388	0.1629	0.1629	0.1629
	1.3609	1.3875	1.3682	1.375	1.4531	0.8688	0.8079	0.8325	0.8237	1.2558	1.2558	1.2558
	1.375	1.3917	1.3691	1.3834	1.3719	1.3342	0.8744	0.8999	0.804	0.8583	0.8583	0.8583
	0.3363	0.2666	0.3274	0.2702	0.2361	0.2246	0.2084	0.1983	0.1743	0.0347	0.0347	0.0347
	1.3392	1.3939	1.3552	1.3642	0.8636	0.8592	0.7661	0.8826	0.7938	0.626	0.626	0.626
	0.2797	0.2229	0.2448	0.2306	0.2788	0.1803	0.1686	0.05	0.0357	0.626	0.626	0.626

19	1.2913	1.3216	1.2627	1.2464	1.2078	1.2009	0.7416	0.7402	0.7552	0.7743	0.7178	0.7266
	1.3408	1.2921	1.3259	1.3405	1.3275	1.3	0.7569	1.3095	0.7588	0.718	0.7554	0.7795
	0.1929	0.2285	0.2007	0.1969	0.1618	0.1319	0.119	0.1602	0.1085	0.1432	0.037	0.0328
	1.3338	1.3354	1.3437	1.3337	1.3405	1.2379	0.7876	0.6683	0.7707	0.7741	0.6961	0.7487
	1.3245	1.3481	1.3709	1.3617	1.3517	0.756	0.7649	0.7892	0.757	0.766	0.7213	0.7869
	0.2233	0.2328	0.2261	0.2192	0.2054	0.1553	0.1442	0.1185	0.1177	0.0354	0.0353	0.0339
	1.3722	1.3209	1.314	1.3806	1.3032	1.2773	1.2326	0.8046	0.7294	0.7105	0.6914	0.7546
	0.1485	0.1911	0.1737	0.2025	0.2069	0.1914	0.042	0.035	0.0337	0.0363	0.0364	0.0398
20	1.3567	1.3406	1.3299	0.8311	1.3325	1.288	0.7028	0.7472	0.6841	0.6193	0.7367	0.8088
	1.3602	1.3607	1.3749	1.4092	1.4087	1.3919	1.2164	0.8033	0.8704	0.8646	0.8345	0.8736
	0.2215	0.211	0.2027	0.165	0.1795	0.1471	0.138	0.1439	0.0344	0.0344	0.1118	0.034
	1.3346	1.3783	1.3558	1.4185	1.3672	0.7289	1.2568	0.8442	0.7434	0.7383	0.7268	0.7591
	1.3558	1.3708	1.4114	1.3694	1.3961	0.7001	0.7099	0.761	1.1725	0.7942	0.8078	0.7825
	0.2104	0.2383	0.1985	0.2021	0.2168	0.2088	0.1584	0.1534	0.0354	0.1078	0.035	0.0347
	1.3548	1.3692	1.3614	1.3619	0.8194	0.8062	0.7979	0.7597	0.681	0.7209	0.7625	1.2516
	0.2228	0.2718	0.2695	0.2771	0.2111	0.1781	0.1339	0.1345	0.0354	0.1326	0.0359	0.036

Absorbance at 620 nm for Blanks – Plates 1 through 20 for 5/12/03

3	0.8283	0.5055	0.7148	0.7805	0.6941	0.6591	0.4516	0.7344	0.6901	0.5954	0.6107	0.5556
	0.0371	0.0368	0.0364	0.034	0.0367	0.0368	0.037	0.0374	0.0347	0.0337	0.0352	0.0347
	0.5976	0.6618	0.7036	0.7151	0.7408	0.6947	0.7018	0.7278	0.7264	0.7048	0.688	0.462
	0.0397	0.0372	0.036	0.036	0.0377	0.0362	0.0371	0.0363	0.0351	0.0378	0.0375	0.0357
	0.4392	0.6509	0.6542	0.7438	0.7538	0.7576	0.7554	0.7178	0.7732	0.7018	0.7169	0.4832
	0.0353	0.0354	0.0353	0.0352	0.0347	0.0348	0.0373	0.036	0.0357	0.0359	0.0351	0.034
	0.0465	0.0462	0.0449	0.0446	0.0456	0.0529	0.0455	0.0476	0.0448	0.0461	0.0486	0.0457
	0.0454	0.0457	0.048	0.046	0.0455	0.0459	0.0453	0.0464	0.0446	0.0474	0.0454	0.0445
6	0.3824	0.5378	0.7284	0.6114	0.5563	0.6115	0.5361	0.4887	0.6846	0.7015	0.7952	0.7151
	0.0344	0.0354	0.0356	0.034	0.0358	0.0371	0.0367	0.0365	0.0344	0.0338	0.0353	0.0348
	0.5615	0.6467	0.7063	0.7268	0.7424	0.7661	0.6817	0.7385	0.667	0.6722	0.7524	0.5098
	0.0354	0.0356	0.0371	0.0353	0.0385	0.0368	0.0358	0.0356	0.0352	0.0376	0.0382	0.0354
	0.5317	0.7843	0.7492	0.793	0.7135	0.7114	0.7247	0.6885	0.7534	0.8192	0.8944	0.8041
	0.035	0.035	0.0365	0.0366	0.0348	0.0347	0.0362	0.0358	0.0355	0.036	0.035	0.09
	0.0463	0.0457	0.0448	0.0438	0.045	0.0527	0.0453	0.0473	0.045	0.0462	0.048	0.0456
	0.0452	0.0454	0.048	0.0456	0.0456	0.0453	0.0447	0.0461	0.0441	0.049	0.0447	0.0445

9	0.67	0.811	0.7318	0.5481	0.7214	0.6213	0.7082	0.8422	0.7818	0.8466	0.7551	0.7681
	0.7894	0.7732	0.776	0.7939	0.7851	0.7738	0.0354	0.0343	0.0353	0.0351	0.0357	0.0351
	0.6447	0.7307	0.7208	0.7094	0.7356	0.7599	0.7633	0.7502	0.7588	0.8012	0.6868	0.5854
	0.0355	0.0349	0.0352	0.0359	0.0378	0.0367	0.0347	0.0357	0.036	0.0352	0.0366	0.0371
	0.6096	0.671	0.7772	0.6841	0.7451	0.836	0.7488	0.7649	0.7755	0.8083	0.7381	0.5413
	0.0355	0.035	0.0391	0.0383	0.0355	0.0353	0.0353	0.038	0.0347	0.0342	0.0349	0.0352
	0.0457	0.0459	0.0463	0.0461	0.0458	0.0474	0.0455	0.0468	0.0457	0.045	0.0495	0.0464
	0.0449	0.0455	0.0474	0.0468	0.0473	0.049	0.0457	0.0455	0.0463	0.0445	0.0459	0.0454
12	0.7865	0.8245	0.7576	0.6086	0.7463	0.7168	0.7182	0.8456	0.8342	0.6705	0.6641	0.7324
	0.3105	0.2386	0.2302	0.1954	0.0353	0.0371	0.0366	0.0369	0.0337	0.0341	0.0355	0.0347
	0.7867	0.6891	0.8425	0.7665	0.7317	0.7712	0.7703	0.7646	0.8056	0.8318	0.7245	0.7814
	0.2479	0.2135	0.2482	0.2803	0.1206	0.057	0.0354	0.0521	0.0354	0.0366	0.042	0.0354
	0.6623	0.7895	0.7327	0.7294	0.7617	0.7953	0.7759	0.7889	0.7649	0.7409	0.7041	0.6736
	0.3014	0.2893	0.3134	0.1348	0.3616	0.0757	0.0365	0.1281	0.0354	0.0352	0.0358	0.0345
	0.047	0.0456	0.0452	0.0446	0.0457	0.0528	0.0455	0.0477	0.0448	0.0462	0.0487	0.0458
	0.045	0.0455	0.0479	0.0468	0.0451	0.045	0.0449	0.0461	0.0442	0.0488	0.0449	0.0441

15	0.6645	0.6771	0.6577	0.7413	0.7917	0.6981	0.7559	0.6775	0.7404	0.5985	0.688	0.6685
	0.0342	0.035	0.0357	0.0338	0.0357	0.0365	0.0366	0.0364	0.0337	0.0338	0.0351	0.0357
	0.7275	0.7716	0.7497	0.7394	0.7412	0.7403	0.7734	0.7555	0.751	0.7184	0.7094	0.7031
	0.0354	0.0359	0.0351	0.0354	0.0371	0.0388	0.0356	0.0357	0.0351	0.0365	0.0374	0.0355
	0.6356	0.6682	0.7668	0.6851	0.7431	0.7217	0.7494	0.7275	0.8009	0.7646	0.6396	0.6615
	0.0347	0.0347	0.0406	0.0349	0.0345	0.0348	0.0362	0.0352	0.0353	0.0357	0.0356	0.0343
	0.0475	0.0458	0.0451	0.0447	0.0456	0.0528	0.0457	0.0474	0.0451	0.0462	0.048	0.0465
	0.0354	0.0354	0.0482	0.0463	0.0458	0.0455	0.0451	0.0473	0.0443	0.0475	0.0466	0.0463
18	0.6336	0.6164	0.6762	0.6903	0.6488	0.7028	0.7008	0.6689	0.7421	0.7571	0.8249	0.7381
	0.034	0.0353	0.0365	0.0338	0.0352	0.0366	0.0363	0.0368	0.0343	0.034	0.0353	0.0348
	0.7056	0.7319	0.7595	0.7578	0.7267	0.7301	0.794	0.7379	0.7303	0.7547	0.7608	0.7137
	0.0352	0.0359	0.0354	0.0358	0.0378	0.0373	0.0371	0.0358	0.0362	0.0381	0.038	0.036
	0.5737	0.7036	0.7596	0.7933	0.7515	0.7648	0.7625	0.7711	0.7595	0.739	0.7849	0.7133
	0.035	0.0349	0.0346	0.036	0.0346	0.0348	0.0365	0.0362	0.0354	0.0355	0.0351	0.0342
	0.0466	0.0459	0.0448	0.0444	0.0455	0.0532	0.0456	0.0477	0.0451	0.0464	0.0481	0.0457
	0.0469	0.046	0.0481	0.0458	0.0453	0.0449	0.0452	0.0462	0.0444	0.0473	0.0448	0.0444

21	0.5827	0.5871	0.6494	0.7422	0.6244	0.5651	0.7198	0.7426	0.6921	0.7969	0.6698	0.6481
	0.0345	0.0353	0.0355	0.0338	0.0354	0.0367	0.0371	0.036	0.0337	0.034	0.0355	0.0894
	0.6235	0.6871	0.6358	0.6887	0.7497	0.7189	0.7151	0.7135	0.6988	0.7532	0.6847	0.7281
	0.0349	0.0356	0.0352	0.0352	0.0377	0.0364	0.0353	0.0352	0.0348	0.0364	0.0377	0.0361
	0.6082	0.6894	0.7368	0.6931	0.7422	0.7077	0.7418	0.7273	0.7341	0.7467	0.7725	0.6724
	0.034	0.0345	0.0358	0.0353	0.0342	0.0347	0.0366	0.0356	0.0373	0.0359	0.0357	0.0427
	0.0464	0.0455	0.0446	0.0439	0.0449	0.0529	0.0453	0.0479	0.0449	0.0459	0.0479	0.0456
	0.0458	0.0457	0.0482	0.0459	0.0458	0.0458	0.045	0.0463	0.0441	0.0477	0.0449	0.0472

Date	1/29/03							
Sample	CAS Settled MLSS							
Size Fraction		Whole	< 1.5 um	< 0.22 um	< 100 kD	< 30 kD	< 10 kD	< 1 kD
Abs. at 280 nm								
Replicate 1		0.1007	0.0931	0.0867	0.087	0.0862	0.0851	0.0404
Replicate 2		0.0995	0.0929	0.0867	0.0868	0.0864	0.0853	0.0408
Replicate 3		0.1021	0.0928	0.0868	0.0868	0.0863	0.085	0.0404
Average		0.1008	0.0929	0.0867	0.0869	0.0863	0.0851	0.0405
TOC Measurements (mg/L)								
Replicate 1		9.29	8.38	7.34	6.89	6.33	5.59	2.98
Replicate 2		9.29	8.48	7.56	6.9	6.39	5.77	3.02
Replicate 3		9.33	8.35	7.53	7	6.4	5.81	2.99
Average		9.3033	8.4033	7.4767	6.9300	6.3733	5.7233	2.9967
Standard Deviation		0.023094	0.0680686	0.1193035	0.06083	0.0379	0.1172	0.0208
95% Confidence Interval		0.0261328	0.0770253	0.135002	0.06883	0.0428	0.1326	0.0236
Calculated COC (mg/L)		6.3067	5.4067	4.4800	3.9333	3.3767	2.7267	2.9967
Calculated e280 (L/mol cm)								
Average		192	206	232	265	307	375	162
Standard Deviation		0.0131502	0.0082653	0.0159707	0.00888	0.0061	0.0206	0.009
95% Confidence Interval		0.0148806	0.0093529	0.0180721	0.01005	0.0068	0.0233	0.0102

Date	3/27/03							
Sample	CAS Settled MLSS							
Size Fraction		Whole	< 1.5 um	< 0.22 um	< 100 kD	< 30 kD	< 10 kD	< 1 kD
Abs. at 280 nm								
Replicate 1		0.2811	0.1704	0.0702	0.1031	0.0615	0.0566	0.0304
Replicate 2		0.2839	0.1726	0.0701	0.1032	0.0617	0.0567	0.0305
Replicate 3		0.282	0.169	0.0701	0.1032	0.0617	0.0567	0.0304
Average		0.2823	0.1707	0.0701	0.1032	0.0616	0.0567	0.0304
TOC Measurements (mg/L)								
Replicate 1		14.1	8.21	4.87	5.35	3.66	3.61	1.92
Replicate 2		16.2	8.95	5.26	5.84	3.73	3.66	1.93
Replicate 3		16.1	8.97	5.27	5.88	3.72	3.67	1.94
Average		15.4667	8.7100	5.1333	5.6900	3.7033	3.6467	1.9300
Standard Deviation		1.1846237	0.4331282	0.2281082	0.29513	0.0379	0.0321	0.01
95% Confidence Interval		1.3405013	0.4901209	0.2581236	0.33396	0.0428	0.0364	0.0113
Calculated COC (mg/L)		13.5367	6.7800	3.2033	3.7600	1.7733	1.7167	1.9300
Calculated e280 (L/mol cm)								
Average		250	302	263	329	417	396	189
Standard Deviation		0.0767592	0.0508518	0.0444443	0.05187	0.0104	0.0089	0.0055
95% Confidence Interval		0.0868595	0.0575431	0.0502924	0.0587	0.0118	0.01	0.0062

Date	4/2/03							
Sample	CAS Settled MLSS							
Size Fraction		Whole	< 1.5 um	< 0.22 um	< 100 kD	< 30 kD	< 10 kD	< 1 kD
Abs. at 280 nm								
Replicate 1		0.2983476	0.0988298	0.0727166	0.07073	0.0718	0.0653	0.0319
Replicate 2		0.3042801	0.098086	0.0727166	0.0703	0.0713	0.0651	0.032
Replicate 3		0.2947367	0.0985436	0.072447	0.07041	0.0718	0.0648	0.0318
Average		0.2991	0.0985	0.0726	0.0705	0.0716	0.0651	0.0319
TOC Measurements (mg/L)								
Replicate 1		14	5.19	4.33	4.22	4.12	3.89	1.85
Replicate 2		15.2	5.24	4.49	4.33	4.23	3.99	1.83
Replicate 3		15.4	5.2	4.48	4.32	4.24	3.99	1.76
Average		14.8667	5.2100	4.4333	4.2900	4.1967	3.9567	1.8133
Standard Deviation		0.7571878	0.0264575	0.0896289	0.06083	0.0666	0.0577	0.0473
95% Confidence Interval		0.8568216	0.0299389	0.1014226	0.06883	0.0753	0.0653	0.0535
Calculated COC (mg/L)		13.0533	3.3967	2.6200	2.4767	2.3833	2.1433	1.8133
Calculated e280 (L/mol cm)								
Average		275	348	333	341	361	364	211
Standard Deviation		0.0534187	0.0063482	0.0203303	0.01453	0.0162	0.0151	0.0262
95% Confidence Interval		0.0604477	0.0071835	0.0230054	0.01644	0.0184	0.017	0.0296

Date	5/12/03							
Sample	CAS Settled MLSS							
Size Fraction		Whole	< 1.5 um	< 0.22 um	< 100 kD	< 30 kD	< 10 kD	< 1 kD
Abs. at 280 nm								
Replicate 1		0.0862	0.091564	0.0532878	0.0900	0.0706	0.0688	0.0158
Replicate 2		0.1077	0.0920203	0.05153	0.0853	0.0729	0.0687	0.0161
Replicate 3		0.1364	0.1154697	0.0516	0.0837	0.0702	0.0692	0.0284
Average		0.1101	0.0997	0.0521	0.0863	0.0713	0.0689	0.0201
TOC Measurements (mg/L)								
Replicate 1		6.878	4.752	5.279	4.176	4.134	4.036	1.826
Replicate 2		6.732	4.675	5.273	4.136	4.169	4.083	1.787
Replicate 3		7.37	4.697	5.292	4.136	4.161	4.014	1.789
Average		6.9933	4.7080	5.2813	4.1493	4.1547	4.0443	1.8007
Standard Deviation		0.3342713	0.0396611	0.0097125	0.02309	0.0183	0.0352	0.022
95% Confidence Interval		0.3782561	0.0448798	0.0109905	0.02613	0.0208	0.0399	0.0249
Calculated COC (mg/L)		5.1927	2.9073	3.4807	2.3487	2.3540	2.2437	1.8007
Calculated e280 (L/mol cm)								
Average		254	411	180	441	363	369	134

VITA

R. David Holbrook

R. David Holbrook was born in Boston, MA, lived briefly in Concord, MA and Rochester, NY before settling in Vestal, NY, where he graduated from Vestal High School in 1988. Dave's undergraduate studies were completed at Cornell University, where he received a B.S. in Agricultural and Biological Engineering in 1992. He remained at Cornell for his master's degree and received a M.Eng degree in Civil (Environmental) Engineering in 1993. After finishing at Cornell, Dave was employed by I. Krüger, Inc. (now Krüger, Inc., part of USFilter, Inc.) as a process engineer where he learned how to design and operate a wide array of wastewater treatment processes including biological nutrient removal systems, biological aerated filters, pure oxygen systems, and thermophilic aerobic digestion. In 1999, Dave left Krüger to start his graduate work at in the Department of Civil and Environmental Engineering at Virginia Tech. At Virginia Tech, he became interested in trace organic contaminant behavior in wastewater and natural systems. Dave defended his dissertation on July 22, 2003.