FACTORS INFLUENCING THE NITRIFICATION EFFICIENCY OF FLUIDIZED BED FILTERS WITH A PLASTIC BEAD MEDIUM

By

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ABSTRACT

Nitrification performance of three, fluidized-bed filters was investigated. Each filter contained 10 L of plastic bead medium (near neutral specific gravity and 2-4 mm diameter and length) and was loaded under conditions of various flow rates and ammonia levels. Bead settled depth and biofilter diameter (12.7, 15.2 and 17.8 cm) were the factors differing among the filters. The experiments were conducted with three replicate recirculating systems. Each system included one of the three different biofilter types, connected in parallel to a reservoir containing 500 L of water. Systems were allowed to acclimate using a synthetic nutrient substrate, which was followed by a comparative analysis of biofilter performance. To evaluate filter performance, ammonia inflow concentration, ammonia loading rates, nitrite, nitrate, temperature, pH, dissolved oxygen levels, hardness, alkalinity and flow rates were monitored. Initially, four different flow rates, ranging from 6 Lpm–12 Lpm, were tested at constant ammonia feed level (8.4 g/day). Here, biofilter D_3 (17.8 cm diameter) showed the best ammonia removal performance at a flow rate of 6 Lpm, followed by the performance of D_1 (12.7 Lpm) and D_2 (15.2 Lpm). The difference in ammonia and nitrite removal performance decreased among the biofilters, as flow rate increased. An increase in flow rate also lowered ammonia level in the systems at a constant ammonia loading, but did not affect the nitrite concentration. Five different ammonia feed rates, ranging from 8.4 – 16.8 g/day, were tested in the second part of the study, at a constant flow rate of 12 Lpm in each column. Different ammonia and nitrite removal performance was observed between biofilter sizes. Ammonia accumulated in the tanks as ammonia loading increased, but nitrite concentration remained relatively constant. The results indicated that nitrification performance improved by 17 % as the applied flow rate was increased. Ammonia concentration decreased slightly, from 0.6 mg/L to 0.5 mg/L. The performance appeared to be limited at higher ammonia loadings, at which time ammonia concentration increased from 0.5 to 0.99 mg/L. Data on biofilm development indicates a reduction in biofilm thickness as flow rate was increase, and significant biofilm accumulation as ammonia supply increased. The results of this work were compared to performance data generated using a steady state biofilm model, developed by Rittmann and McCarty (1980). The model predicted higher values of biofilm thickness (L_f) than those seen in this study.

Fluidized bed filters with plastic bead medium proved to be effective in removing ammonia and nitrite from a synthetic aquaculture water.

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CHAPTER I

BIOFILTER DYNAMICS IN RECIRCULATING AQUACULTURE SYSTEMS

A. RECIRCULATING AQUACULTURE

Seafood consumption is rising worldwide while the available natural catch is diminishing because of aggressive harvesting practices (Youngs and Timmons, 1991). Aquaculture production of catfish ponds in the southern United States, salmon in northern United States and Canada, tilapia in recirculating indoor systems and shrimp culture in South America have helped to fill the void by providing a dependable high quality supply of seafood. The culture systems employed have proven themselves economical, however, they require a large amount of high quality water, which is finite in supply. Clean water supply whether from surface or ground water sources becomes a critical issue (Easter, 1992). Additionally, concerns regarding detrimental impacts of aquaculture production to the environment (Rosenthal, 1994), increased regulations on aquaculture effluents, and the need to conserve water resources (Klontz, 1979) and energy has pushed the aquaculture industry to focus on development and refinement of water recycling technologies, particularly recirculating aquaculture systems (RAS). These systems can generally be defined as an assemblage of parts used for the husbandry of aquatic organisms in which water is continuously cleaned and recycled (Libey, 1996). RAS offer solutions for intensification of fish culture controlling the water quality. Water is cleaned via mechanical and biological filtration. Mechanical filtration removes particulates, while biological filtration removes dissolved wastes via biochemical reactions that occur during bacterial metabolism (Hall, 1999). These processes allow water to be cleaned and reused several times prior to discharge. These processes conserve water by reducing the amount of water needed (from an external source) to maintain a biologically suitable culture environment. Water recycling allows the majority of recirculating systems to exchange approximately 10% of total system volume per day

while recycling 90% of the culture water. Owsley (1993) reported five aquaculture facilities maintaining a daily exchange rate of approximately 10%, and Westerman et al. (1996) reported 9 to 11% exchange rates for four recirculating systems employed in a filter evaluation study.

In addition to water conservation, recirculating systems allow large fish yields to be obtained in a relatively small area, enhance environmental control, and increase growth on a year around production schedule, thereby avoiding the seasonal limitations suffered in outdoor systems (Van Gorder, 1994).

B. BIOFILTRATION AND NITRIFICATION

Biological filtration in the broadest sense includes any filtration technique that utilizes biological (living) organisms to remove impurities from the water (Wheaton et al., 1990). Although biological filtration can include living plant filters, nitrification, denitrification, extended aeration systems and a host of other types of filters or unit processes, only ammonia (NH_3) removal (nitrification) process will receive consideration in this section.

In aquaculture, biofiltration is an important process employed by water recirculating systems. In order to maintain a clean environment, it is necessary to remove ammonia, a toxic compound released into the water column as a product of the animal metabolism. Although less toxic than ammonia, nitrite (NO_2^-) are toxic to fish, while nitrate (NO_3^-), the final oxidized form in nitrification, are relatively nontoxic to fish unless high concentrations are sustained for an extended period of time (Spotte, 1979).

One of the most useful techniques in treating ammonia-laden water (particularly at low levels of ammonia) has been the biological fixed film process (Brune and Gunther, 1981). Biological growth occurs on the surface of the media and the attached film of microorganisms oxidizes the dissolved ammonia. This transformation is known as nitrification.

Kinetics

To ensure prolonged fish survival, high levels of sustained nitrification must be achieved. Therefore, ecological requirements of the bacteria must be met within biofilters for effective nitrification to occur (Malone et al., 1993). Autotrophic bacteria are credited for performing nitrification (Wedemeyer, 1996). The primary microorganisms responsible for this are two genera of chemolythotrophic bacteria in the family Nitrobacteraceae. *Nitrosomonas sp.*, which oxidize ammonia to nitrite, and *Nitrobacter sp.*, which oxidize nitrite to nitrate, are the most common and well-studied genera (Gaudy and Gaudy, 1988).

In addition to *Nitrosomonas*, other genera are known to oxidize ammonia, including *Nitrosospira sp., Nitrosogloea sp., Nitrosocystis sp.* and *Nitrosococcus sp.* (O'Shaughnessy et al., 1982).

Nitrosomonas and *Nitrobacter* are considered to be of primary importance in fixed-film nitrification of wastewater. These two genera are primarily autotrophic in that the only required source of carbon is inorganic CO_2, while they are capable of using "at least certain organic compounds", if available (Gaudy and Gaudy, 1988). They are chemotrophic in that the energy source is a chemical reduction/oxidation (redox) reaction and lithotrophic in that the electron donor in the redox reaction is an inorganic compound (nitrogen) (Easter, 1992).

Wheaton et al., (1990) presented a simplified schematic of nitrogen cycle (Figure 1.1). Within a nitrifying filter, the objective is to convert ammonia to nitrate (relatively non toxic). The way to prevent anaerobic conversion of nitrate to N_2 as shown in Figure 1.1., is to maintain dissolved oxygen levels that are greater than 2 – 3 mg/L.

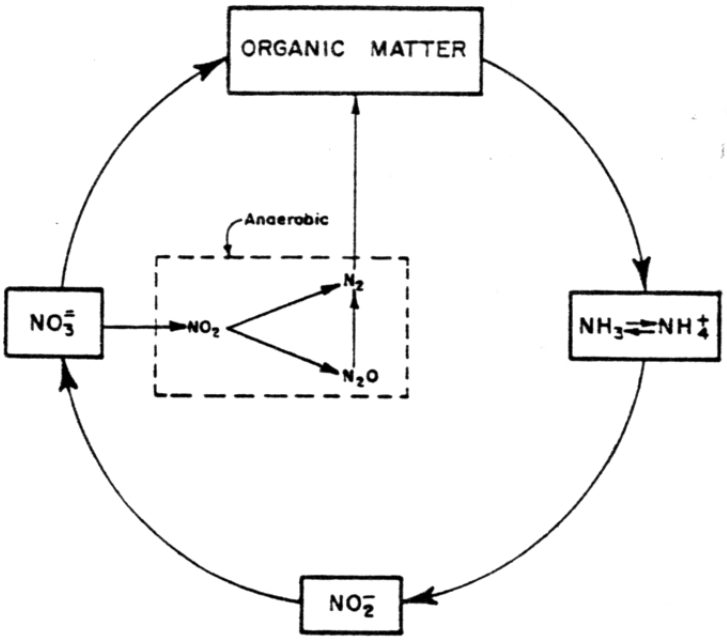

Figure 1.1. Nitrogen cycle: the reactions within the dotted box are anaerobic; all other reactions are aerobic (Wheaton et al., 1990).

Heterotrophic bacteria rely on organic compounds for their energy source, as compared to autotrophic bacteria that rely on inorganic compounds as an energy source. The quality of water in an aquacultural system, as measured by chemical oxygen demand (COD) will profoundly affect the nitrifying performance of a nitrification filter. Heterotrophic bacteria grow at a significantly greater rate than *Nitrosomonas* and *Nitrobacter* (Water Pollution Control Federation, 1983). Thus, attempting to combine the conversion of organic compounds and the inorganic forms of nitrogen in the same nitrification filter leads to competition for growing space between the heterotrophs and the autotrophs (Wheaton et al., 1990).

The denitrifying conversions of NO_3 to NO_2 (nitrate to nitrite), NO_3 to N_2O or N_2 (nitrate to dinitrogen oxide or nitrogen gas, respectively), and N_2O to N_2 occur only under anoxic conditions. Thus, these reactions are not expected to occur in a well-aerated filter.

Equations 1 and 2 show the basic chemical conversion occurring in a nitrification filter (USEPA, 1975: Water Pollution Control Federation, 1983).

$$NH_4^+ + 1.5\ O_2 \Rightarrow 2H^+ + H_2O + NO_2^- \qquad (1)$$

$$NO_2^- + 0.5\,O_2 \Rightarrow NO_3^- \qquad (2)$$

Energy released by the conversion described in Equations 1 and 2 is used by *Nitrosomonas* and *Nitrobacter* to drive their life process. Additionally, these reactions require oxygen, produce hydrogen ions (lowering pH), and produce nitrite as an intermediate product (Wheaton et al., 1990). Cell growth of *Nitrosomonas* and *Nitrobacter* are important considerations in nitrification filter design because large amounts of cell growth can clog some types of filters (e.g., trickling filters). Equation 3 describes cell growth and ammonia oxidation to nitrate (Water Pollution Control Federation, 1983; Gujer and Boller, 1986).

$$NH_4^+ + 1.83\,O_2 + 1.98\,HCO_3^- \Rightarrow 0.021\,C_5H_7O_2N + 0.98\,NO_3^- + 1.041\,H_2O + 1.88\,H_2CO_3^- \qquad (3)$$

From equation 3, oxygen and alkalinity requirements and cell biomass production can be calculated. For every gram of NH_4^+ -N oxidized to NO_3^- -N, 4.18 g of O_2 and 7.14 g of alkalinity (as Ca CO_3) are used, and 8.59 g of carbonic acid and 0.17 g of cells are produced.

The rate of oxidation of ammonia in a biological filter has been described by the Monod equation (Equation 4). The Monod equation approximates the curve that describes the relationship between the specific growth rate of bacteria and substrate concentration (Grady and Lim, 1980).

$$\mu_n = (\mu_n' \times S) / (S + K_s) \qquad (4)$$

μ_n = specific growth rate of nitrifiers (day^{-1})

μ_n' = maximum specific growth rate of microorganisms (l/day)

S = limiting substrate concentration

K_s = half saturation constant which is equal to the growth limiting substrate concentration at one-half the maximum growth rate (g/m^3 of NH_4^+ -N or NO_3^- -N)

If micronutrients requirements are met, nitrite is usually the growth limiting substrate of *Nitrobacter*, while ammonium is the limiting substrate of *Nitrosomonas*. Because the

growth rate of *Nitrobacter* is greater than that of *Nitrosomonas*, oxidation of ammonia usually is the rate-limiting step in the conversion of ammonia in nitrate (Water Pollution Control Federation, 1983). As such, the ammonia coefficients in Equation 4 for *Nitrosomonas* are the rate limiting parameters to use in describing nitrification.

Although nitrification can occur throughout the culture system (e.g., in biofilms, on pipe, and tank walls), the majority of biochemical reactions pertaining to heterotrophic and autotrophic bacteria occur within biofilters (Losordo, 1991).

C. BIOFILTERS

Biofilters, often referred as biological filters or nitrification filters, are commonly used in recirculating aquaculture production systems to remove ammonia and convert it first to nitrite and then into the less toxic nitrate. Biofilters are biological entities composed of some types of solid media on which nitrifying bacteria grow. The biofilters are specifically designed for concentrated bacteria attachment and nitrification via fixed-film kinetics. The nitrifying bacteria use energy released in the conversion of ammonia to nitrite to drive their life process. Since biofiltration is the principal unit process used for treating fish metabolites, biofilters can be considered major components in intensive recirculating aquaculture systems (Libey and Miller 1985). A biofilter's efficiency in ammonia removal is a key-limiting factor for the carrying capacity of the system (Zhu and Chen, 1999). They must remove the waste produced in the aquaculture system at a rate sufficient to maintain water quality at a level that prevents stress in the crop. Biofilters design is, in simple terms, a mass balance between the waste produced by the crop and the waste removed by the filter.

Many different biofilter configurations (e.g., downflow filters, rotating biological drums, and fluidized bed filters) are used in aquaculture systems (Wheaton et al., 1990).

1. Chemical Factors Affecting Filter Performances

1.1. pH

The optimum pH for nitrification filters is in the range of 6 to 9. However, for a specific filter the optimum range is narrower; the specific range being a function of adaptation of the bacteria in the filter to pH. Haug and McCarty (1972) observed dramatic reduction of nitrification in a fixed film system below pH 6 and cessation of nitrification at pH 5.5. Thus, maintaining pH levels above six is a recommended practice (Wheaton et al., 1990). However, since the percentage of unionized ammonia (the toxic form to fish) increases as pH increases, it is also a good practice to maintain a pH near the lower end of the optimum pH range for the nitrifying bacteria.

1.2. Alkalinity

The conversion of NH_4^+ to nitrate consumes alkalinity as shown in Equation 3. Malone and Burden (1988) in their work on pH and alkalinity in nitrification filters showed that bicarbonate alkalinity might be critical for nitrifiers growth. Alkalinity in the form of carbonate and bicarbonate is a nutrient for nitrifying bacteria. Alkalinity is also necessary to prevent pH changes due to acid production in the nitrification process (Equation 4).

1.3. Oxygen

Nitrification rates in nitrifying filters decrease when insufficient oxygen is available to the nitrifying bacteria (Kaiser and Wheaton, 1983; Nagel and Haworth, 1969). The limiting concentration of oxygen is a function of several variables including temperature, concentration of organics in the filter feed water, and bacterial biomass present. In practice, filters contain heterotrophs as well as nitrifiers. Because the nitrogen cycle moves from organic compounds to ammonia to nitrite to nitrate, a filter receiving culture water first tends to have a high percentage of heterotrophs. Further, into the filter, the *Nitrosomonas* population increases to remove the ammonia, and further still into the filter, the *Nitrobacter* population increases, as nitrite concentrations increases. Position of bacterial strains in the filter dictates that oxygen is used first by the heterotrophs, then by

the *Nitrosomonas* and then by *Nitrobacter*. In filters where the water must supply all of the oxygen, *Nitrobacter* and /or *Nitrosomonas* often become oxygen starved.

Most information on limiting oxygen concentration in nitrification filters comes from research on municipal wastewater treatment. Some authors suggest, based primarily on stochiometric requirements, that oxygen concentrations above 1 mg/L are sufficient (Water Pollution Control Federation, 1983). However, the same reference states that 2 mg/l and above is a safe operating level for wastewater nitrification filters. Thus, aquacultural nitrification filters are often ammonia or nitrite limited, rather than oxygen limited. If this is the case, increasing oxygen levels will have little effect on filter performance until oxygen becomes the limiting factor.

1.4. Ammonia / Nitrite Concentrations

It has been shown that excessively high ammonia and/or nitrite concentrations are toxic to nitrification filter operation. Anthonie (1976) concluded that nitrous acid and un-ionozed ammonia (NH_3) were the inhibitory agents. Unionized ammonia inhibits *Nitrosomonas* at concentrations higher than those that inhibit *Nitrobacter* (10 – 150 mg/L of unionized ammonia as compared to 0.1 – 1.0 mg/L of unionized ammonia, respectively). Nitrous acid was inhibitory at concentrations of 0.22 to 218 mg/L. It appears that acclimation to unionized ammonia and/or to nitrous acid, temperature, and the number of active nitrifiers also influence the inhibitory effects of unionized ammonia and nitrous acid (Hochheimer, 1990).

Aquacultural nitrification filters operate at very low ammonia and nitrite concentrations as compared to wastewater nitrification filters. The ammonia and/or nitrite concentrations often are so low, that these compounds become the rate-limiting factor in filter nitrification (Hochheimer, 1990). Thus, the availability of ammonia and/or nitrite may determine the rate of nitrification in an aquacultural nitrification filter, rather than oxygen as is often assumed.

2. Physical Factors

2.1. Temperature

There appears to be considerable disagreement in the literature concerning the specific effects of temperature on nitrification. For example, Jones and Morita (1985) isolated an ammonia oxidizing bacteria capable of growth at -5°C. Optimal growth for these cells occurred at 22°C, while their lethal temperature was about 29°C. However, bacteria adapted to 25°C had an optimal temperature of 30°C and a lethal temperature of about 38°C (Jones and Morita, 1985). Their results suggest that nitrification bacteria will adapt to a variety of temperatures provided they are given time to do so. Haug and McCarty (1972) reported the following relationship between the ammonia removal rate, concentration and temperature for a submerged bed filter:

$$ACR = (0.11T - 0.2)\ (S/10)$$

Where:

ACR = ammonia consumption rate (mg/L/min)

S = ammonia concentration (mg/L)

T = water temperature (ºC).

2.2. Media Type and Size

Media is a solid material placed into a nitrification filter to provide surface area on which bacteria growth can occur. There are hundreds of media types that may be used for nitrification filters, ranging from sand and rocks to plastic media. Almost any solid material that is non-toxic to the nitrifiers and the crop can be used as media. The type of media selected usually is a function of desired media diameter and specific surface area, cost, availability and weight per unit volume. Carbonated base rocks, such as limestone and marble, provide buffering capacity to the system, at least until they become totally coated with bacteria. Plastic materials often have high void ratios, are light in weight, will last almost indefinitely if not exposed to sunlight, but are expensive per unit volume. Plastic media offer no buffering capacity.

2.3. Specific Surface Area

The specific surface area is the surface area of the media per unit volume. Because it costs more to build a large filter than a small filter, the specific surface area of media is an important design parameter. The media size, void ratio (volume of air left in the filter after it is filled with media divided by the total volume of the empty filter) and specific surface area are often related to each other. Small diameter media usually have a higher specific surface area and a lower void ratio than does a large diameter media of the same type. Specific surface area is usually a function of the media type selected. The higher the specific surface area of the media, the more bacteria that can grow in a unit volume of media, and the greater will be the total ammonia removal capacity per unit volume of filter.

2.4. Hydraulic and Mass Loading

Hydraulic loading in a submerged or trickling filter is a measure of the amount of water pumped onto the filter per unit cross-sectional area per unit time (expressed as m^3/m^2 –day). Maximum allowable flow rate (maximum irrigation rate) in all types of nitrification filters is usually set by the velocity that scours the bacteria off the media or produces excessive head loss. For fluidized filters, low flow is set by the minimum water flow that will supply the filter oxygen demand. Maximum velocity or flow rate in a fluidized bed is sometimes set by the need to keep the media in the filter container (Roberts, 1985). It is obvious that both the upper and the lower irrigation limits vary considerably with media size, manufacture, material and other variables.

Kaiser and Wheaton (1983) observed that for low ammonia concentrations shorter filter detention times (higher flow rates) produced higher ammonia mass removal rates. Kaiser and Wheaton (1983) argued that the nitrifiers growth rate and the nitrification rate were not a function of the concentration of the limiting substrate, but were a function of the mass loading of the limiting substrate. A similar finding was reported by Cook and Kincannon (1971) in their study of trickling filter configuration. They determined that COD removal efficiency depended on the total COD applied, and not its concentration or volumetric flow rate.

2.5. Film and Biofilm Thickness

The film refers to the stagnant layer of water surrounding the bacterial layer growing on the media surface. The film thickness is a function of water velocity over the media, water viscosity, and temperature. Hochheimer (1990) in his modeling work allowed film thickness to vary between 1.0 and 100 microns. Williamson and McCarty (1976) measured the film layer thickness and reported a thickness of 56 microns for aquaculture conditions.

A biofilm is an aggregate of bacteria and extracellular polymer that is attached to a solid surface. The biofilm thickness is usually subject of mechanical and hydrodynamic control in biofilters used in aquaculture, but its performance depends also by microbial composition and density. Atkinson and Fowler (1974) found that the total biofilm thickness could range between 0.07 and 4 mm. Under hydrodynamic control (i.e., flow rates), the biofilm thickness keeps usually under 0.2 mm.

D. FLUIDIZED BED FILTER

Fluidized bed filters have traditionally consisted of a bed of granular media maintained in a constant state of expansion or fluidization. They fall under the broad heading of mobile bed bioreactors (Lazarova and Manem, 1994). The media consists of particles with water negative buoyancy (e.g., sand, plastic beads, glass beads, crushed shells). Granular filters can be desirable due to their high specific surface area and ability to capture solids while performing nitrification (Chen et al., 1993; Losordo and Timmons, 1994; Westerman et al., 1996). Fluidized bed reactors can achieve superior performance to fixed bed reactors because the biofilm is evenly distributed throughout the reactor, while the liquid regime has plug-flow characteristics (Rittmann, 1982). In fluidized bed, each particle becomes coated with biofilm that grows as organics and other nutrients are extracted from the passing water. Excess biofloc, which develops during periods of high loading, tends to be abraded off by the continual collision between media particles (Jeris et al., 1974; Fan et al., 1987). Fluidized beds present an ideal fixed film bioreactor, benefiting from inherently high specific surface area and a natural resistance

to biofouling. Additionally, thin biofilms and high velocity regimes that surround each grain favor high rates for mass transfer for critical nutrients such as ammonia and oxygen. Because the bed is submerged, all oxygen to the nitrifiers must be supplied as dissolved oxygen in the culture water (Wheaton et al., 1994).

Bead filters are typically cylindrical in shape, with fittings on one end to admit water and on the other to release water. In fluidized bed filters, water is injected upwards through the bed at increasing flux rates (or superficial velocity) until the interstitial velocities generate hydraulic drag forces sufficient enough to overcome the submerged weight of an individual media particle. The media bed then expands, reducing the interstitial velocities as the void ratio of the bed increases until a balance point is reached between the uprising water velocity and the sinking rate of the media particles. In practice, however, the expanding bed height of media particles remains constant, but individual particles slowly bounce and roll about as the turbulence induced by the water injection system is dissipated. The expansion characteristics change as the biofilm develops, increasing the drag forces without contributing significantly to the particle weight. As a result, an acclimated fluidized bed often exhibits expanded bed heights different then predicted for clean media (Atkinson et al., 1981). Atkinson and Dave (1972) found that for particles exposed to constant attrition in a fluidized bed, the steady state microbial layer thickness depends on substrate concentration, i.e., growth rate. Conversely, Atkinson and Knights (1975) found that microbial layer thickness depends upon bed expansion (attrition) when exposed to a fixed substrate concentration (constant growth rate). Therefore, a steady biomass hold-up of a particle represents a balance between the overall growth rate and the degree of attrition, e.g., particle/ particle/ wall contacts, allowing a better control of biofilm thickness. Water velocity varies with reactor diameter at a given flow rate and directly influences the bed expansion level. The maximum bed expansion level in biofilter limits the flow rate through the biofilter. A high flow rate through the bead filter causes a high-energy demand. An ideal bed filter exerts little backpressure on pumps, thereby minimizing energy requirements. A reduction of bed expansion limitation and of energy consumption is achieved by using biofilter diameters that allow a lower water velocity and a high flow rate.

In optimizing fluidized bed design, all of above variables are usually considered. On this project, negative buoyant plastic beads were used as media.

E. MODELING OF FLUIDIZED BED FILTERS

Use of a biofilm model is necessary to ascertain whether the observed results correlate with the fundamental phenomena and assumptions that form the basis of the biofilm model. The model can then be used to cautiously to extrapolate beyond the range of values considered in the study. The model offers a pseudo-analytical solution for steady state biofilm kinetics through which substrate flux into a biofilm can be obtained from simple algebraic expressions that require only kinetic and physical parameters and a bulk substrate concentration (Saez and Rittmann, 1987). Model considerations and assumptions include: a) biological reactions based on Monod kinetics, b) application of Fick's law for diffusion of substrate across the bulk liquid/biofilm boundary and within the biofilm, c) a total biofilm mass balance including growth, decay and shear loss, d) a homogenous biofilm matrix with uniform local cell density and thickness, e) one limiting substrate that changes only in the direction perpendicular to the biofilm surface, f) all required nutrients (other than the one specified to be limiting) are in excess concentration, and g) a steady-state biofilm, which is defined as a biofilm that is not growing or decaying over time (Rittmann and McCarty, 1980).

A biofilm model is useful in biofilter design because it can predict the substrate flux (J), and the thickness of the biofilm (L_f) starting from the substrate concentration (S) and fundamental biofilm parameters. Rittmann and McCarty (1980) developed a model in which they based their predictions on the kinetics and energetics of substrate utilization and biofilm growth. Although the biofilm thickness can be nonsteady-state, a steady-state concentration profile within the biofilm can be assumed because the substrate concentration changes much more quickly than the biofilm can grow or decay. Therefore, the concentration profile is considered steady state even though the biofilm thickness can be changing. The steady-state biofilm was defined as one that is not growing or decaying over time. In their model, Rittmann and McCarty determined simultaneously the rate of substrate utilization and the biofilm mass, from the bulk liquid concentration and the

appropriate fundamental parameters. For modeling purpose, a biofilm is idealized as a homogenous matrix of bacteria and the extracellular polymers that bind the bacteria together and to the solid surface (Characklis, 1973). An idealized biofilm concept for biofilm model is presented in Figure 1.2.

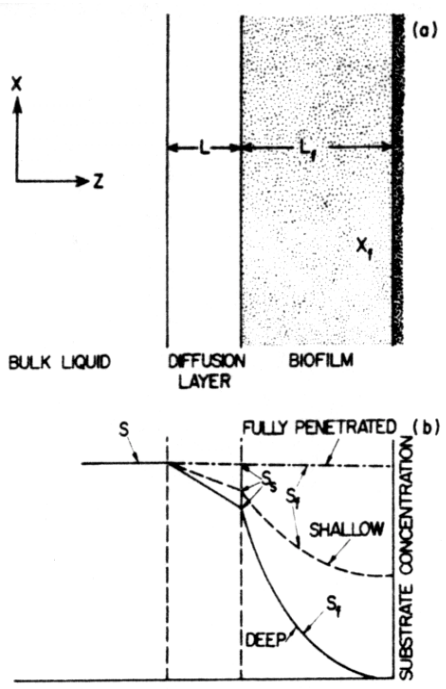

Figure 1.2. Conceptual basis for biofilm model. (a) Physical concepts; (b) Substrate concentration profiles (Rittmann and McCarty, 1980).

As shown in Fig. 1.2(a), the idealized biofilm has a uniform cell density of X_f (M_xL^{-3}) and a locally uniform thickness of L_f. Substrate concentration within the biofilm changes only in the *z* direction, which is normal to the surface of the biofilm.

It is assumed that all required nutrients are in excess concentration, except one, which is herein called the rate limiting substrate, or simply the substrate. The substrate utilization reaction within a control volume of biofilm is assumed to follow a Monod relation accumulation due to reaction

$$(M_xL^{-3}T^{-1}) = (kX_fS_f)/(K_s + S_f) \qquad (5)$$

where S_f is the rate limiting substrate concentration at that point in the biofilm (M_sL^{-3}), Ks is the half velocity coefficient (M_sL^{-3}), k is the maximum specific rate of substrate utilization ($M_sM_x\,T^{-1}$), and *t* is time. Rittman and McCarty (1978) have presented solutions for the flux (J) of the substrate into the biofilm for the general Monod reaction.

Development of a general solution for substrate flux into a steady-state biofilm requires three steps. The first two define substrate flux for extreme cases, for which explicit solutions can be derived. The third step provides a general solution that is applicable for situations between two extremes. Fig. 1.2(b) illustrates the three characteristics substrate-concentration profiles. Within the deep biofilm, the substrate concentration S_f decreases asymptotically to zero. The deep biofilm has the maximum possible flux for a given S_s, the substrate concentration at the liquid / biofilm interface. At the other extreme is fully penetrated biofilm, in which S_s penetrates to the full thickness of the biofilm. All cases that are not deep ($S_s \geq S_f > 0$) at the solid surface are called *shallow* biofilms. For all cases, the substrate-concentration gradient becomes zero at or before the solid-surface boundary.

In order to apply bacterial growth kinetics to a steady-state biofilm, a fundamental assumption related to the conservation of energy, must be made. Therefore, in order to determine the steady-state-biofilm mass, it is assumed that the total amount of biofilm mass is just equal to that which can be supported by the substrate flux. The steady-state-biofilm thickness can then be computed by equating the available and maintenance energy rates, where the steady-state thickness of biofilm is related to the flux:

$$L_f = JY / bX_f \qquad (6)$$

JY is the net energy that becomes available to the biofilm per unit time and per unit cross sectional area. Equation (6) requires that the steady-state biofilm be a dynamic one. The decaying portion continually gives up space to the growing portion in such a manner that the net biofilm thickness remains constant.

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CHAPTER II

FACTORS INFLUENCING THE NITRIFICATION EFFICIENCY OF FLUIDIZED BED WITH PLASTIC BEAD MEDIUM

A. INTRODUCTION

Due to concerns regarding the potential detrimental impacts of aquaculture production on the environment, increased regulations on aquaculture effluents and the need to conserve water resources and energy, the aquaculture industry is focusing on development and refinement of water recycling technologies (Klontz, 1979; Rosenthal, 1994). A recirculating aquaculture system (RAS) is an assemblage of parts and devices used for the culture of aquatic organisms where water is continuously cleaned and reused (Libey, 1996). Mechanical filtration removes particulate wastes, while biological filtration removed dissolved wastes via biochemical reactions that occur during bacterial metabolism. Ammonia is released into the water column as a final product of animal metabolism. Ammonia removal constitutes the main concern for biofiltration in aquaculture, in order to ensure prolonged fish survival. Biofiltration employs strains of bacteria (*Nitrosomonas sp.*), which oxidize ammonia to the nitrite (NO_2 –N), and Nitrobacter *sp*., which oxidize the nitrite to relatively nontoxic nitrate (NO_3^- -N), during a process known as nitrification. A commonly used technique in RAS for nitrification is the biological fixed film process (Brune and Gunther, 1981). Biofilters, defined as biological entities composed of a solid medium on which nitrifying bacteria grow, are commonly used in production RAS to remove ammonia. They are considered major components in intensive RAS (Libey and Miller, 1985) and are specifically designed for concentrated bacteria attachment and nitrification via fixed-film kinetics.

Many different biofilter physical configurations, i.e., downflow filters, rotating biological contactors (RBC), fluidized bed filters or fluidized bed biological reactor (FBBR), etc., are used in RAS (Wheaton et al., 1990). Fluidized beds consist of a bed of granular media maintained in a constant state of expansion of fluidization, in which the biofilm grows attached to small carrier particles that remain suspended in the fluid; i.e., fluidized by the drag forces associated with the upward flow of water. The medium (e.g.,

sand, plastic beads, glass beads, etc.) has specific gravity heavier than water. Granular filters are generally desired for the following advantages: allow for better control of biofilm thickness, have superior mass transfer characteristics, are not subject to clogging, and provide very high surface area for biofilm development while maintaining low pressure drops (Grady Jr. and Lim, 1980). Negative buoyant (sinking) plastic granular media have been used recently in fluidized beds, but still have limited applications due to limited knowledge about its performance. Improving the understanding of one type of this relatively new fluidized bed media, and evaluating its potential under conditions of constant and different ammonia supply, was the first objective of this study. The second and the third objectives were to relate filter performance to filter design and operational characteristics, and to compare the results to performance data generated using a current biofilter model.

B. EXPERIMENTAL METHODS

1. Test System Design

Preliminary experiments were conducted with a laboratory scale biofilter (15.2 cm diameter and 100 cm height), filled with the plastic beads under consideration. Different influent flow rates and water velocities were then tested and constituted the basis for the design of larger columns used in the experiment.

Three water-recirculating systems, each including three-biofilters of different diameters were built and operated at the Virginia Tech Aquaculture Center Lab. The configurations of the three systems were identical. A schematic diagram of a system is shown in Figure 2.1.

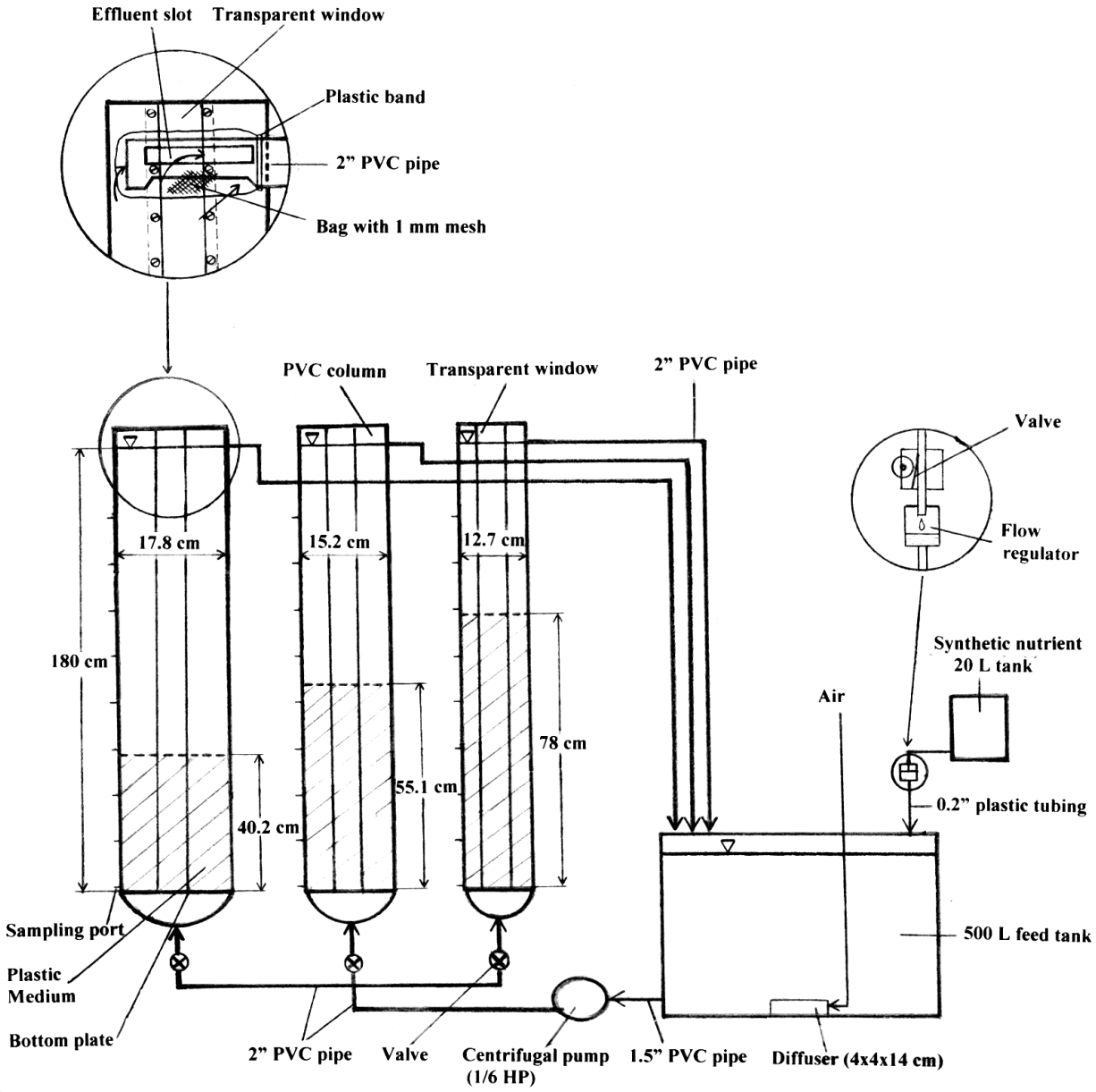

Figure 2.1. Schematic diagram of one biofiltration system. Height of filtration media is for the static (unfluidized) bed.

Each system consisted of a 500 L oval tank (0.6m x 0.7m x 1.2m) made from polyvinyl chloride (PVC) (Rubbermaid®), three cylindrical PVC biofiltration reactors, one 1/6 HP pump, an aeration device, and a dripping system for addition of a nutrient mixture. Wooden frames supported the biofilters in a vertical position. The feed tank was the site for aeration, nutrients addition and daily water exchange. Flow through each filter was individually controlled to ensure that each filter received the same hydraulic loading. Flow rates used ranged from 6 Lpm / filter to 12 Lpm / filter, which yielded between two to four system turnovers per hour. This corresponded to a volumetric hydraulic loading rate of between 600 Lpm / m^3 / filter and 1200 Lpm / m^3 / filter. The aeration device consisted of four stone diffusers (14 cm x 4cm x 4 cm) that sat on the tank bottom and were connected to a supply airline. The dripping system, a Kent – Marine AquaDoze$_{TM}$ product with a reservoir of 20 L capacity, provided constant nutrient addition over a 24-hour period.

The three systems were maintained in low lighting conditions to provide optimal light conditions for bacterial growth in the biofilters. Throughout the study, an exhaust fan and four propane gas heaters were used to regulate ambient air temperature. Submersible emergency heaters, Visi-Therm VTN300, were also available one for each system, to maintain the temperature over 25ºC.

2. Biofilter Characterization

The filters used in this study were up-flow fluidized bed biological reactors (FBBR). Each filter consisted of a cylindrical vessel with a transparent window about 8 cm wide along the cylinder body, which allowed for light penetration and visual media inspection. The depth of water in each column was 180 cm. The diameters of biofilters tested were D_1 = 12.7 cm, D_2 = 15.2 cm, and D_3 = 17.8 cm. The water volumes of each biofilters were 23.04 L, 32.64 L and 45.26 L, respectively. Nine collecting ports were installed vertically, every 20 cm along the cylinders. A perforated PVC distribution plate located at the bottom assured distribution of inflow water and a screen at the top prevented the loss of the beads. Gate valves were mounted on the water supply pipes (2” diameter) at the bottom inflow connections for adjustment and control of water flow rates. The filter medium used consisted of 2 x 4 mm ABS (acrylonitrile, butadiene and styrene) plastic

beads with a specific gravity of 1.06 (International Polymer Corp., Allentown, Pennsylvania). The average specific surface area of medium was 1600 m^2 / m^3. Each biofilter held 10 L of beads, which provided a biofiltration surface area of 16 m^2. At no flow, beads occupied 44 %, 31 % and 22 % of the reactor volume, corresponding to a bed height of approximately 80 cm, 55 cm and 40 cm. The submerged inner walls of the tank and the pipes supplemented the surface area contributing to treatment.

3. Acclimation of Biofilters

The biofilters were acclimated by introducing the synthetic mixture described in Table 2.1.

Table 2.1. Composition of test substrate (Zhu and Chen, 1999).

Ingredients	Mass* (g)
NH_4Cl	1377
$NaHCO_3$	3500
$MgSO_4 \times 7H_2O$	36
Na_2HPO_4	159
KH_2PO_4	153
$FeCl_3 \times 6H_2O$	5

Mass*(g) - 100g of the synthetic mixture contains 8.64 g NH_3.

Commercial nitrifying bacterial seed provided by BACTA-PUR^{TM} was used to inoculate the systems. Concentrations of total ammonia nitrogen (TAN) and nitrite-nitrogen (NO_2^- –N) were monitored daily to assess nitrifier establishment and activity. Daily ammonia mass loading was corrected, based in feed tank concentration, in order to maintain a level of 2 mg /L NH_3-N. Biofilters were considered fully acclimated when a steady-state culture was established, capable of keeping TAN and NO_2^- –N levels below 0.7 mg / L in each system, under conditions of 8.64 g/day mass ammonia loading for 7 consecutive days. Following acclimation, studies on biofilter performances were initiated.

4. Performance and Water Quality Monitoring

4.1. Daily Operation

Initially, all systems were filled with municipal water, and a combination of municipal water and well water was used later for water replacements (3 : 1 ratio). New water (10 % replacement) was introduced into the systems each day, following water sampling. Sodium bicarbonate ($NaHCO_3$) additions were made as needed to maintain pH and alkalinity at levels characteristic of culture systems (Table 2.2). The targeted ranges for other basic water quality parameters are also given in Table 2.2.

Table 2.2. General water quality parameters for recirculating fish culture systems.

Parameter	Target range	Reference
NH_3 –N, mg / L	< 1.0	Malone 1999
NO_2^- –N, mg / L	<1.0	Malone 1999
NO_3^- -N, mg / L	< 1000	Losordo 1991
Disolved oxygen, mg / L	> 5	Kaiser and Wheaton 1983; Losordo 1991
pH	6.7 - 9	Klontz et al., 1979
Temperature, °C	20 - 30	Malone 1999
Alkalinity, mg / L	> 80	Klontz et al., 1979 Meade 1989
Hardness, mg / L	> 100	Meade 1989

Equal amounts of the synthetic solution were added to each system constantly (approximately 5 L/day), 24 hours per day. The feed tanks were aerated continuously, which served to mix the nutrients, provide oxygen and strip CO_2. Table 2.3.a and 2.3.b

present the operational parameters used in the experimental design for Part 1 and for Part 2 studies, respectively.

Table 2.3. Experimental design of Parts 1 and 2 of study. A) Operational parameters of Part 1, with flow rate variation; B) Operational parameters of Part 2, with NH_3-N loading variation. R_1, R_2 and R_3 are the reactors types used in the study.

A)

Stage period, days	Flow rate/ filter, Lpm	Water velocity, cm/sec R_1*	R_2**	R_3***	Daily NH_3-N added, g	Nutrients added/day, g	Influent NH_3-N conc., mg/L	Recirculation freq., pass/hr
7	6	0.80	0.60	0.40	8.64	100	0.60	2.00
7	8	1.06	0.80	0.53	8.64	100	0.57	2.66
7	10	1.33	1.00	0.67	8.64	100	0.53	3.33
7	12	1.60	1.20	0.80	8.64	100	0.50	4.00
B)								
7	12	1.60	1.20	0.80	8.64	100	0.50	4.00
7	12	1.60	1.20	0.80	10.80	125	0.52	4.00
7	12	1.60	1.20	0.80	12.96	150	0.69	4.00
7	12	1.60	1.20	0.80	15.12	175	0.83	4.00
7	12	1.60	1.20	0.80	17.28	200	0.99	4.00

R_1* Reactor with diameter = 12.7 cm

R_2** Reactor with diameter = 15.2 cm

R_3*** Reactor with diameter = 17.8 cm

Headloss measurements were performed at the end of each stage. A hose was connected at the bottom plate level of each biofilter and kept vertically, recording the water level difference at the top of the biofilter.

4.2. Water Quality Monitoring

The study period lasts 135 days. Daily water samples were collected at 12:00 a.m., prior to the daily water exchange, to monitor the loading of nitrogenous compounds. Samples were taken prior to the biofilter, from the feed tank, and from immediately above the fluidized bed through collection ports. These samples were analyzed for TAN and NO_2^- –N. TAN analyses were performed in duplicate. Nitrate-nitrogen (NO_3^- -N), alkalinity (as $CaCO_3$), temperature (°C), dissolved oxygen (DO) and pH were measured daily by sampling the feed tank water. Water hardness (as $CaCO_3$) in the systems and DO

of filter outflow were tested periodically. All tests followed protocols presented in Standard Methods (APHA et al., 1995). A YSI, model 58, dissolved oxygen meter (YSI Co., Yellow Spring, Ohio) was used for temperature and DO measurements, and a Hanna Instruments, model HI 1270, pH probe (Hanna Instruments, Woonsoccket, Rhode Island) was used to monitor pH. Concentrations of TAN, NO_2^--N and NO_3^--N were measured using a Hach DR/2000 spectrophotometer (Hach Co., Loveland, Colorado). Total alkalinity and hardness both were analyzed via Hach titration. Calculations of NH_3–N were made using equations presented by Emmerson et al. (1975).

Media samples were collected during the experiments by inserting a core sampler down from the top of the columns. Samples of the medium were collected at different bed heights. Samples were collected from each biofilter on the last day of each experimental stage. The samples were transported to the Environmental Biotechnology Laboratory and processed immediately for biofilm thickness measurements, by evaporation methods (modified from Rittmann and McCarty, 1980). Samples were weighted using an analytical scale before and after a 24-hour period at 105 °C.

5. Statistical Analysis

All statistical tests were performed using The SAS System® software package. The TAN and NO_2^--N removal statistical design for flow rate variation and concentration variation was a repeated measures split plot. Splits plot design without the repeated factor (called also a within-subject factor) was applied for biofilm thickness results analysis. The repeated factor was considered the day, the replication factor was the system, and treatment factor was the filter. Since the concentrations and flow rates were applied to the same nine filters, the filter (experimental unit) was divided into concentration or flow rate stages (split plots) in the design. Since each concentration or flow rate stage was measured repeatedly on each of at least seven days, the day factor was considered a repeated (or within-subjects) factor. The correlation structure for the day repeated measures factor was modeled with an autoregerssive structure.

For flow rate and TAN concentration:

(3 systems) x (3 filters) x (4 flow rates) x (7 days) = 252 observations

(3 systems) x (3 filters) x (5 TAN concentrations) x (7 days) = 315 observations

For biofilm thickness:

(3 systems) x (3 filters) x (4 flow rates) = 36 observations

(3 systems) x (3 filters) x (5 TAN concentrations) = 45 observations

Due to the time lag between the stages (2+ days), the modeling of correlation structure for the stages was not a concern.

C. RESULTS AND DISCUSSION

1. Biofilters Acclimation

Each group of three biofilters connected to the same water bath was exposed to similar conditions of acclimation. The acclimation process was started in all three systems at the same time. The water flow rate was maintained at 6 Lpm for each biofilter throughout the entire period of acclimation. TAN and NO_2^--N levels increased to a peak prior to decreasing to steady state conditions (Figure 2.2 and 2.3).

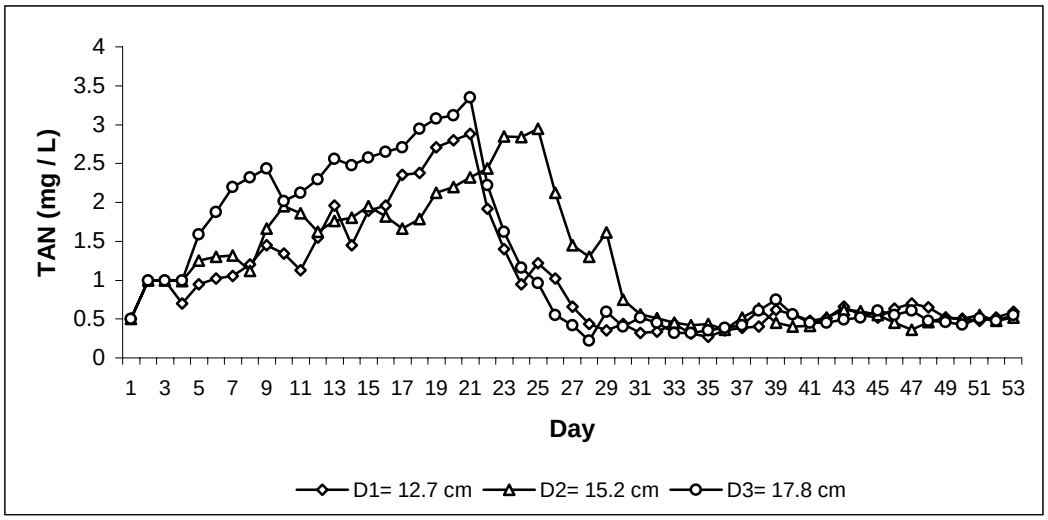

Figure 2.2. Biofilter microbial acclimation shown using total ammonia nitrogen (TAN) to indicate first stage nitrifier population establishment.

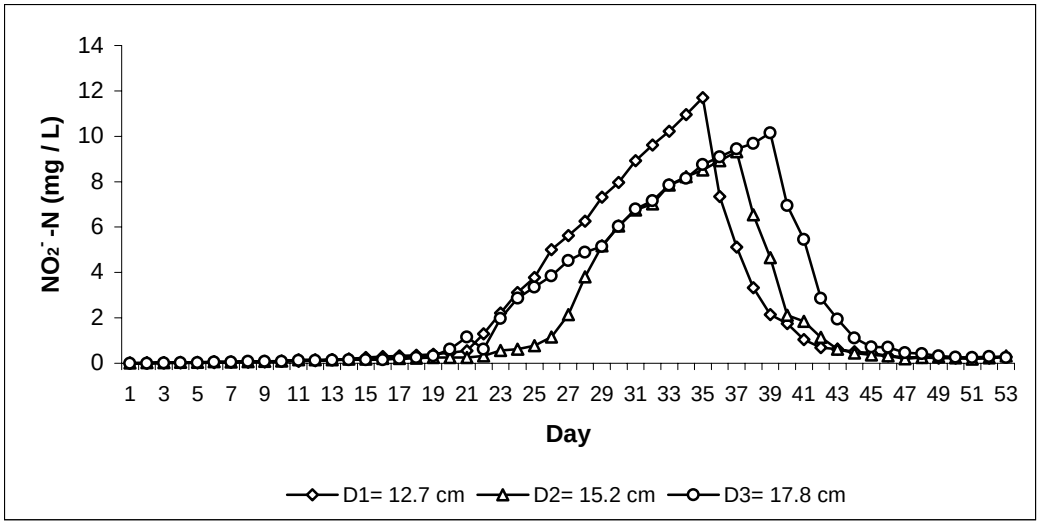

Figure 2.3. Biofilter microbial acclimation shown using nitrite nitrogen (NO_2^--N) as an indicator of second-stage nitrifier population establishment.

The TAN and NO_2^--N accumulation was building up before peak, despite a 10% volume of daily water exchange. The amount of synthetic mixture added daily was increased in steps, targeting a less than 2 mg / L TAN concentration in any system at peak, according to Malone and Burden, (1998). However, at peak concentration this limit was passed in all systems. The maximum TAN concentration per system was 2.88 mg / L in the first, 2.95 mg / L in the second, and 3.35 mg / L in the third, respectively. TAN concentrations for all systems peaked between days 21 and 25. Similar dynamics occurred in NO_2^--N concentrations, where the peaks were observed between days 35 and 39. The rate of decline to steady state conditions was similar among all the filters. Time to TAN and NO_2^--N stabilization exceeded the typical 20 to 35 day stabilization period for a new biofilter reported by Wheaton et al. (1991). More time was necessary in this study for stabilization due to the continuous increases in TAN loading made after the peak, in order to acclimate the systems to a flux of ammonia of 180 mg / m^2 –day. This TAN flux level was the value targeted for the first part of the study. All systems reached TAN steady state conditions around day 33, whereas NO_2^--N stabilization occurring around day 45. Between days 46 and 53, all systems were able to treat this flux of ammonia, keeping both TAN and NO_2 –N concentrations under 0.7 mg / L (Fig. 2.2 and Fig. 2.3).

2. Filter Performances and Basic Water Quality Analysis

2.1. TAN and NO_2^--N Mass Removal Analysis

Part 1. Nitrification rates during each trial were evaluated by the percent TAN and NO_2^--N removal efficiency. The percent of TAN removed per pass by each biofilter decreased significantly ($p<0.0001$) as flow rate increased in each consecutive stage (Figure 2.4). The greatest difference was measured in the biofilter with diameter D_3, where the range of mean TAN removed was 62.76% at 6 Lpm, and 18.33% at 12 Lpm. Biofilter D_1 was the second, removing between 37.14 and 15.94% TAN at the two flow rates, respectively, whereas biofilter D_2 showed the lowest performance change with 28.96 and 13.68%. The complete data set, with standard deviations, is presented in Table 2.4. As the removal efficiency decreased when flow rate increased, the TAN concentration in tanks decreased significantly in each stage ($p<0.0001$), suggesting that higher recirculating rates increased the average TAN level in the biofilter but, consequently, lowered TAN concentration in the tank (Table 2.5, and Figure 2.5). The mean ammonia concentration in the feed tank was found to decrease continously (17% between the first and the fourth stage) as flow rate was increased from 6 to 12 Lpm. These findings are in agreement with those reported previously (Rogers and Klemeston, 1985; Malone et al., 1999). Increasing the mean TAN concentration in the biofilter by increasing the recirculating rate resulted in proportional improvements in a filter's conversion capabilities.

The difference in percent TAN removal was highly significant among the three-biofilter types at flow rates of 6 Lpm, 8 Lpm, and 10 Lpm ($p<0.0001$), but the difference became less significant ($p=0.0023$) at 12 Lpm in Stage 4 (Table 2.5, and Figure 2.5). Malone et al. (1993) reported that high removal efficiencies imply large decreases in TAN concentrations within the biofilter, exposing large sections of the biofilter to unnecessary low TAN levels, reducing the nitrification efficiency of the filter as a whole. Cooper and Weldon (1981) reported from sand fluidized bed experiments that nitrification occurs in the first segment of the biofilter, whereas the upper segment is where denitrification predominantly occurs. These reports suggest an explanation for the differences recorded between biofilters in TAN removal efficiency at the same flow rates

applied. D_3 had the largest surface area exposed to the TAN inflow and the lowest bead bed thickness, allowing for a larger portion of the biofilter media to be exposed to a higher TAN concentration. Therefore, more intense nitrification might have occurred in biofilter D_3 than in biofilters D_1 and D_2, which were narrower and had thicker beds.

In biofilter D_3 the TAN removal was highest at 62.76% for a flow rate of 6 Lpm, but decreased rapidly to 39.74%, when the flow rate was increased to 8 Lpm. At 6 Lpm (0.4 cm/s water velocity), the bed volume increased by 19% over the no flow volume (Figure 2.14), but only slight vibrations of the beads were observed. This lack of active movement would reduce attrition and the microbial layer grew thicker than in biofilters D_1 and D_2, where an active fluidization occurred at 6 Lpm. This is in agreement with Atkinson and Knights (1975) who found that microbial layer thickness depended upon bed expansion and attrition, when the biofilm was exposed to a constant concentration of substrate. Biofilm thickness measurements revealed that the biofilm was always thickest in biofilter D_3, (Figure 2.16).

Table 2.4. Mean percent TAN removal for each stage and biofilter in Part 1 of the study. P-values show the difference between the filters for each flow rate applied.

Flow Rate	Biofilter D_1* (%)	SD	D_2** (%)	SD	D_3*** (%)	SD	p-value (Diameter)
Stage 1 (6 Lpm)	37.14	± 2.56	28.96	± 1.42	62.76	± 3.04	p<0.0001
Stage 2 (8 Lpm)	29.11	± 1.30	27.09	± 1.34	39.74	± 4.21	p<0.0001
Stage 3 (10 Lpm)	21.15	± 1.27	19.03	± 1.48	27.04	± 2.82	p<0.0001
Stage 4 (12 Lpm)	15.94	± 2.65	13.68	± 2.54	18.33	± 2.46	p=0.0023

D_1* = 12.7 cm diameter
D_2** = 15.2 cm diameter
D_3*** = 17.8 cm diameter

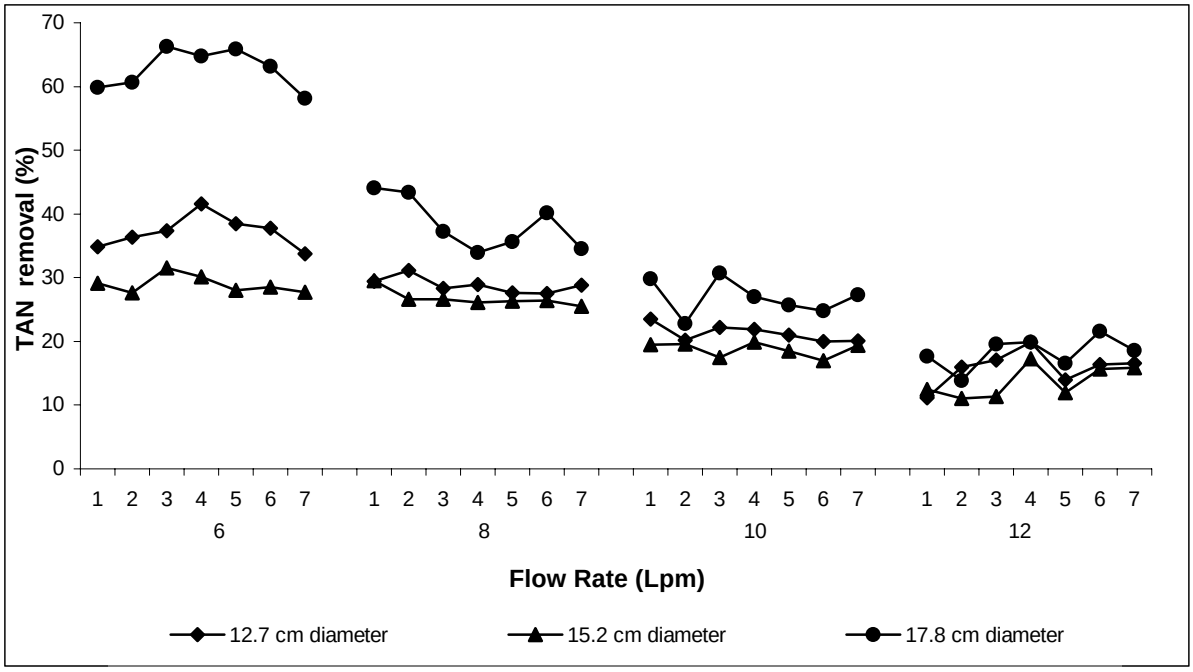

Figure 2.4. Percent TAN removal versus time and flow rate in the various biofilters in Part 1 of the study.

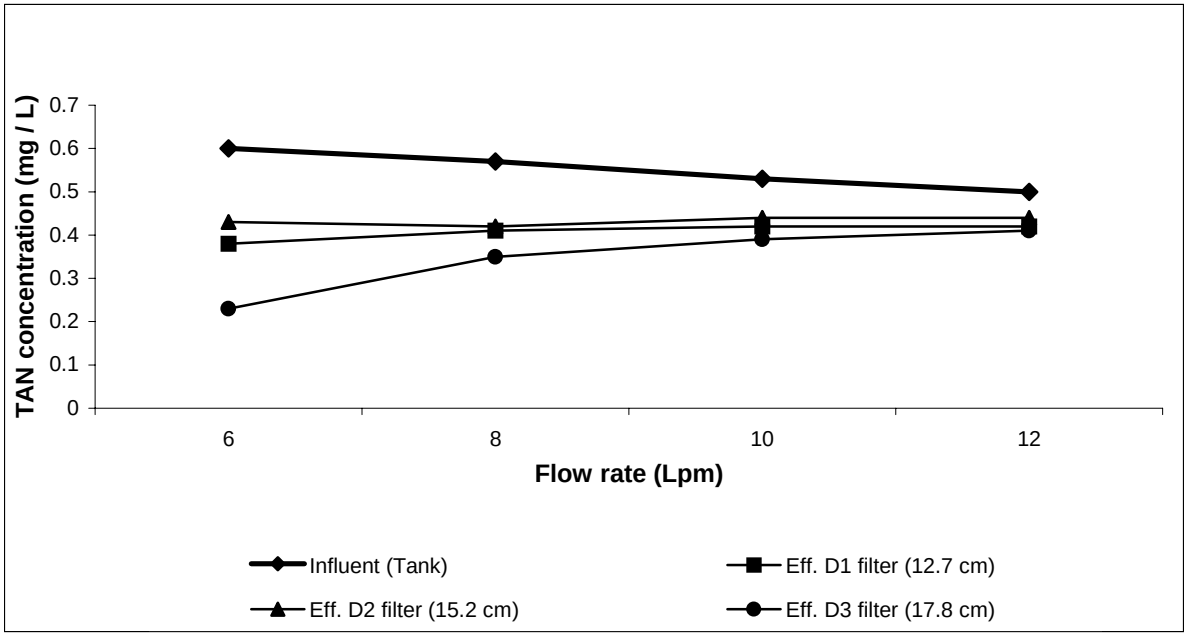

Figure 2.5. Mean influent and effluent TAN concentrations for each biofilter versus various flow rates applied in Part 1 of the study.

Mean NO_2^- –N removal efficiency (oxidation to NO_3^- -N) decreased slightly in the first part of the experiment as flow rate was increased, in all biofilters. The NO_2^- -N removal was based on NO_2^- -N concentrations into and out of the filter. The negative values (Figure 2.6 and Figure 2.10) represent addition of NO_2^- -N into the filter by NH_3 -N oxidation (Westerman et al., 1996). This suggested that *Nitrobacter* populations may have difficulty adjusting to increasing NO_2^- -N levels, resulting in a lag between loading and microbial community response. This phenomenon has been regarded as one of the problems associated with biofiltration in recirculating aquaculture systems (Luchetti and Gray, 1988; Hall, 1999). No significant difference in NO_2^- –N removal efficiency was observed between biofilters D_1 and D_2 ($p=0.0116$). Biofilter D_3 removed NO_2^- –N more efficiently than the other two at 6 Lpm ($p<0.0001$), but became less consistent at higher flow rates. The greatest efficiency of NO_2^- –N removal was observed at 6 Lpm on day one, with a maximum value of 49.84% in biofilter D_3. The minimum, -30.42%, was observed in the same biofilter at 12 Lpm on day seven.

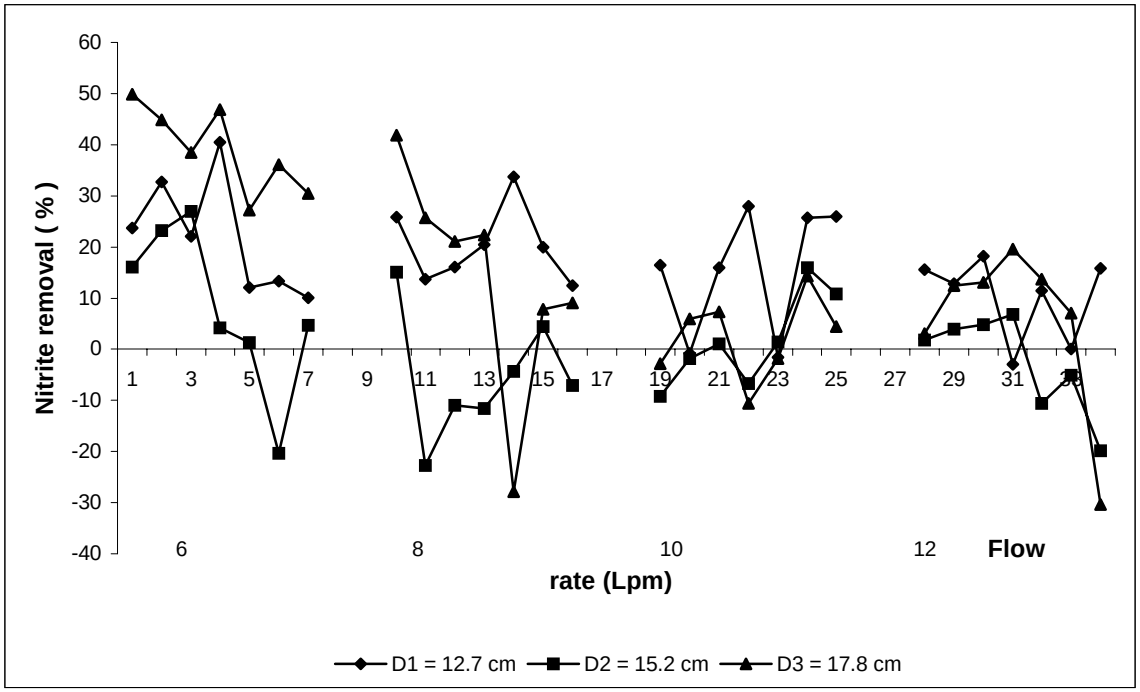

Figure 2.6. Percent NO_2^- –N removal versus time and flow rate applied in Part 1 of the study.

Mean influent (tank) and effluent NO_2^- –N concentrations for each biofilter increased slightly from 6 to 10 Lpm, but remained constant at 12 Lpm, except in filter D_3, where

the concentration decreased (Figure 2.7). These results suggest that the biofilters were able to remove the increasing NO_2^- –N production from the systems as tank TAN concentration decreased, despite the daily fluctuations. The values recorded were well under the harmful limits (> 1.0 mg/L NO_2^- –N, Malone et al., 1999) for a fish population. Mean tank NO_2^- –N levels were not significantly different between various flow rates applied ($p > 0.1$), reaching values between 0.17 - 0.20 mg/L (Table 2.5). The mean effluent NO_2^- –N ranged between 0.11 mg/L for biofilter D_3 in Stage 1, and 0.20 mg/L for biofilter D_2, in Stage 3.

However, based on TAN removal, biofilter D_3 was found to be the most reliable nitrifying biofilter in the first part of this study. In terms of NO_2^- –N removal, all three biofilters were similar except biofilter D_3 in the first stage.

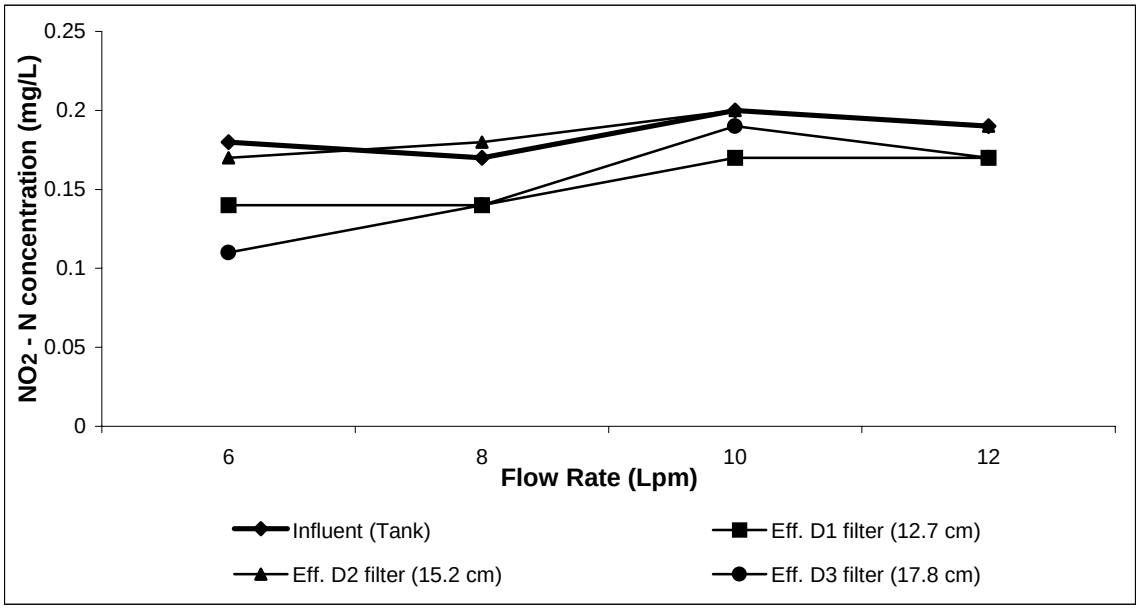

Figure 2.7. Mean influent and effluent NO_2^- -N concentrations for each biofilter versus various flow rates applied in Part 1 of the study.

Table 2.5. Average TAN and NO_2^-- N levels at various flow rates applied.

Part /	Influent, mg/L		Flow Rate,	Effluent D_1, mg/L		Effluent D_2, mg/L		Effluent D_3, mg/L	
Stage	TAN	NO_2^--N	Lpm	TAN	NO_2^--N	TAN	NO_2^--N	TAN	NO_2^--N
1/1	0.60	0.18	6	0.38	0.14	0.43	0.17	0.23	0.11
1/2	0.57	0.17	8	0.41	0.14	0.42	0.18	0.35	0.14
1/3	0.53	0.20	10	0.42	0.17	0.44	0.20	0.39	0.19
1/4	0.50	0.19	12	0.42	0.17	0.44	0.19	0.41	0.17

Part.2. At 180 mg TAN /m^2-day applied, the difference in percent removal TAN concentrations was significant among the biofilters ($p<0.0001$), but the difference decreased in the fifth stage, becoming not significant at 360 mg /m^2-day TAN ($p=0.5686$). At a TAN loading of 180 mg/m^2-day, biofilter D_3 showed the best performance, removing 18.33% TAN, followed by D_1 with15.94% and D_2 with 13.68%. At 360 mg TAN /m^2-day applied, D_3 removed 14.08%, D_1 14.01% and D_2 13.21%. The complete data for the experiment, including standard deviations, are presented in Table 2.6. For each filter the percent TAN removal increased as TAN supply increased, reaching a maximum value at 225 mg/m^2-day TAN for filter D_1 and at 270 mg/m^2 -day TAN for filters D_2 and D_3. Percent removal then decreased again at higher TAN loadings (Figure 2.8).

Table 2.6. Mean percent TAN removal for each stage and biofilter in Part 2 of the study. P-values show the difference between the filters for each TAN concentration applied.

TAN	Biofilter						p-value
applied	D_1* (%)	SD	D_2** (%)	SD	D_3 ***(%)	SD	(Diameter)
Stage 1 (180 mg/m^2-d)	15.94	± 2.65	13.68	± 2.54	18.33	± 2.46	p<0.0001
Stage 2 (225 mg/m_2-d)	20.02	± 2.04	17.07	± 2.55	19.96	± 2.35	p=0.0014
Stage 3 (270mg/m^2-d)	18.43	± 1.07	17.75	± 1.39	19.96	± 1.68	p=0.0689
Stage 4 (315 mg/m^2-d)	18.43	± 1.11	16.96	± 1.47	18.90	± 1.69	p=0.2107
Stage 5 (360 mg/m^2-d)	14.01	± 2.33	13.21	± 2.01	14.08	± 2.38	p=0.5686

D_1* = 12.7 cm diameter
D_2** = 15.2 cm diameter

D_3*** = 17.8 cm diameter

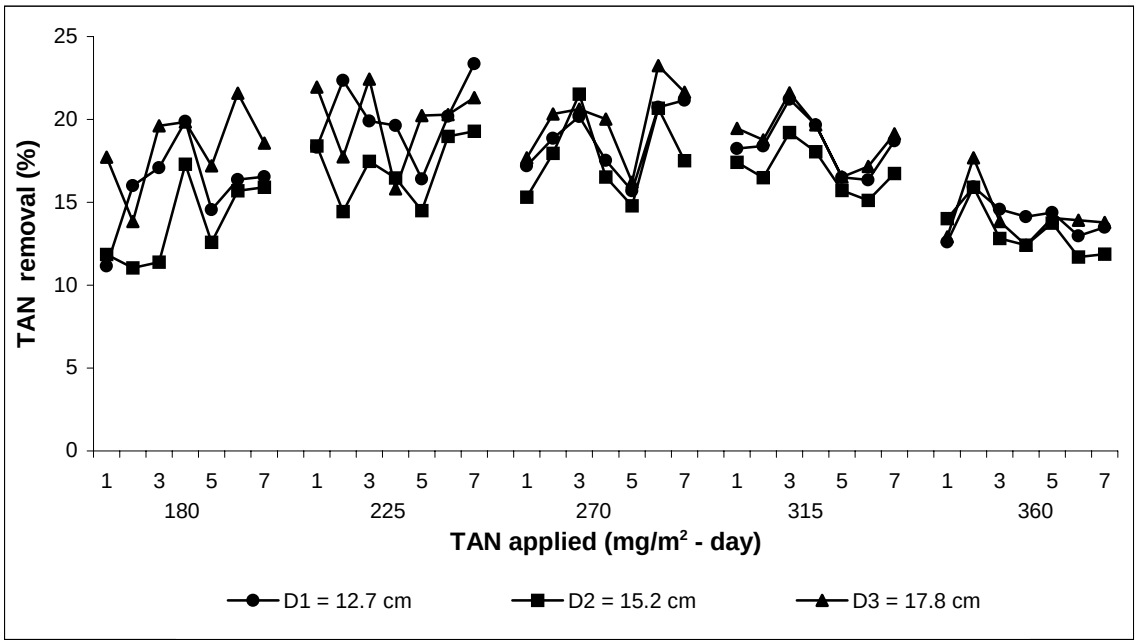

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Figure 2.8. Percent TAN removal versus time and TAN applied in Part 2 of the study.

The range in mean percent ammonia removal accomplished by the filters was between 13.21% for D_2 at 360 mg/m^2-day TAN loading and 20.02% for D_1 at 225 mg/m^2-day TAN loading (Table 2.6).

The dynamics of TAN removal efficiency in Figure 2.8 suggested that it was not dependent upon TAN loading. Instead, the results showed that increasing TAN loading from 180 to 360 mg/m^2-day affected the influent (tank) TAN concentration, increasing the level with 0.5 mg/L (Table 6 and Figure 2.9). These results are partially in agreement with those reported by Malone et al. (1999) for bead bed biofilters experiments. Malone et al. reported that increase in influent TAN concentration increased the mean TAN concentration into the biofilter, therefore improving the gradient of diffusion into the biofilm and enhancing conversion. The results from this experiment suggested that there is a limit to this phenomenon in plastic beads systems, due to lack of a correlation between nitrification rate and TAN concentration applied, resulting in TAN accumulation. Atkinson et al. (1981) reported from fluidized sand biofilters experiments, that for a given reactor configuration under conditions of constant attrition, the microbial (biofilm) growth rate depended upon the substrate concentration in the reactor. Those

findings are supported by the biofilm thickness measurements made in this study (Figure 2.16 and Figure 2.17).

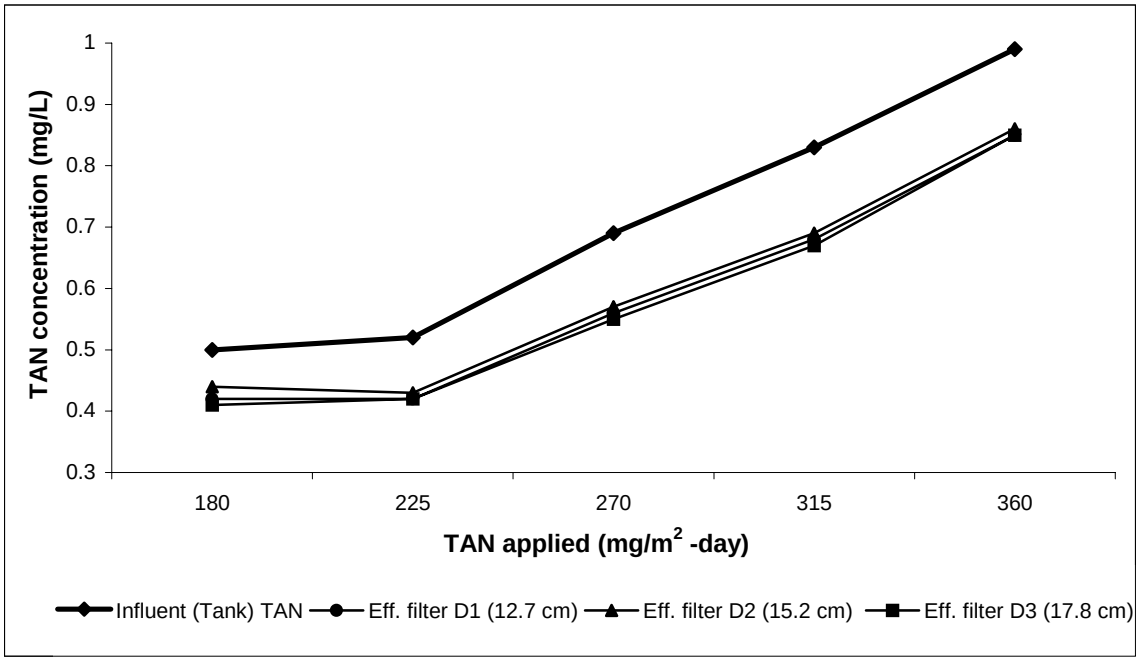

Figure 2.9. Mean influent and effluent TAN concentrations for each biofilter versus various TAN concentrations applied in Part 2 of the study.

The mean NO_2^- -N reduction increased in Part 2 of the experiment, as TAN concentration applied increased from 180 to 360 mg/m^2-day (Figure 2.10). The results showed a better adjustment of *Nitrobacter* populations to higher NO_2^- -N production; i.e., removal efficiencies were all positive at 270, 315 and 360 mg/m^2-day applied. No significant difference in NO_2^- -N removal efficiency was observed between biofilters ($p=0.0784$), except at 180 mg/m^2-day, where the NO_2^- -N reduction ranged between 13.68% in biofilter D_2 and 18.83% in biofilter D_3 ($p<0.0001$). The complete data set is presented in Table 2.7. The maximum efficiency of NO_2^- -N removal (49.84%) was observed in biofilter D_3 on day one at 180 mg/m^2-day TAN, and the minimum value (-30.42% addition of NO_2^- -N in the filter) was observed on day seven at 360 mg/m^2-day in the same biofilter.

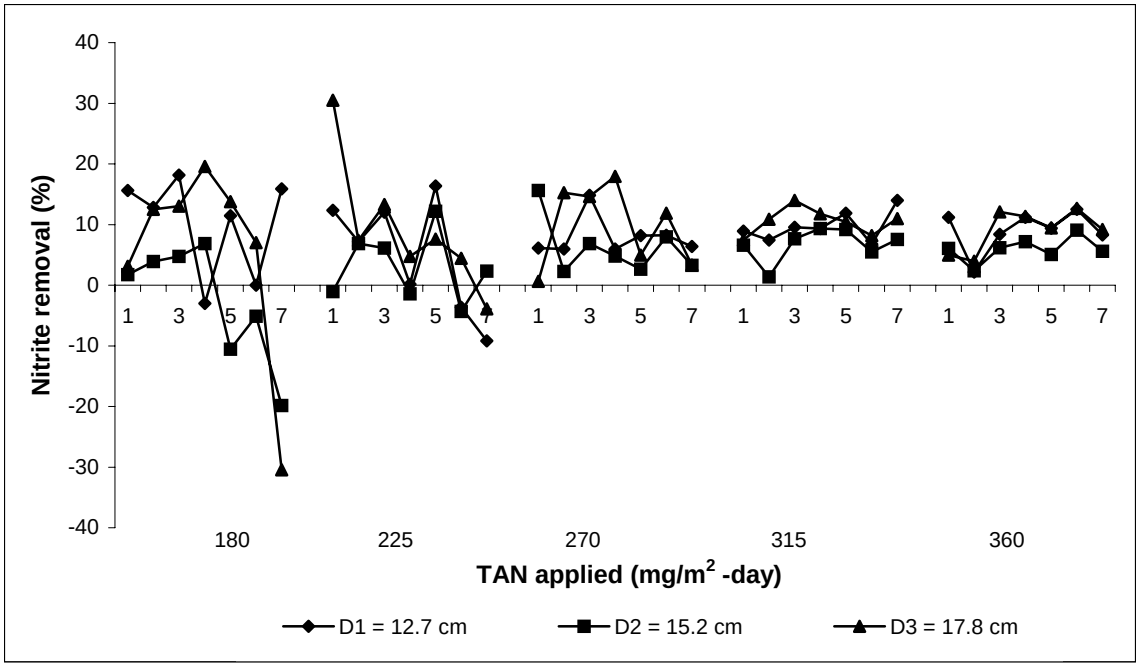

Figure 2.10. Percent NO_2^- -N removal versus time and TAN concentrations applied in Part 2 of the study.

Mean influent (tank) and effluent NO_2^- -N concentrations for each biofilter increased significantly at 180, 225 and 270 mg/m^2-day TAN loading, then decreased unexpectedly at 315 mg/m^2-day TAN and increased again at the highest loading (Figure 2.11). The mean NO_2^- -N level in tank was significantly different ($p<0.0001$) between various TAN concentrations applied, reaching values between 0.19 - 0.29 mg/L (Table 2.7). The mean effluent NO_2^- -N ranged between 0.17 mg/L for filter D_1 and D_3 at 180 mg/m^2-day TAN, and 0.28 mg/L for filter D_2 at 270 mg/m^2-day TAN.

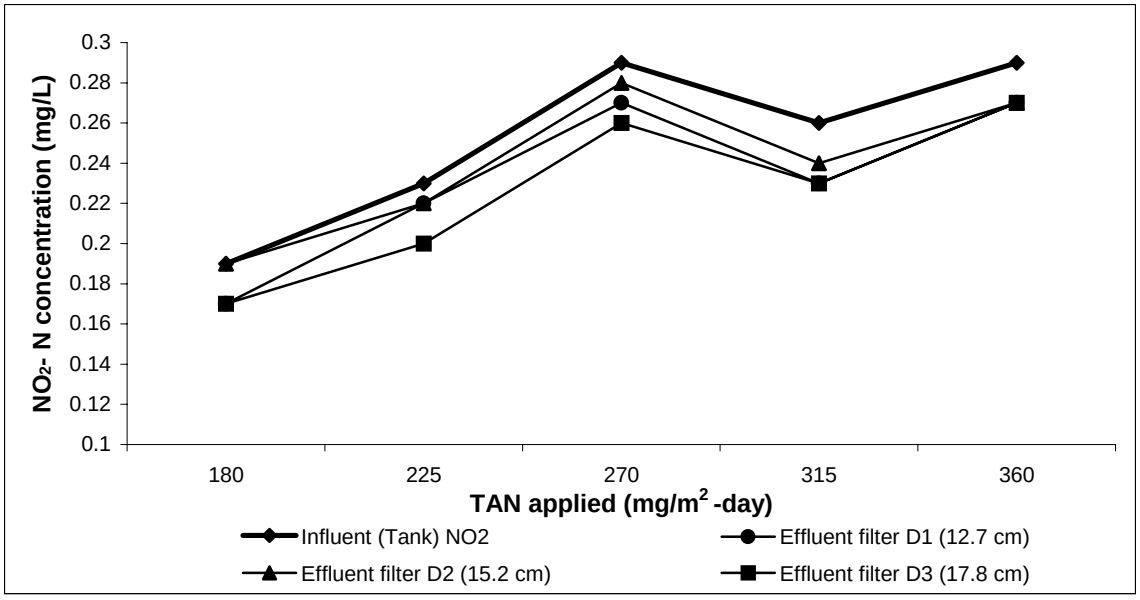

Figure 2.11. Mean influent and effluent NO_2^- -N concentrations for each biofilter versus various TAN concentrations applied in Part 2 of the study.

Table 2.7. Average TAN and NO_2^- -N levels at various TAN concentrations applied.

Part/ Stage	Influent, mg/L TAN	NO_2^--N	TAN applied, mg/m^2-day	Effluent D_1, mg/L TAN	NO_2^--N	Effluent D_2, mg/L TAN	NO_2^--N	Effluent D_3, mg/L TAN	NO_2^--N
2/1	0.50	0.19	180	0.42	0.17	0.44	0.19	0.41	0.17
2/2	0.52	0.23	225	0.42	0.22	0.43	0.22	0.42	0.20
2/3	0.69	0.29	270	0.56	0.27	0.57	0.28	0.55	0.26
2/4	0.83	0.26	315	0.68	0.23	0.69	0.24	0.67	0.23
2/5	0.99	0.29	360	0.85	0.27	0.86	0.27	0.85	0.27

2.2. Other Water Quality Analysis

NO_3^- -N level steadily increased in the systems throughout the study (Figure 2.12), NO_3^- -N reaching the maximum mean value of 163 mg/L in Part 1 at 12 Lpm applied, and 238 mg/L in Part 2 at 360 mg/m^2/day TAN applied. These values are below the upper limit of the target range used for this study (Table 2.2).

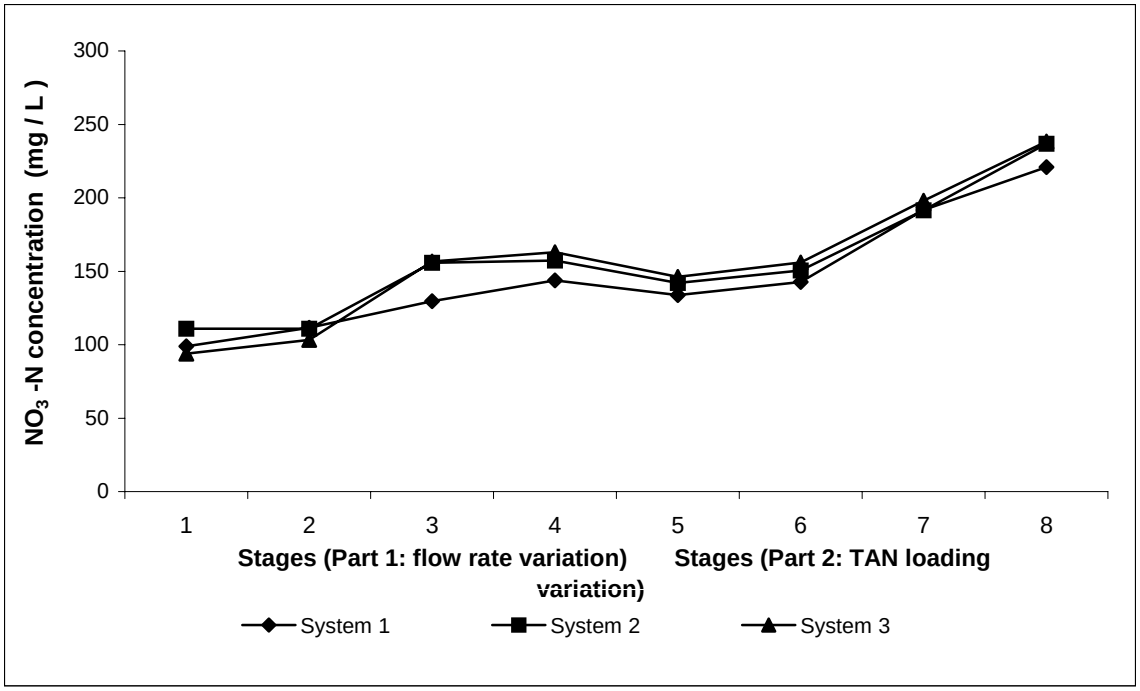

Figure 2.12. Nitrate (NO_3^- -N) concentration means seen in the Part 1 and in Part 2 of the experiment. In Part 1, 180 mg/L–day TAN was applied constantly at four different flow rates: 1 (6 Lpm), 2 (8 Lpm), 3 (10 Lpm), and 4 (12 Lpm), respectively. In Part 2, 12 Lpm flow rate was applied constantly at five different influent TAN concentrations: 4 (180 mg/L-day), 5 (225 mg/L-day), 6 (270 mg/L-day), 7 (315 mg/l-day), and 8 (360 mg/L-day), respectively.

NO_3^- -N concentration was directly controlled by the daily exchange of water (10%). Water exchange also contributed directly to TAN and NO_2^- -N reductions, but TAN and NO_2^- -N control was probably more a function of microbial oxidation.

Temperature and pH values for all systems typically ranged from 25.1 – 30.7 °C and 7.4 – 8.3, respectively. The temperature fluctuations were similar in all treatments. The high values of pH maintained throughout the study were a function of the bicarbonate added and the carbon dioxide stripping due to aeration of the feed tanks. D.O. values were maintained between 6.4 – 7.6 mg/L during the experiment. Despite the high water temperature, these values were possible due to the strong aeration of the tanks. D.O. was not a limiting factor for biofilter performance at these concentrations (Losordo, 1991; Kaiser and Wheaton, 1993). Alkalinity and hardness values ranged from 90 – 150 and 128 – 142 mg/L , respectively, in all systems. Alkalinity fluctuations resulted from nitrification effects, and repeated $NaHCO_3$ addition was necessary. These ranges are

considered biologically suitable for both fish and nitrifiers (Meade, 1989; Klontz et al., 1979).

2.3. Hydraulic Analysis

Pumping up through a column mean that it has to be pumped against the headloss in the bottom plate, the headloss in the beads and the headloss in the water column. Filter headloss recorded in this experiment varied as a function of flow rate and the degree of media fluidization. The values ranged between 4.6 cm in biofilter D_1 at 6 Lpm and 1.8 cm in biofilter D_3 at 12 Lpm (Table 2.8). These values are lower than those reported in the literature for sand beds, (15 to 50 cm; Chang, 1999), and are in agreement with previous findings that headloss is linearly related to filter flow rate.

Table 2.8. The differences in headloss values (cm) for each biofilter.

Biofilter	Flow rate, Lpm			
	6	8	10	12
D_1	4.6	4.3	4.0	3.8
D_2	3.6	3.3	3.0	2.8
D_3	2.6	2.3	2.0	1.8

Hydraulic residence time (HRT) ranged between 2.31 minutes in biofilter D_1 at 12 Lpm (37.14% TAN removal) to 1.19 minutes in biofilter D_3 at 6 Lpm (62.76% TAN removal). At 2.31 minutes HRT, the water velocity was 57.6 m/h in filter D_1, and 14.4 m/h in filter D_3 at 1.19 minutes HRT. Figure 2.13 show how HRT varied in the three columns at four different flow rates. The TAN percent removals were lower in this experiment than seen in the literature. Gauntlett (1981) reported 93% TAN removal from drinking water, using a raw water containing 2 mg /L TAN, and an upflow rate of 12 m/h. The better performance could be due to the larger surface area, larger HRT, and the higher concentration of TAN in the bulk water of Gauntlett's experiments.

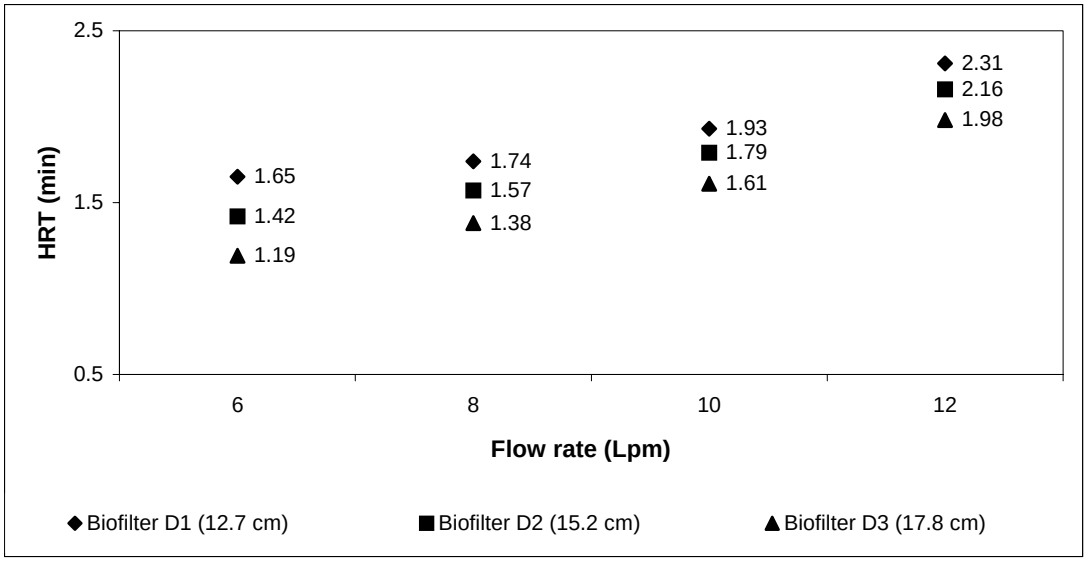

Figure 2.13. HRT versus various flow rates in Part 1.

The percent bed expansion increased approximately linearly with water velocity for each biofilter. The bed levels recorded at different flow rates are presented in Figure 2.14. Among biofilters, the highest water velocities were measured in biofilter D_1, which resulted in the highest increase in percent bed expansion (Figure 2.15). The small density difference between the particles, biofilm and the suspending fluid (water), showed no effect on bed fluidization level. This was more obvious in the second part, when the flow rate was kept constant, and the biofilm accumulated. These observations are in agreement with the findings of Atkinson et al. (1981) in their experiments with plastic media. Atkinson et al. (1981) also reported differences of up to 260% between the initial buoyancy of sand and the final buoyancy of sand particles in a fluidized filter that were coated with biofilm. These findings suggested that the fluidization of plastic beads is more constant than in sand biofilters.

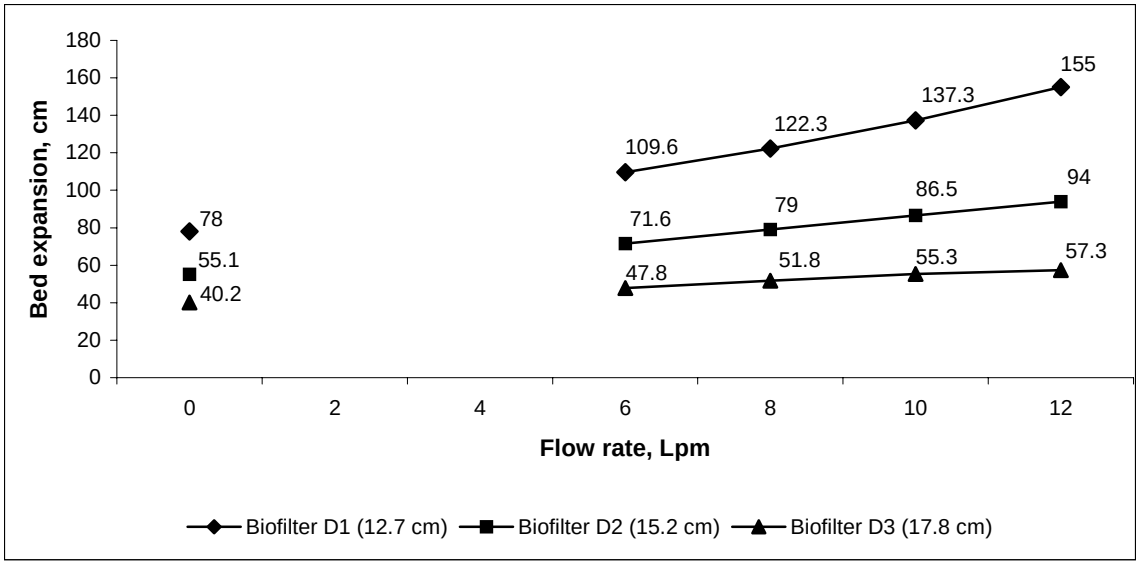

Figure 2.14. Bed height (cm) at various flow rates (Lpm).

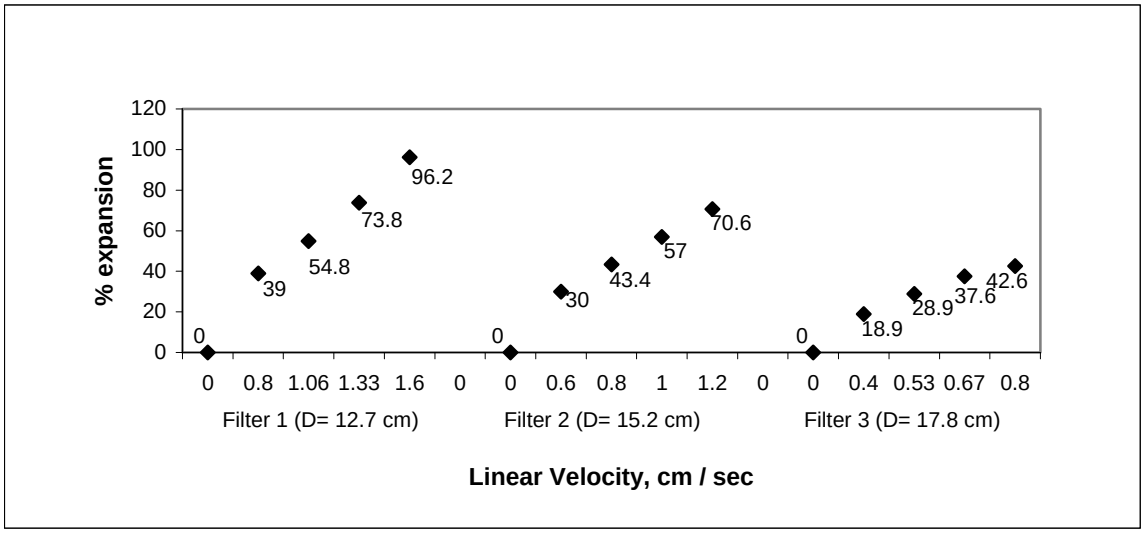

Figure 2.15. Percent bed expansion at various water velocities (cm/sec) in the three biofilters.

2.4. Biofilm Thickness Analysis

Two trends were noted from biofilm measurements. During Part 1, the average biofilm thickness decreased significantly ($p<0.0001$) as flow rate was increased and the average TAN loading was decreased (Figure 2.17). The biofilm thickness varied between 76.4 microns in biofilter D_3 at 6 Lpm, and 44.2 microns in biofilter D_1 at 12 Lpm.

Obviously, the attrition was higher in biofilter D_1, which experienced the highest water velocities. The balance between overall growth rate and the degree of attrition resulted in a thinner biofilm. During Part 2, when influent TAN flux increased under conditions of constant flow rate (12 Lpm), the biofilm accumulated. The average biofilm thickness increased significantly ($p<0.0001$), as shown in Figure 2.18. The biofilm thickness varied between 44.2 microns in biofilter D_1 at 180 mg/L TAN applied, and 90.1 microns in biofilter D_3 at 360 mg/L applied. The results obtained in Part 1 of this experiment are in agreement with Atkinson and Knights (1975), who reported that microbial layer thickness depends upon bed expansion (attrition) when exposed to a fixed TAN concentration (constant growth rate). The results found in Part 2 agreed with Atkinson and Davies (1972) who found that for particles exposed to constant attrition in a fluidized bed, microbial layer thickness depends on substrate concentration; i.e., depends on grow rate.

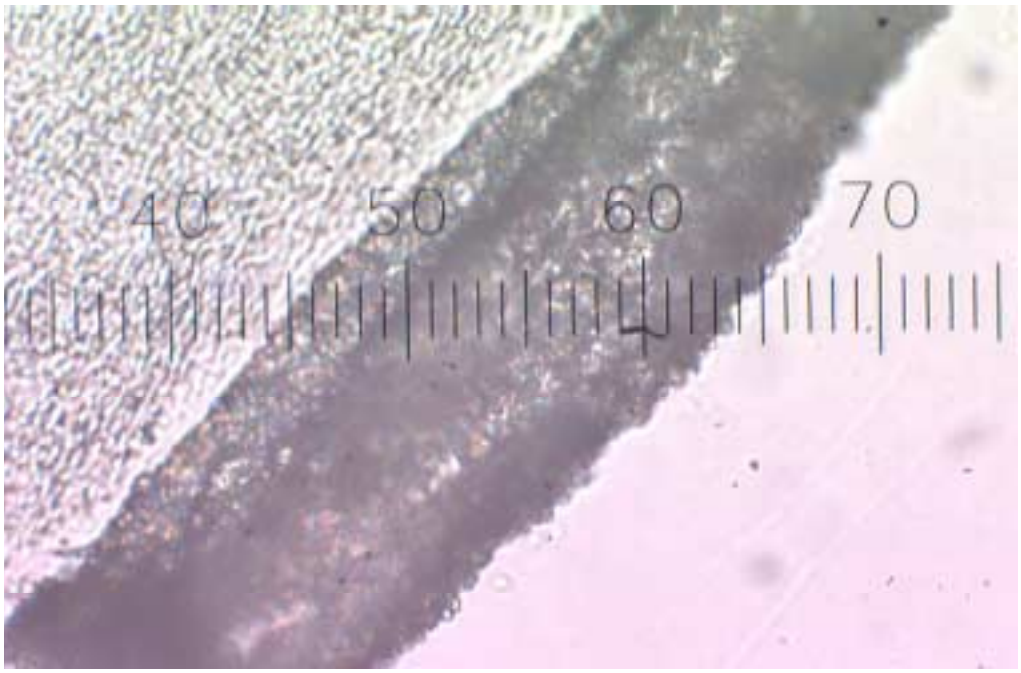

Figure 2.16. Cross-section of a nitrifying biofilm attached to a plastic bead surface. Scale: 1 unit = 4 μm.

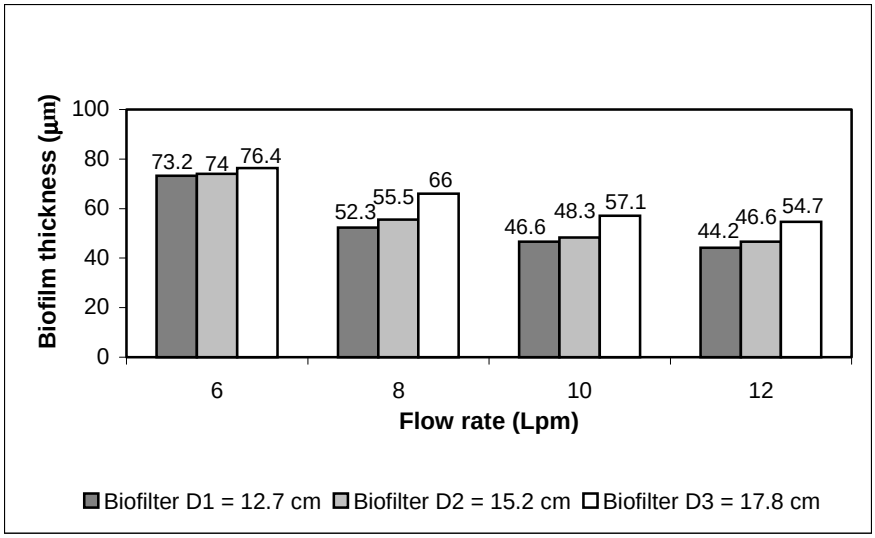

Figure 2.17. Biofilm thickness variation at varied flow rates applied in Part 1.

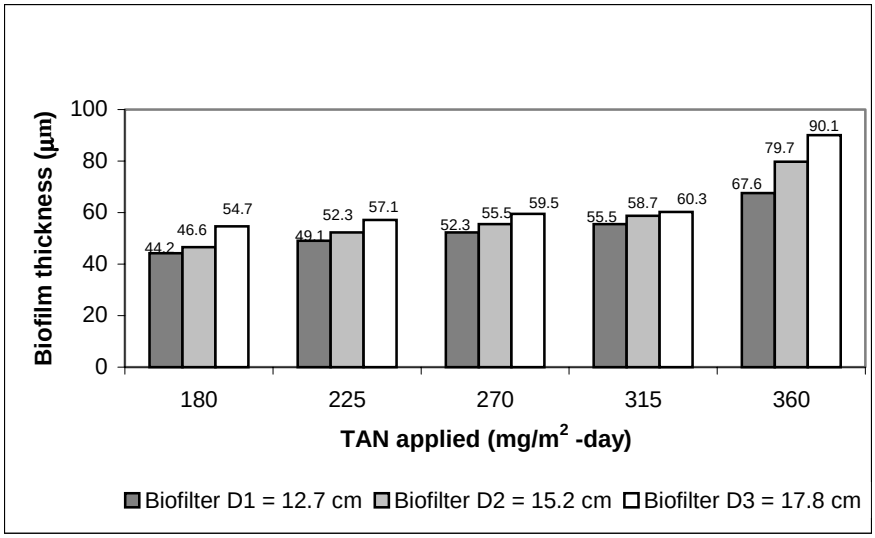

Figure 2.18. Biofilm thickness variation at varied TAN concentrations applied in Part 2.

3. Compatibility with an Existing Model

A comparison was made between the biofilm thickness recorded in this experiment and the theoretical model for a steady state biofilm developed by Rittmann and McCarty (1981). The steady state biofilm was based on diffusion transport and mass balance principles. The model assumed uniform cell density in the biofilm, locally uniform biofilm thickness, and all required nutrients are in excess concentration with the exception of the rate-limiting substrate (TAN, in this case). Three steady-state biofilms (biofilters D_1, D_2, and D_3) were compared with the model predictions. Table 2.9 presents

the influent concentrations (S), the flux of substrate into the biofilm (J), biofilm thickness predicted (L_f), and the biofilm thickness measured in the three biofilters (L_{fD1}, L_{fD2}, and L_{fD3}) in four stages of Part 1, and the five stages of Part 2 experiments.

To obtain the values of J and L_f from Table 2.9, a complex iterative calculation process was needed (Rittmann and McCarty, 1981). Thirteen parameters were used for the calculation by a computer program, including: substrate (TAN), coefficient of total biomass loss (b'), substrate mass diffusivity in water (D) and in biofilm (D_f), maximum specific rate of substrate utilization (k), half saturation constant (K_s), water film thickness (L), bacterial density in biofilm (X_f), true yield of bacterial mass per unit of substrate mass utilized (Y), minimum bulk concentration of rate-limiting substrate able to sustain a steady-state biofilm (S_{min}), particle diameter (d_p), absolute viscosity of liquid (μ), mass density of liquid (ρ) and hydrodynamic diffusivity (D_H). A detailed description of the procedure can be found in Rittmann and McCarty (1980 a) and Rittmann (1982). The values of the parameters selected for the simulation in this study are provided in Table 2.10. A standard deviation, as found for multiple evaluations, is given for K, K_s and S_{min} (Table 2.10). Steady-state results for L_f for biofilters D_1, D_2 and D_3 at varied flow rates and TAN concentration applied are presented in Figures 2.19 and 2.20, respectively, illustrating the comparison between predicted L_f and the biofilm thickness found in the experiment. The predicted L_f was higher than the data points recorded for each biofilter, in both parts (1 and 2) of the experiment. Among the biofilters, L_f decreased with water velocity increase, from biofilter D_3 to D_1. This suggests that L_f depends on the attrition intensity among the beads. Rittmann (1982) found that S_{min} and L_f are reduced and substrate-removal efficiency decline, when shear stress became important.

The results seen in this experiment represent the values for ideal nitrification conditions of a single substrate-limiting factor and a single type of bacteria (nitrifiers). However, waters in actual recirculating aquaculture systems usually contain high biochemical oxygen demand (BOD) concentrations that provide substrate for heterotrophic bacteria, which compete with nitrifiers for growing space, oxygen and other nutrients. This tends to lower the ntirifier density value (X_f) in the biofilm. Heisenbroek and Kamstra (1990) found a lower ammonia removal rate in the trickling filters of eel farms than in a biofilm fed with a synthetic, non-organic wastewater. During this

experiment, only tap and well water and necessary nitrification substrate nutrients were used in the parallel reactor systems. The experimental results were obtained without the influence of elevated BOD concentrations. TAN removal data in this study were therefore much higher than would be expected in a commercial system.

Table2.9. Substrate flux (J) and biofilm thickness (L_f) predicted through steady-state modeling.

Flow rate applied (Lpm)	S (mg/l)	J (mg/cm^2-day)	Predicted	L_f, mμ Biofilter D_1*	Biofilter D_2**	Biofilter D_3***
6	0.6	0.092	72.3	73.2	74.0	76.4
8	0.57	0.086	68.0	52.3	55.5	66.0
10	0.53	0.079	62.2	46.6	48.3	57.1
12	0.5	0.074	57.9	44.2	46.6	54.7
TAN flux applied (mg/m^2-day)	S(mg/l)	J (mg/cm^2-day)	Predicted	L_f, mμ Biofilter D_1*	Biofilter D_2**	Biofilter D_3***
180	0.5	0.074	57.9	44.2	46.6	54.7
225	0.52	0.077	60.8	49.1	52.3	57.1
270	0.69	0.108	85.5	52.3	55.5	59.5
315	0.83	0.133	105.2	55.5	58.7	60.3
360	0.99	0.162	127.3	67.6	79.7	90.1

D1* = 12.7 cm diameter
D2** = 15.2 cm diameter
D3***= 17.8 cm diameter

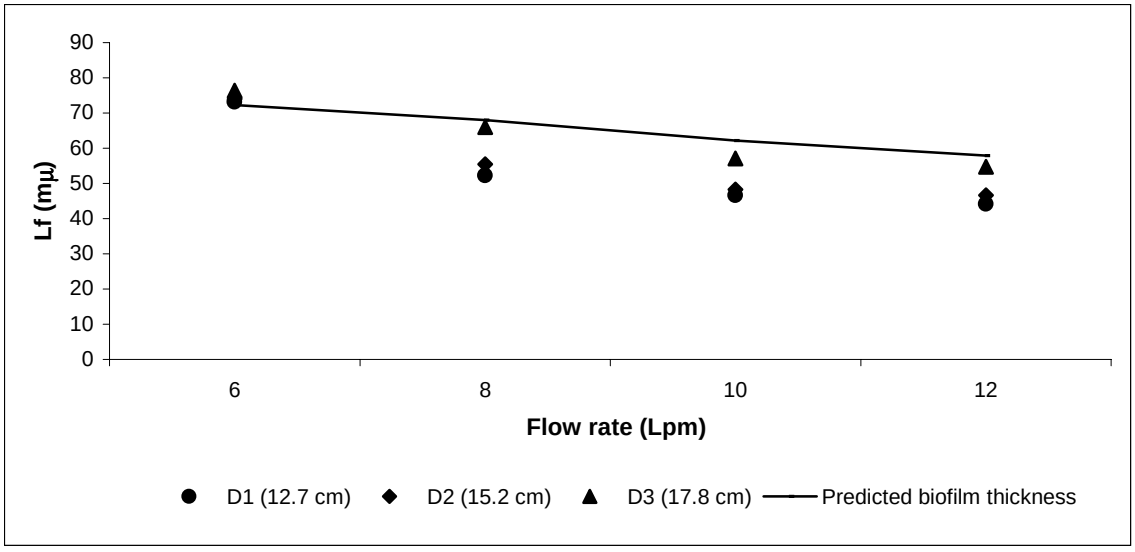

Figure 2.19. Experimental (symbol) and predicted (line) biofilm thickness profile from Part 1 of the study.

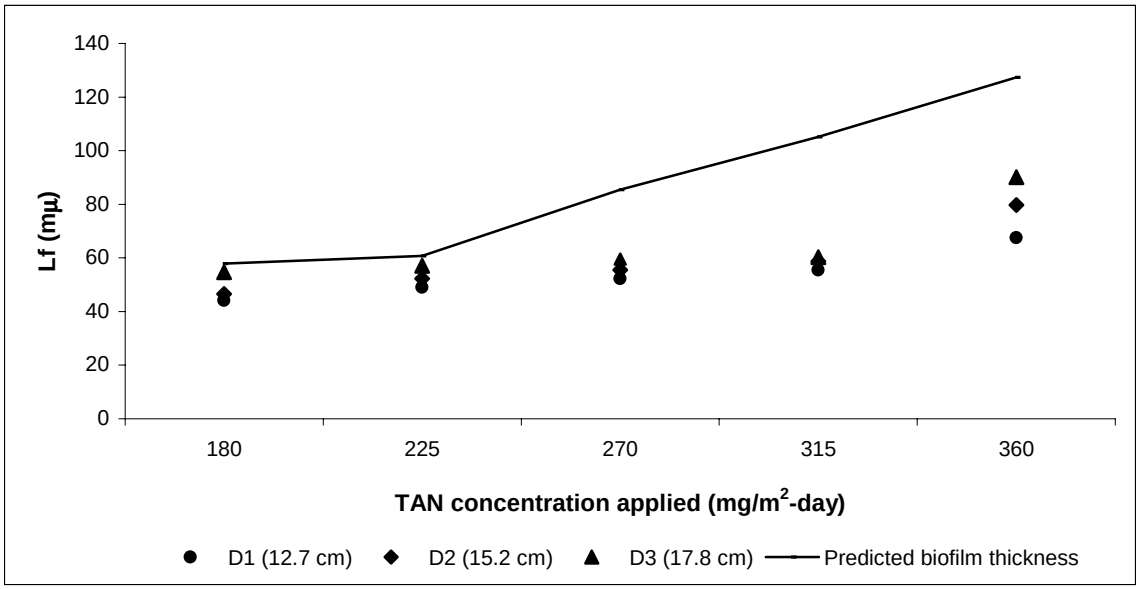

Figure 2.20. Experimental (symbol) and predicted (line) biofilm thickness profile from Part 2 of the study.

Table 2.10. Conditions for comparison of ammonia nitrification.

Parameter	Value used	Literature value	Reference
Substrate	Ammonia		
b' (1/day)	0.205 ± 0.049	0.05	Kissel et al. (1984)
		0.205	Rittmann and McCarty (1980)
D (m2/day)	0.00013	0.00013	Ritmmann and Manem (1992)
		0.00015	Chen et al. (1989)
		0.000157	Kissel et al (1984)
D_f (m2/day)	0.000104	0.000104	Rittmann and Manem (1992)
		0.00012	Chen et al. (1989)
		0.000126	Kissel et al (1984)
k (g/g TAN-day)	20	20 ± 4.4	Rittmann and McCarty (1980)
K_s (mg/L)	0.0035	0.0039 ± 0.0008	Rittmann and McCarty (1980)
		2	Zhu and Ken (1999)
L (m)	0.000054	0.000054	Rittmann and Manem (1992)
		0.00012	Chen et al. (1989)
X_f (mg/L)	12700	12700	Rittmann and Manem (1992)
		50100	Chen et al. (1989)
Y (g cell/g TAN)	0.2	0.2	Rittmann and Manem (1992)
		0.17	WPCF (1983)
		0.21	Gujer and Boller (1986)
S_{min} (mg/cm3)	0.00066	0.00066	Rittmann (1982)
		0.00066 ± 0.28	Rittmann and McCarty (1980)
d_p (cm)	0.3	0.3 (fluidized bed)	Rittmann (1982)
μ (g/cm-day)	864	864	Rittmann (1982)
ρ (g/cm3)	1	1	Rittmann (1982)
D_H (cm2/day)	10	10 (complete mixt)	Rittmann (1982)

4. Performance versus Filter Design and Operational Characteristics

TAN and NO_2^--N conversion rates are used principally for evaluation and comparison of biofilter performance in RAS. Fluidized bed filter performance can vary dramatically with loading and management. A good biofilm management plan must address the needs of the nitrifying bacteria in terms of water quality, nutrient transport and biofilm harvesting. Periodic monitoring of the biofilters performance facilitates optimization in situations where peak nitrification performance is demanded. Approaching the sizing of floating bead (and other) biofilters in terms of volumetric nitrification capacity, Malone et al. (1999) reported area conversion rates with of about 300 mg TAN/m^2-day in RAS with effluent TAN and nitrite levels between 0.5 and 1.0 mg N/L. Other values reported

for TAN conversion rate were between 60 mg TAN/m^2-day (DeLosReyes and Lawson, 1996) and 440 mg TAN/m^2-day (Wimberly, 1990). The values from this study (between 180 and 360 mg TAN/m^2-day volumetric conversion rates) corresponding to maximum TAN and NO_2^- -N concentrations in the feed tank of 0.99 and 0.29, respectively, suggest that using fluidized bed filters with plastic medium can be at least as efficient for aquaculture as other biofilter types. However, volumetric conversion rates are assumed to decline with decreasing substrate concentration, i.e., TAN diffusion limitation. The values can be expected to hold for the systems where the temperature is maintained over 25 °C and effluent D.O. is higher than 2 mg/L. The actual level of nitrification occurring in the RAS filters may be higher since TAN also results from the breakdown of nitrogenous organic compounds by heterotrophs. Thus, this aspect should also be considered in sizing and designing fluidized filters.

Based on the results of this study, a lower fluidizing velocity (given by a large reactor diameter) and a high flow rate yielded the best nitrification efficiency. The lower fluidization rate showed a better protection of the biofilm from destruction by attrition. A higher flow rate allowed for faster recirculation of water (a maximum of four passes per hour in this study), through yielding an improved nitrification efficiency. The small size of the beads and their density close to that of water were also important factors in biofilm conservation. The small size of the beads used (2 – 4 mm) provided an expanded nitrification surface per unit volume (1600m^2/m^3), which is approximately 25% greater than plastic beads used in floating bead filters. The neutral density also favored very small headlosses for a bed of this material (Table 2.8). As previously stated, each system utilized a 1/6 Hp pump with flow rate being adjusted to achieve similar flows between all filter types. The pump output was actually much restricted.

With regard to biofilter design, the largest diameter reactor (biofilter D_3) had a significantly higher TAN and NO_2^- -N removal efficiency than the other two diameters at a the lowest flow rate (6 Lpm); but, all three showed similar removal efficiencies as the flow rate was increased to 12 Lpm. Despite this similarity, a larger diameter of filter is still be desired, because a lower water velocity yields less bed expansion and the need for less volume. In this study, at similar flow rates, a column diameter of 17.8 cm (D_3) had a percent bed expansion at fluidization of only 42.6%, as compared to 96.2% with the 12.7

cm diameter column (D_1). Also, the head loss in biofilter D_3 was 15% less than in biofilter D_1 at 6 Lpm. The percent difference was even larger (30%) at 12 Lpm. Based on these data, an ideal fluidized bed filter with plastic medium would be filled to about 40 – 50% capacity with the medium (no flow) and 1 to 2 cm/sec at the desired flow rate. A biofilter with these characteristics should be able to achieve TAN removal of 25%, which is usually targeted when high nitrification performance is demanded (Malone et al., 1999). The biofilter design should combine the economics of construction with nitrification performances, while maintaining TAN and NO_2^- -N levels at values not harmful for the crop in a RAS.

In this study, the three biofilters of each system shared the maximum surface area calculated as necessary for nitrification. Under actual conditions, for safety reasons and flexibility, the entire volume of plastic medium necessary should be distributed among three or four columns operated in parallel. However, a supplementation of the necessary surface area is recommended. The amount of the medium should be large enough that the failure of one of the filters does not significantly impact on overall system performance. Another important consideration is the presence of organic substances and solids in the RAS, which could decrease the density of nitrifiers in biofilm due to the proliferation of heterotrophs. Heterotrophs exhibit faster growth rates than nitrifiers in organic-laden waters. According to Manem and Rittmann (1992), heterotrophs will growth on top of existing biofilm layer forcing nitrifiers to reside at deeper biofilm depths. Malone et al. (1993) attributed inhibition of oxygen and nutrient transport to deeper biofilm depths as the cause of nitrification reduction.

However, for a better understanding of the economics of this technology, an evaluation at a larger scale under presence of aquatic organism is suggested.

D. SUMMARY AND CONCLUSIONS

Fluidized bed filters with plastic bead medium proved to be effective in removing ammonia and nitrite from a synthetic aquaculture water. Ammonia and nitrite were maintained within ranges considered biologically suitable for both fish and nitrifiers. The results of this study demonstrated that an improvement in nitrification performance occurs in fluidized bed filters with increased hydraulic loading conditions (6 to 12 Lpm) and with increased ammonia supply (180 mg/m^2-day to 360 mg/m^2-day). These results were produced under conditions of no organic carbon loading, except for the carbon associated with biomass. Under fish production conditions, an increase in organic loading will occur in the nitrifying reactor and consequently will lower nitrification.

The following statements summarize the major findings of this study:

1. Influent TAN levels slightly decreased (from 0.6 to 0.5 mg/L) under conditions of constant ammonia flux (180 mg/m^2-day) and flow rate increase from 6 to 12 Lpm. TAN removal efficiency was between 13.68 and 62.76 %. Influent NO_2^- -N followed a similar pattern, but the effluent concentrations for all biofilters showed more variability from day to day than TAN. The NO_2^- -N fluctuations were similar in all filters and remained at a level lower than 0.3 mg/L. The largest biofilter (D_3) removed ammonia best at the lowest flow rate. However, the difference in TAN removal performance for the three biofilters decreased as flow rate increased.

2. Under conditions of ammonia flux variations from 180 mg/ m^2-day to 360 mg/ m^2-day and flow rate of 12 Lpm, influent TAN accumulated from 0.5 to 0.99 mg/L and removal efficiency was between 13.21 and 20.02 %. TAN removals increased slightly as influent TAN flux was increased and then began to decrease as TAN flux was increased above 270 mg/m^2-day. Influent NO_2^- -N was between 0.19 and 0.29 mg/L and effluent NO_2^- -N ranged between 0.17 and 0.28 mg/L. Percent NO_2^- -N removal ranged between 49.8 and –30.4 % (negative values

indicating NO_2^- -N addition in the filter). Essentially, there was no difference in performances of the three column diameters.

3. Biofilm thickness showed deterioration as flow rate was increased and accumulated as ammonia supply increased. The model used (Rittmann and McCarty, 1980) predicted higher values of the biofilm thickness (L_f) for given influent concentrations (S) at substrate flux (J) than measured in this study.

4. A fluidized bed filter with plastic beads provides good removal efficiency of TAN and low head loss if designed as a large diameter column operating at flow rates that allow for a water velocity of 1 - 2 cm/sec. The ideal system will operate with three or four reactors in parallel, that are each filled to about 40 - 50% of capacity at no flow.

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APPENDIX A: RAW DATA

Table A1. Acclimation period raw data; TAN and NO_2^--N variation.

	TAN variation			NO_2 variation		
Day	System 1	System 2	System 3	System 1	System 2	System 3
1	0.50	0.50	0.50	0.00	0.01	0.00
2	1.00	1.00	1.00	0.00	0.03	0.00
3	1.00	1.00	1.00	0.02	0.03	0.01
4	0.70	0.99	1.00	0.02	0.04	0.03
5	0.95	1.25	1.59	0.02	0.04	0.03
6	1.02	1.30	1.88	0.03	0.07	0.05
7	1.05	1.32	2.20	0.04	0.06	0.06
8	1.20	1.12	2.32	0.04	0.07	0.08
9	1.45	1.66	2.44	0.06	0.08	0.08
10	1.34	1.95	2.02	0.08	0.10	0.09
11	1.13	1.86	2.12	0.09	0.15	0.13
12	1.55	1.62	2.30	0.11	0.16	0.12
13	1.96	1.76	2.56	0.15	0.16	0.14
14	1.45	1.80	2.48	0.18	0.18	0.15
15	1.89	1.95	2.58	0.26	0.19	0.15
16	1.96	1.82	2.65	0.29	0.21	0.16
17	2.35	1.66	2.71	0.33	0.21	0.21
18	2.38	1.79	2.95	0.35	0.23	0.26
19	2.71	2.12	3.08	0.39	0.26	0.32
20	2.80	2.20	3.12	0.46	0.25	0.62
21	2.88	2.32	3.35	0.55	0.25	1.15
22	1.92	2.44	2.22	1.30	0.33	0.62
23	1.40	2.85	1.62	2.22	0.56	1.98
24	0.95	2.84	1.16	3.12	0.62	2.86
25	1.22	2.95	0.96	3.78	0.78	3.35
26	1.02	2.12	0.55	5.00	1.15	3.85
27	0.66	1.45	0.42	5.62	2.14	4.52
28	0.44	1.30	0.22	6.26	3.80	4.89
29	0.35	1.61	0.59	7.32	5.18	5.15
30	0.44	0.75	0.40	7.96	6.05	6.03
31	0.32	0.56	0.52	8.92	6.75	6.80
32	0.34	0.51	0.45	9.62	7.02	7.16
33	0.44	0.45	0.32	10.22	7.85	7.86
34	0.31	0.42	0.32	10.95	8.22	8.15
35	0.27	0.44	0.35	11.70	8.52	8.76
36	0.35	0.36	0.39	7.35	8.92	9.10
37	0.39	0.52	0.42	5.12	9.32	9.45

38	0.40	0.63	0.61	3.33	6.54	9.68
39	0.62	0.45	0.75	2.15	4.65	10.15
40	0.56	0.40	0.56	1.75	2.11	6.95
41	0.48	0.41	0.45	1.05	1.85	5.45

Table A 1. (cont.) Acclimation period raw data; TAN and NO_2 variation.

	TAN variation			NO2 variation		
Day	System 1	System 2	System 3	System 1	System 2	System 3
42	0.51	0.52	0.45	0.69	1.14	2.86
43	0.66	0.62	0.49	0.62	0.62	1.95
44	0.55	0.60	0.52	0.51	0.45	1.12
45	0.52	0.55	0.61	0.45	0.36	0.72
46	0.63	0.45	0.55	0.33	0.33	0.70
47	0.70	0.36	0.61	0.27	0.20	0.46
48	0.65	0.46	0.48	0.26	0.25	0.42
49	0.52	0.52	0.46	0.23	0.28	0.32
50	0.50	0.50	0.43	0.23	0.24	0.27
51	0.48	0.55	0.51	0.26	0.19	0.26
52	0.52	0.48	0.48	0.22	0.24	0.29
53	0.59	0.52	0.55	0.28	0.32	0.26

Table A2. TAN removal performance experiments row data; Part 1 (180 mg/m^2-d TAN applied at various flow rates).

6 Lpm
SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.62	32.2	0.42	29.0	0.44	58.0	0.26
2	0.60	33.3	0.40	25.0	0.45	60.0	0.24
3	0.62	35.4	0.40	32.2	0.42	64.5	0.22
4	0.47	40.4	0.28	25.5	0.35	59.5	0.19
5	0.54	35.1	0.35	25.9	0.40	66.6	0.18
6	0.49	38.7	0.30	24.4	0.37	61.2	0.19
7	0.59	33.8	0.39	28.8	0.42	61.0	0.23
AVG.	0.56	35.6	0.36	27.2	0.40	61.5	0.21

8 Lpm
SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.59	28.8	0.42	27.1	0.43	47.4	0.31
2	0.57	26.3	0.42	24.5	0.43	38.5	0.35
3	0.59	30.5	0.41	23.7	0.45	35.5	0.38
4	0.61	27.8	0.44	22.9	0.47	34.4	0.40
5	0.52	25.0	0.39	21.1	0.41	30.7	0.36
6	0.61	22.9	0.47	21.3	0.48	36.0	0.39
7	0.57	26.3	0.42	24.5	0.43	47.3	0.30
AVG.	0.58	26.8	0.42	23.6	0.44	38.6	0.33

10 Lpm
SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.51	19.6	0.41	21.5	0.40	33.3	0.34
2	0.51	15.6	0.43	17.6	0.42	23.5	0.39
3	0.54	22.2	0.42	18.5	0.44	31.4	0.37
4	0.53	24.5	0.40	24.5	0.40	30.1	0.37
5	0.51	21.5	0.40	17.6	0.42	25.4	0.38
6	0.56	21.4	0.44	21.4	0.44	30.3	0.39
7	0.51	19.6	0.41	17.6	0.42	29.4	0.36
AVG.	0.52	20.6	0.41	19.8	0.42	29.1	0.37

12 Lpm
SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.50	10.0	0.45	10.0	0.45	14.0	0.43
2	0.44	13.6	0.38	6.81	0.41	9.0	0.40
3	0.52	15.3	0.44	11.5	0.46	19.2	0.42
4	0.51	21.5	0.40	15.6	0.43	17.6	0.42
5	0.48	14.5	0.41	12.5	0.42	16.6	0.40

6	0.50	18.0	0.41	16.0	0.42	22.0	0.39
7	0.50	16.0	0.42	14.0	0.43	18.0	0.41
AVG.	0.49	15.5	0.41	12.3	0.43	16.6	0.41

6 Lpm
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.67	37.3	0.42	29.8	0.47	62.6	0.25
2	0.71	40.8	0.42	30.9	0.49	66.1	0.24
3	0.66	39.3	0.40	34.8	0.43	66.6	0.22
4	0.71	43.6	0.40	35.2	0.46	66.1	0.24
5	0.68	38.2	0.42	32.3	0.46	63.2	0.25
6	0.48	33.3	0.32	27.0	0.35	60.4	0.19
7	0.64	32.8	0.43	26.5	0.47	57.8	0.27
AVG.	0.65	37.9	0.40	30.9	0.44	63.3	0.23

8 Lpm
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.54	33.3	0.36	33.3	0.36	42.5	0.31
2	0.59	32.2	0.40	25.4	0.44	44.0	0.33
3	0.62	27.4	0.45	29.0	0.44	37.0	0.39
4	0.60	30.0	0.42	26.6	0.44	31.6	0.41
5	0.57	31.5	0.39	31.5	0.39	36.8	0.36
6	0.58	29.3	0.41	29.3	0.41	43.1	0.33
7	0.60	35.0	0.39	30.0	0.42	43.3	0.34
AVG.	0.58	31.2	0.40	29.3	0.41	39.8	0.35

10 Lpm
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.58	24.1	0.44	20.6	0.46	27.5	0.42
2	0.53	24.5	0.4	24.5	0.40	22.6	0.41
3	0.54	20.3	0.43	18.5	0.44	29.6	0.38
4	0.49	16.3	0.41	20.4	0.39	24.4	0.37
5	0.58	22.4	0.45	20.6	0.46	27.5	0.42
6	0.53	22.6	0.41	18.8	0.43	26.4	0.39
7	0.51	21.5	0.4	19.6	0.41	29.4	0.36
AVG.	0.53	21.7	0.42	20.4	0.42	26.8	0.39

12 Lpm
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.52	13.4	0.45	11.5	0.46	21.1	0.41
2	0.49	18.3	0.40	14.2	0.42	20.4	0.39
3	0.52	19.2	0.42	11.5	0.46	19.2	0.42
4	0.52	19.2	0.42	19.2	0.42	21.1	0.41
5	0.51	13.7	0.44	13.7	0.44	15.6	0.43
6	0.51	17.6	0.42	17.6	0.42	23.5	0.39
7	0.50	16.0	0.42	20.0	0.40	22.0	0.39
AVG.	0.51	16.8	0.42	15.4	0.43	20.47	0.40

6 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.63	34.9	0.41	28.5	0.45	58.7	0.26
2	0.63	34.9	0.41	26.9	0.46	58.7	0.26
3	0.62	37.0	0.39	27.4	0.45	67.7	0.20
4	0.64	40.6	0.38	29.6	0.45	67.1	0.21
5	0.62	41.9	0.36	25.8	0.46	67.7	0.20
6	0.56	41.0	0.33	33.9	0.37	67.8	0.18
7	0.61	34.4	0.40	27.8	0.44	55.7	0.27
AVG.	0.61	37.8	0.38	28.6	0.44	63.3	0.22

8 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.51	27.4	0.37	29.4	0.36	43.1	0.29
2	0.57	33.3	0.38	29.8	0.40	47.3	0.30
3	0.59	27.1	0.43	27.1	0.43	38.9	0.36
4	0.59	30.5	0.41	28.8	0.42	35.5	0.38
5	0.61	26.2	0.45	26.2	0.45	39.3	0.37
6	0.63	30.1	0.44	28.5	0.45	41.2	0.37
7	0.60	30.0	0.42	28.3	0.43	40.0	0.36
AVG.	0.58	29.2	0.41	28.3	0.42	40.8	0.34

10 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.56	26.7	0.41	16.0	0.47	28.5	0.40
2	0.54	20.3	0.43	16.6	0.45	22.2	0.42
3	0.58	24.1	0.44	15.5	0.49	31.0	0.40
4	0.53	20.7	0.42	18.8	0.43	26.4	0.39
5	0.58	18.9	0.47	17.2	0.48	24.1	0.44
6	0.57	15.7	0.48	10.5	0.51	17.5	0.47
7	0.53	20.7	0.42	22.6	0.41	26.4	0.39
AVG.	0.50	21.0	0.43	16.7	0.46	25.1	0.41

12 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.50	10.0	0.45	14.0	0.43	18.0	0.41
2	0.50	16.0	0.42	12.0	0.44	12.0	0.44
3	0.54	16.6	0.45	11.1	0.48	20.3	0.43
4	0.53	18.8	0.43	16.9	0.44	20.7	0.42
5	0.52	15.3	0.44	11.5	0.46	19.2	0.42
6	0.52	13.4	0.45	13.4	0.45	19.2	0.42
7	0.51	17.6	0.42	13.7	0.44	15.6	0.43
AVG.	0.51	15.4	0.43	13.2	0.44	17.8	0.42

Table A3. TAN removal performance experiment row data; Part 2 (12 Lpm applied at varied TAN loadings).

225 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.56	19.6	0.45	17.8	0.46	21.4	0.44
2	0.50	22.0	0.39	10.0	0.45	16.0	0.42
3	0.51	19.6	0.41	17.6	0.42	23.5	0.39
4	0.52	21.1	0.41	17.3	0.43	15.3	0.44
5	0.50	18.0	0.41	14.0	0.43	22.0	0.39
6	0.49	20.4	0.39	20.4	0.39	24.4	0.37
7	0.48	27.0	0.35	27.0	0.35	25.0	0.36
AVG.	0.50	21.1	0.40	17.7	0.41	21.1	0.40

270 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.67	16.4	0.56	14.9	0.57	16.4	0.56
2	0.69	18.8	0.56	18.8	0.56	21.7	0.54
3	0.73	21.9	0.57	23.2	0.56	21.9	0.57
4	0.68	14.7	0.58	14.7	0.58	19.1	0.55
5	0.73	13.6	0.63	9.5	0.66	10.9	0.65
6	0.67	20.8	0.53	22.3	0.52	22.3	0.52
7	0.66	21.2	0.52	18.1	0.54	19.6	0.53
AVG.	0.69	18.2	0.56	17.4	0.57	18.8	0.56

315 mg/m^2-d
SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.84	16.6	0.70	16.6	0.70	17.8	0.69
2	0.89	17.9	0.73	15.7	0.75	19.1	0.72
3	0.85	24.7	0.64	22.3	0.66	24.7	0.64
4	0.80	20.0	0.64	18.7	0.65	20.0	0.64
5	0.81	18.5	0.66	19.7	0.65	20.9	0.64
6	0.81	16.0	0.68	14.8	0.69	17.2	0.67
7	0.84	16.6	0.70	15.4	0.71	17.8	0.69
AVG.	0.83	18.6	0.67	17.6	0.68	19.6	0.67

360 mg/m^2-d
SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.99	9.0	0.90	10.1	0.89	11.1	0.88
2	0.97	14.4	0.83	15.4	0.82	16.4	0.81
3	0.98	11.2	0.87	10.2	0.88	13.2	0.85
4	0.95	13.6	0.82	11.5	0.84	12.6	0.83
5	1.03	12.6	0.90	11.6	0.91	13.5	0.89
6	1.06	13.2	0.92	13.2	0.92	14.1	0.91
7	1.08	12.0	0.95	13.8	0.93	14.8	0.92

AVG.	1.00	12.3	0.88	12.3	0.88	13.7	0.87

225 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.55	16.3	0.46	21.8	0.43	25.4	0.41
2	0.49	22.4	0.38	16.3	0.41	18.3	0.40
3	0.52	21.1	0.41	19.2	0.42	23.0	0.40
4	0.53	22.6	0.41	16.9	0.44	18.8	0.43
5	0.50	14.0	0.43	14.0	0.43	18.0	0.41
6	0.50	22.0	0.39	22.0	0.39	20.0	0.40
7	0.48	25.0	0.36	18.7	0.39	22.9	0.37
AVG.	0.51	20.5	0.40	18.4	0.41	20.9	0.40

270 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.70	17.1	0.58	15.7	0.59	18.5	0.57
2	0.70	18.5	0.57	18.5	0.57	21.4	0.55
3	0.74	20.2	0.59	22.9	0.57	21.6	0.58
4	0.67	19.4	0.54	16.4	0.56	19.4	0.54
5	0.70	15.7	0.59	18.5	0.57	18.5	0.57
6	0.64	21.8	0.50	18.7	0.52	23.4	0.49
7	0.63	23.8	0.48	15.8	0.53	20.6	0.50
AVG.	0.68	19.5	0.55	18.1	0.55	20.5	0.54

315 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.82	18.2	0.67	17.0	0.68	19.5	0.66
2	0.85	17.6	0.70	16.4	0.71	17.6	0.70
3	0.83	22.8	0.64	20.4	0.66	25.3	0.62
4	0.78	20.5	0.62	21.7	0.61	20.5	0.62
5	0.83	16.8	0.69	14.4	0.71	15.6	0.70
6	0.81	18.5	0.66	17.2	0.67	18.5	0.66
7	0.85	21.1	0.67	17.6	0.70	20.0	0.68
AVG.	0.82	19.4	0.66	17.8	0.67	19.5	0.66

360 mg/m^2-d
SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.95	14.7	0.81	14.7	0.81	14.7	0.81
2	0.93	17.2	0.77	16.1	0.78	17.2	0.77
3	0.95	17.8	0.78	14.7	0.81	13.6	0.82
4	0.98	15.3	0.83	13.2	0.85	14.2	0.84
5	1.01	15.8	0.85	16.8	0.84	14.8	0.86
6	1.04	11.5	0.92	9.6	0.94	12.5	0.91
7	1.03	14.5	0.88	9.7	0.93	13.5	0.89
AVG.	0.98	15.2	0.83	13.4	0.85	14.3	0.84

225 mg/m^2-d

SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.58	18.9	0.47	15.5	0.49	18.9	0.47
2	0.53	22.6	0.41	16.9	0.44	18.8	0.43
3	0.58	18.9	0.47	15.5	0.49	20.6	0.46
4	0.53	15.0	0.45	15.0	0.45	13.2	0.46
5	0.58	17.2	0.48	15.5	0.49	20.6	0.46
6	0.55	18.1	0.45	14.5	0.47	16.3	0.46
7	0.50	18.0	0.41	12.0	0.44	16.0	0.42
AVG.	0.55	18.4	0.44	15.06	0.467143	17.8	0.45

270 mg/m^2-d

SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.72	18.0	0.59	15.2	0.61	18.0	0.59
2	0.73	19.1	0.59	16.4	0.61	17.8	0.60
3	0.71	18.3	0.58	18.3	0.58	18.3	0.58
4	0.65	18.4	0.53	18.4	0.53	21.5	0.51
5	0.68	17.6	0.56	16.1	0.57	19.1	0.55
6	0.67	19.4	0.54	20.8	0.53	23.8	0.51
7	0.65	18.4	0.53	18.4	0.53	24.6	0.49
AVG.	0.68	18.5	0.56	17.7	0.56	20.4	0.54

315 mg/m^2-d

SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.81	19.7	0.65	18.5	0.66	20.9	0.64
2	0.87	19.5	0.70	17.2	0.72	19.5	0.7
3	0.81	16.0	0.68	14.8	0.69	14.8	0.69
4	0.81	18.5	0.66	13.5	0.70	18.5	0.66
5	0.85	14.1	0.73	12.9	0.74	12.9	0.74
6	0.83	14.4	0.71	13.2	0.72	15.6	0.70
7	0.82	18.2	0.67	17.0	0.68	19.5	0.66
AVG.	0.82	17.2	0.68	15.3	0.70	17.4	0.68

360 mg/m^2-d

SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT3
1	0.93	13.9	0.80	17.2	0.77	12.9	0.81
2	0.93	16.1	0.78	16.1	0.78	19.3	0.75
3	0.96	14.5	0.82	13.5	0.83	14.5	0.82
4	0.97	13.4	0.84	12.3	0.85	10.3	0.87
5	1.02	14.0	0.87	12.7	0.89	13.7	0.88
6	1.06	14.1	0.91	12.2	0.93	15.0	0.90
7	1.08	13.8	0.93	12.0	0.95	12.9	0.94
AVG.	0.99	14.3	0.85	13.6	0.85	14.1	0.85

Table A4. NO_2^--N removal performance experiments row data; Part 1 (180 mg/m^2-d TAN applied at various flow rates).

6 Lpm

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.21	19.0	0.17	9.5	0.19	47.6	0.11
2	0.16	50.0	0.08	37.5	0.10	56.2	0.07
3	0.24	25.0	0.18	45.8	0.13	41.6	0.14
4	0.15	40.0	0.09	0.0	0.15	46.6	0.08
5	0.13	0.0	0.13	-7.6	0.14	30.7	0.09
6	0.18	0.0	0.18	-11.1	0.20	33.3	0.12
7	0.20	25.0	0.15	30.0	0.14	10.0	0.18
AVG.	0.18	22.7	0.14	14.8	0.15	38.0	0.11

8 Lpm

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.23	30.4	0.16	39.1	0.14	60.8	0.09
2	0.15	6.6	0.14	-13.3	0.17	26.6	0.11
3	0.13	-7.7	0.14	-7.6	0.14	30.7	0.09
4	0.20	15.0	0.17	-5.0	0.21	15.0	0.17
5	0.20	60.0	0.08	50.0	0.10	5.0	0.19
6	0.15	13.3	0.13	26.6	0.11	-6.6	0.16
7	0.16	12.5	0.14	-6.2	0.17	25.0	0.12
AVG.	0.17	18.6	0.13	11.9	0.14	22.3	0.13

10 Lpm

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.15	13.3	0.13	6.6	0.14	-13.3	0.17
2	0.16	18. 5	0.13	6.2	0.15	12.5	0.14
3	0.18	16.6	0.15	5.5	0.17	5.5	0.17
4	0.13	23.0	0.10	7.7	0.12	-30.7	0.17
5	0.21	-38.0	0.29	0.0	0.21	4.7	0.2
6	0.12	25.0	0.09	16.6	0.10	25.0	0.09
7	0.14	35.7	0.09	14.2	0.12	-14.3	0.16
AVG.	0.15	13.5	0.14	8.1	0.14	-1.5	0.15

12 Lpm

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.19	15.7	0.16	5.2	0.18	5.2	0.18
2	0.14	21.4	0.11	0.0	0.14	14.2	0.12
3	0.19	10.5	0.17	0.0	0.19	10.5	0.17
4	0.19	-31.5	0.25	-21	0.23	5.2	0.18
5	0.17	17.6	0.14	5.8	0.16	17.6	0.14

6	0.19	15.7	0.16	10.5	0.17	21.0	0.15
7	0.18	16.6	0.15	-27.7	0.23	-27.7	0.23
AVG.	0.17	9.4	0.16	-3.8	0.18	6.6	0.16

6 Lpm

SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.19	15.7	0.16	15.7	0.16	47.3	0.10
2	0.20	15.0	0.17	10.0	0.18	45.0	0.11
3	0.20	10.0	0.18	10.0	0.18	30.0	0.14
4	0.16	37.5	0.10	-12.5	0.18	37.5	0.10
5	0.17	29.4	0.12	11.7	0.15	17.6	0.14
6	0.20	30.0	0.14	-40.0	0.28	40.0	0.12
7	0.19	5.2	0.18	-10.5	0.21	36.8	0.12
AVG.	0.18	20.4	0.15	-2.2	0.19	36.3	0.12

8 Lpm

SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.16	25.0	0.12	6.25	0.15	31.2	0.11
2	0.22	22.7	0.17	-13.6	0.25	9.0	0.20
3	0.17	29.4	0.12	-11.7	0.19	5.8	0.16
4	0.21	28.5	0.15	-23.8	0.26	28.5	0.15
5	0.12	33.3	0.08	-16.6	0.14	-50.0	0.18
6	0.15	26.6	0.11	-13.3	0.17	20.0	0.12
7	0.20	25.0	0.15	-15.0	0.23	30.0	0.14
AVG.	0.17	27.2	0.13	-12.5	0.19	10.6	0.15

10 Lpm

SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.24	20.8	0.19	-4.7	0.25	0.0	0.24
2	0.19	-21.0	0.23	0.0	0.19	5.2	0.18
3	0.27	-14.8	0.31	-18.5	0.32	0.0	0.27
4	0.14	35.7	0.09	-21.4	0.17	-7.1	0.15
5	0.29	20.6	0.23	0.0	0.29	-10.3	0.32
6	0.27	25.9	0.20	25.9	0.20	7.4	0.25
7	0.22	27.2	0.16	18.1	0.18	22.7	0.17
AVG.	0.23	13.5	0.20	-0.1	0.23	2.5	0.22

12 Lpm

SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.25	16.0	0.21	0.0	0.25	4.0	0.24
2	0.18	11.1	0.16	0.0	0.18	5.5	0.17
3	0.23	21.7	0.18	8.6	0.21	17.4	0.19
4	0.22	-18.1	0.26	4.5	0.21	9.0	0.2
5	0.18	16.6	0.15	0.0	0.18	11.1	0.16
6	0.19	-15.7	0.22	-10.5	0.21	0.0	0.19

7	0.18	16.6	0.15	-38.8	0.25	-27.7	0.23
AVG.	0.20	6.8	0.19	-5.1	0.21	2.7	0.20

6 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.22	36.3	0.14	22.7	0.17	54.5	0.10
2	0.18	33.3	0.12	22.2	0.14	33.3	0.12
3	0.16	31.2	0.11	25.0	0.12	43.75	0.09
4	0.16	43.7	0.09	25.0	0.12	56.25	0.07
5	0.15	6.6	0.14	0.0	0.15	33.3	0.10
6	0.20	10.0	0.18	-10.0	0.22	35.0	0.13
7	0.18	0.0	0.18	-5.5	0.19	44.4	0.10
AVG.	0.18	23.0	0.13	11.3	0.16	42.9	0.10

8 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.18	22.2	0.14	0.0	0.18	33.3	0.12
2	0.17	11.7	0.15	-41.1	0.24	41.1	0.10
3	0.15	26.6	0.11	-13.3	0.17	26.6	0.11
4	0.17	17.6	0.14	-5.8	0.18	23.5	0.13
5	0.13	7.7	0.12	-46.1	0.19	-38.4	0.18
6	0.20	20.0	0.16	0.0	0.20	10.0	0.18
7	0.18	0.0	0.18	0.0	0.18	-27.7	0.23
AVG.	0.17	15.1	0.14	-15.2	0.19	98.0	0.15

10 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.20	15.0	0.17	-30.0	0.26	5.0	0.19
2	0.17	0.0	0.17	-11.7	0.19	0.0	0.17
3	0.37	45.9	0.20	16.2	0.31	16.2	0.31
4	0.16	25.0	0.12	-6.25	0.17	6.25	0.15
5	0.24	12.5	0.21	4.1	0.23	0.0	0.24
6	0.19	26.3	0.14	5.2	0.18	10.5	0.17
7	0.20	15.0	0.17	0.0	0.20	5.0	0.19
AVG.	0.22	19.9	0.17	-3.2	0.22	6.1	0.20

12 Lpm
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.20	15.0	0.17	0.0	0.2	0.0	0.20
2	0.17	5.8	0.16	11.7	0.15	17.6	0.14
3	0.18	22.2	0.14	5.5	0.17	11.1	0.16
4	0.27	40.7	0.16	37.0	0.17	44.4	0.15
5	0.16	0.0	0.16	-37.5	0.22	12.5	0.14
6	0.13	0.0	0.13	-15.4	0.15	0.0	0.13

7	0.14	14.2	0.12	7.1	0.13	-35.7	0.19
AVG.	0.17	14.0	0.14	1.2	0.17	7.1	0.15

Table A5. NO_2^--N removal performance experiment row data; Part 2 (12 Lpm applied at various TAN concentrations applied).

225 mg/m^2-d

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.22	13.6	0.19	4.5	0.21	9.0	0.2
2	0.21	-9.5	0.23	-14.3	0.24	-4.7	0.22
3	0.26	3.8	0.25	3.8	0.25	7.7	0.24
4	0.25	-20.0	0.30	0.0	0.25	4.0	0.24
5	0.28	7.1	0.26	-7.1	0.30	3.5	0.27
6	0.26	3.8	0.25	-7.7	0.28	3.8	0.25
7	0.21	-4.7	0.22	-4.7	0.22	0.0	0.21
AVG.	0.24	-0.8	0.24	-3.6	0.25	3.3	0.23

270 mg/m^2-d

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.28	10.7	0.25	14.3	0.24	0.0	0.28
2	0.25	4.0	0.24	0.0	0.25	16.0	0.21
3	0.27	18.5	0.22	11.1	0.24	11.1	0.24
4	0.31	6.4	0.29	3.2	0.30	16.1	0.26
5	0.29	-3.4	0.30	-6.9	0.31	0.0	0.29
6	0.29	13.8	0.25	10.3	0.26	13.8	0.25
7	0.28	7.1	0.26	3.5	0.27	7.1	0.26
AVG.	0.28	8.1	0.25	5.0	0.26	9.1	0.25

315 mg/m^2-d

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.29	10.3	0.26	3.4	0.28	10.3	0.26
2	0.20	5.0	0.19	0.0	0.20	10.0	0.18
3	0.29	10.3	0.26	3.4	0.28	6.9	0.27
4	0.28	7.1	0.26	14.2	0.24	7.1	0.26
5	0.24	12.5	0.21	8.3	0.22	8.3	0.22
6	0.26	7.7	0.24	3.8	0.25	7.7	0.24
7	0.20	15.0	0.17	10.0	0.18	15.0	0.17
AVG.	0.25	9.7	0.22	6.2	0.23	9.3	0.23

360 mg/m^2-d

SYSTEM 1

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.28	7.1	0.26	10.7	0.25	3.5	0.27
2	0.30	6.6	0.28	3.3	0.29	0.0	0.30
3	0.36	8.3	0.33	5.5	0.34	11.1	0.32
4	0.37	10.8	0.33	8.1	0.34	10.8	0.33

5	0.35	11.4	0.31	8.5	0.32	11.4	0.31
6	0.32	12.5	0.28	9.3	0.29	9.3	0.29
7	0.31	6.4	0.29	6.4	0.29	9.6	0.28
AVG.	0.32	9.0	0.29	7.4	0.30	7.9	0.3

225 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.39	-5.1	0.41	25.6	0.29	53.8	0.18
2	0.26	19.2	0.21	23.0	0.20	26.9	0.19
3	0.30	10.0	0.27	3.3	0.29	10.0	0.27
4	0.23	8.7	0.21	-4.3	0.24	4.3	0.22
5	0.19	15.7	0.16	26.3	0.14	10.5	0.17
6	0.24	-4.1	0.25	0.0	0.24	4.1	0.23
7	0.20	5.0	0.19	-5.0	0.21	5.0	0.19
AVG.	0.26	7.0	0.24	9.8	0.23	16.4	0.20

270 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.21	0.0	0.21	9.5	0.19	9.5	0.19
2	0.31	3.2	0.3	3.2	0.30	22.5	0.24
3	0.31	22.5	0.24	12.9	0.27	25.8	0.23
4	0.26	7.6	0.24	3.8	0.25	19.2	0.21
5	0.35	14.2	0.3	5.7	0.33	5.7	0.33
6	0.36	5.5	0.34	2.7	0.35	8.3	0.33
7	0.35	5.7	0.33	0.0	0.35	2.8	0.34
AVG.	0.30	8.4	0.28	5.4	0.29	13.4	0.26

315 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.25	8.0	0.23	8.0	0.23	12.0	0.22
2	0.24	12.5	0.21	4.1	0.23	8.3	0.22
3	0.30	10.0	0.27	0.0	0.30	10.0	0.27
4	0.27	3.7	0.26	0.0	0.27	7.4	0.25
5	0.26	3.8	0.25	7.6	0.24	7.6	0.24
6	0.23	0.0	0.23	4.3	0.22	4.3	0.22
7	0.27	11.1	0.24	7.4	0.25	7.4	0.25
AVG.	0.26	7.0	0.24	4.5	0.24	8.1	0.23

360 mg/m^2-d
SYSTEM 2

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.27	11.1	0.24	3.7	0.26	7.4	0.25
2	0.26	3.8	0.25	3.8	0.25	3.8	0.25
3	0.22	9.0	0.20	9.0	0.20	13.6	0.19
4	0.32	15.6	0.27	6.2	0.30	12.5	0.28

5	0.30	13.3	0.26	6.6	0.28	10.0	0.27
6	0.28	14.2	0.24	10.7	0.25	14.2	0.24
7	0.29	10.3	0.26	10.3	0.26	13.7	0.25
AVG.	0.27	11.0	0.24	7.2	0.25	10.8	0.24

225 mg/m^2-d
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.21	28.5	0.15	-33.3	0.28	28.5	0.15
2	0.17	11.7	0.15	11.7	0.15	0.0	0.17
3	0.18	22.2	0.14	11.1	0.16	22.2	0.14
4	0.17	11.7	0.15	0.0	0.17	5.8	0.16
5	0.23	26.0	0.17	17.3	0.19	8.6	0.21
6	0.19	-10.5	0.21	-5.2	0.20	5.2	0.18
7	0.18	-27.7	0.23	16.6	0.15	-16.6	0.21
AVG.	0.19	8.8	0.17	2.6	0.18	7.7	0.17

270 mg/m^2-d
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.26	7.6	0.24	23.0	0.20	-7.6	0.28
2	0.28	10.7	0.25	3.5	0.27	7.1	0.26
3	0.29	3.4	0.28	-3.4	0.30	6.8	0.27
4	0.27	3.7	0.26	7.4	0.25	18.5	0.22
5	0.22	13.6	0.19	9.0	0.20	9.0	0.2
6	0.37	5.4	0.35	10.8	0.33	13.5	0.32
7	0.32	6.25	0.30	6.2	0.30	0.0	0.32
AVG.	0.28	7.2	0.26	8.1	0.26	6.7	0.26

315 mg/m^2-d
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.24	8.3	0.22	8.3	0.22	0.0	0.24
2	0.21	4.7	0.20	0.0	0.21	14.2	0.18
3	0.36	8.3	0.33	19.4	0.29	25.0	0.27
4	0.29	17.2	0.24	13.7	0.25	20.6	0.23
5	0.26	19.0	0.21	11.5	0.23	15.3	0.22
6	0.24	12.5	0.21	8.3	0.22	12.5	0.21
7	0.19	15.7	0.16	5.2	0.18	10.5	0.17
AVG.	0.24	12.3	0.22	9.5	0.23	14.0	0.21

360 mg/m^2-d
SYSTEM 3

DAY	IN	% REMOV.	OUT 1	% REMOV.	OUT 2	% REMOV.	OUT 3
1	0.26	15.3	0.22	3.8	0.25	3.8	0.25
2	0.25	-4.0	0.26	0.0	0.25	8.0	0.23
3	0.26	7.6	0.24	3.8	0.25	11.5	0.23
4	0.28	7.1	0.26	7.1	0.26	10.7	0.25
5	0.29	3.4	0.28	0.0	0.29	6.8	0.27

6	0.28	10.7	0.25	7.1	0.26	14.2	0.24
7	0.25	8.0	0.23	0.0	0.25	4.0	0.24
AVG.	0.26	6.9	0.24	3.1	0.25	8.4	0.24

Table A6. NO_3^--N raw rata. Part 1: Values recorded at 180 mg/m^2-d TAN and various flow rates. Part 2: Values recorded at 12 Lpm flow rate and various TAN loadings.

		PART 1			PART 2	
		6 Lpm			225 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	82	98	77	135	128	135
2	91	92	82	135	138	153
3	87	131	98	111	135	134
4	103	122	93	129	156	144
5	107	133	114	146	137	139
6	106	101	81	158	153	160
7	116	99	112	122	147	158
AVG.	98.8	110.8	93.8	133.7	142	146.1
		8 Lpm			270 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	124	114	90	142	175	164
2	136	114	103	126	138	145
3	112	98	94	110	140	146
4	92	120	102	146	173	168
5	95	114	109	152	136	152
6	108	110	101	168	144	163
7	114	107	124	155	148	154
AVG.	111.5	111	103.2	142.7	150.5	156
		10 Lpm			315 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	129	141	161	181	202	195
2	130	170	159	172	181	175
3	154	167	160	210	205	195
4	115	154	172	174	168	190
5	147	162	158	182	175	192
6	127	159	151	224	213	234
7	106	138	135	199	196	205
AVG.	129.7	155.8	156.5	191.7	191.4	198
		12 Lpm			360 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	148	165	179	250	246	255
2	136	159	174	236	233	264
3	148	146	169	196	220	230
4	140	164	167	236	259	251

5	140	139	148	242	268	239
6	158	160	159	230	247	241
7	136	168	145	208	212	220
AVG.	143.7	157.2	163	220.2	236.7	238.8

Table A7. D.O. Raw Data. Part 1: Values recorded at 180 mg/m^2-d TAN applied and various flow rates. Part 2: Values recorded at 12 Lpm flow rate and various TAN loadings.

	PART 1 (180 mg/m2-d)			PART 2 (12 Lpm)		
	6 Lpm			225 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	6.40	6.39	6.40	6.94	6.82	6.93
2	7.31	7.30	7.27	7.04	7.01	7.07
3	7.39	7.32	7.29	7.02	7.02	7.14
4	6.93	6.91	6.92	7.15	7.07	7.11
5	7.30	7.30	7.31	6.97	6.98	6.98
6	6.60	6.57	6.63	7.44	7.37	7.35
7	6.78	6.66	6.67	7.26	7.23	7.24
AVG.	6.95	6.92	6.92	7.11	7.07	7.11
	8 Lpm			270 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	7.00	7.05	7.08	7.00	7.04	7.02
2	6.78	6.90	6.95	7.61	7.51	7.48
3	6.94	6.94	6.95	7.61	7.50	7.57
4	6.57	6.59	6.62	7.24	7.16	7.15
5	6.90	6.88	6.78	7.45	7.26	7.44
6	6.70	6.71	6.64	7.45	7.42	7.44
7	6.58	6.75	6.71	7.41	7.22	7.38
AVG.	6.78	6.83	6.81	7.39	7.30	7.35
	10 Lpm			315 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	6.77	6.71	6.75	7.48	7.40	7.42
2	7.28	7.24	7.21	7.44	7.40	7.40
3	6.97	6.96	6.98	7.46	7.20	7.22
4	7.45	7.40	7.44	7.60	7.43	7.45
5	7.30	7.22	7.26	7.48	7.42	7.44
6	7.24	7.26	7.28	7.18	7.05	7.06
7	7.24	7.20	7.25	7.25	7.15	7.19
AVG.	7.17	7.14	7.16	7.41	7.29	7.31
	12 Lpm			360 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	7.30	7.27	7.26	7.20	7.12	7.16
2	7.03	7.02	7.05	7.41	7.32	7.32
3	7.05	7.03	7.06	7.20	7.15	7.14
4	6.84	6.81	6.88	7.28	7.16	7.23
5	6.97	6.96	7.01	7.55	7.41	7.44
6	6.94	6.91	7.00	7.18	7.04	7.10

7	7.00	7.02	7.10	7.28	7.17	7.18
AVG.	7.02	7.00	7.05	7.30	7.19	7.22

Table A8. pH raw data. Part 1: Values recorded at 180 mg/m^2-d TAN applied and various flow rates; Part 2: Values recorded at 12 Lpm flow rate and various TAN loadings.

	PART 1 (180 mg/m2-d)			PART 2 (12 Lpm)		
	6 Lpm			225 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	7.87	7.82	7.97	8.05	8.03	8.07
2	7.75	7.60	7.87	8.03	8.07	8.08
3	7.68	7.55	7.58	7.92	7.90	7.95
4	7.87	7.75	7.75	8.05	8.07	8.11
5	7.90	7.75	7.80	8.00	8.08	8.10
6	7.90	7.75	7.75	7.84	7.89	7.83
7	7.75	7.58	7.54	8.00	7.92	7.94
AVG.	7.81	7.68	7.75	7.98	7.99	8.01
	8 Lpm			270 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	7.75	7.95	7.95	8.00	7.95	7.97
2	8.12	8.04	8.10	7.92	7.83	7.73
3	8.34	8.18	8.32	7.85	7.79	7.87
4	8.29	8.30	8.22	7.92	7.85	7.79
5	7.87	7.85	7.87	7.75	7.72	7.73
6	7.90	7.90	7.86	7.82	7.80	7.81
7	7.62	7.51	7.55	8.00	7.90	7.90
AVG.	7.98	7.96	7.98	7.89	7.83	7.82
	10 Lpm			315 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	7.97	7.84	7.96	7.77	7.80	7.72
2	7.98	7.88	7.96	7.44	7.40	7.40
3	7.99	7.89	7.92	7.81	7.72	7.75
4	8.14	8.04	8.01	8.11	7.81	7.93
5	7.90	7.84	7.97	7.87	7.88	7.89
6	7.87	7.90	7.92	8.19	8.15	8.10
7	7.80	7.77	7.70	8.20	8.24	8.19
AVG.	7.95	7.88	7.92	7.91	7.85	7.85
	12 Lpm			360 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	7.91	7.86	7.96	7.91	7.89	7.77
2	7.89	7.90	7.90	8.12	8.10	7.98
3	7.95	7.99	8.00	8.17	8.18	8.15
4	7.94	7.92	7.92	8.10	7.97	8.05
5	8.20	8.22	8.20	7.85	7.76	7.69
6	8.25	8.16	8.18	8.11	8.17	8.20

7	8.18	8.12	8.16	7.95	7.97	8.00
AVG.	8.04	8.02	8.04	8.03	8.00	7.97

Table A9. Temperature (°C) raw data. Part 1: Values recorded at 180 mg/m^2-d TAN applied and various flow rates; Part 2: Values recorded at 12 Lpm flow rate and various TAN loadings.

	PART 1 (180 mg/m2-d)			PART 2 (12 Lpm)		
	6 Lpm			215 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	26.9	27.0	27.1	30.5	30.7	30.3
2	26.0	26.0	26.1	30.5	30.6	30.2
3	25.1	25.2	25.2	30.4	30.6	30.2
4	28.2	28.1	28.0	30.1	30.2	29.8
5	28.2	28.2	28.1	29.1	29.2	28.9
6	29.5	29.5	29.1	27.9	28.0	28.0
7	29.8	29.8	29.6	29.2	29.4	29.1
AVG.	27.6	27.6	27.6	29.6	29.8	29.5
	8 Lpm			270 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	29.0	28.9	28.7	29.2	29.4	29.0
2	28.7	28.7	28.4	27.8	28.0	27.9
3	27.8	27.8	27.6	25.6	25.8	25.7
4	27.7	27.7	27.6	29.3	29.6	29.4
5	28.4	28.4	28.2	25.8	26.3	26.1
6	28.5	28.6	28.4	26.8	26.6	27.7
7	28.5	28.4	28.6	26.5	26.8	26.7
AVG.	28.3	28.3	28.2	27.2	27.5	27.5
	10 Lpm			315 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	28.0	28.1	28.0	26.1	26.5	26.3
2	27.4	27.4	27.2	27.4	27.8	27.5
3	27.9	28.0	27.8	28.1	28.8	28.6
4	27.5	27.6	27.4	27.7	28.3	28.1
5	27.6	27.6	27.4	27.2	27.6	27.3
6	28.8	27.8	27.7	28.5	28.9	28.8
7	28.0	28.1	27.8	28.4	28.8	28.7
AVG.	27.8	27.8	27.6	27.6	28.1	27.9
	12 Lpm			360 mg/m^2-d		
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	28.9	29.0	28.9	28.4	28.8	28.8
2	29.7	29.8	29.6	28.8	29.2	29.0
3	29.5	29.6	29.4	28.4	28.9	28.7
4	30.3	30.4	30.2	28.2	28.7	28.5
5	29.8	29.9	29.5	27.1	27.6	27.4
6	29.9	30.9	29.6	29.2	29.6	29.4

7	30.1	30.2	30.0	27.8	28.3	28.1
AVG.	29.7	29.9	29.6	28.3	28.7	28.5

Table A10. Alkalinity raw data. Part 1: Values recorded at 180 mg/m^2-d TAN applied and various flow rates; Part 2: Values recorded at 12 Lpm flow rate and various TAN loadings.

		PART 1			PART 2	
		6 Lpm			225 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	105	95	100	110	112	115
2	110	115	105	105	105	110
3	110	105	96	105	105	110
4	108	105	105	95	100	105
5	140	125	125	105	110	115
6	105	105	110	118	125	112
7	140	140	135	115	130	115
AVG.	116.8	112.8	110.8	107.5	112.4	111.7
		8 Lpm			270 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	115	120	110	125	115	115
2	95	110	95	110	110	95
3	135	135	140	105	105	110
4	125	110	133	120	115	100
5	105	108	110	107	105	108
6	115	110	115	115	95	100
7	95	110	100	130	120	120
AVG.	112.1	114.7	114.7	116	109.3	106.8
		10 Lpm			315 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	130	120	130	105	115	105
2	130	120	125	105	115	115
3	105	115	105	105	105	105
4	120	115	120	105	115	115
5	128	105	120	120	125	125
6	105	110	105	120	120	120
7	95	95	90	140	150	145
AVG.	116.1	111.4	113.5	114.2	120.7	118.5
		12 Lpm			360 mg/m^2-d	
DAY	System 1	System 2	System 3	System 1	System 2	System 3
1	125	115	125	120	125	115
2	130	135	130	120	120	115
3	130	130	125	140	145	145
4	130	125	135	145	140	140
5	120	130	120	115	100	115
6	135	125	125	140	145	145

7	120	115	118	115	115	120
AVG.	127.1	125	125.4	127.8	127.1	127.8

Table A11. Biofilm weight values (g) raw data. Part 1: Values recorded at 180 mg/m^2-d TAN applied and various flow rates; Part 2: Values recorded at 12 Lpm flow rate and various TAN loadings.

	PART 1(180 mg/m^2-d)				PART 2 (12 Lpm)			
	6 Lpm	8 Lpm	10 Lpm	12 Lpm	225 mg/m^2-d	270 mg/m^2-d	315 mg/m^2-d	360 mg/m^2-d
Rep. 11	0.0072	0.0076	0.0065	0.0046	0.0056	0.0067	0.0066	0.0082
	0.0075	0.0073	0.0056	0.0056	0.0057	0.0063	0.0069	0.0074
Average	0.0074	0.0075	0.0061	0.0051	0.0057	0.0065	0.0068	0.0078
Rep. 12	0.0091	0.0060	0.0047	0.0061	0.0067	0.0074	0.0065	0.0112
	0.0077	0.0059	0.0056	0.0049	0.0057	0.0062	0.0073	0.0102
Average	0.0084	0.0060	0.0052	0.0055	0.0062	0.0066	0.0069	0.0107
Rep. 13	0.0091	0.0079	0.0066	0.0060	0.0064	0.0070	0.0074	0.0140
	0.0095	0.0063	0.0065	0.0071	0.0061	0.0068	0.0068	0.0112
Average	0.0093	0.0071	0.0066	0.0066	0.0063	0.0069	0.0071	0.0126
Rep. 21	0.0096	0.0045	0.0057	0.0062	0.0058	0.0063	0.0064	0.0710
	0.0093	0.0061	0.0061	0.0058	0.0063	0.0062	0.0063	0.0084
Average	0.0095	0.0053	0.0059	0.0060	0.0061	0.0063	0.0064	0.0078
Rep. 22	0.0098	0.0064	0.0050	0.0053	0.0066	0.0067	0.0075	0.0093
	0.0103	0.0060	0.0057	0.0061	0.0068	0.0071	0.0066	0.0085
Average	0.0101	0.0062	0.0054	0.0057	0.0067	0.0069	0.0071	0.0089
Rep. 23	0.0089	0.0066	0.0063	0.0087	0.0076	0.0076	0.0077	0.0098
	0.0095	0.0097	0.0069	0.0063	0.0079	0.0077	0.0075	0.0092
Average	0.0092	0.0082	0.0066	0.0075	0.0076	0.0077	0.0076	0.0095
Rep. 31	0.0101	0.0066	0.0059	0.0051	0.0063	0.0068	0.0079	0.0096
	0.0105	0.0070	0.0062	0.0057	0.0067	0.0065	0.0073	0.0094
Average	0.0103	0.0068	0.0061	0.0054	0.0065	0.0067	0.0076	0.0095
Rep. 32	0.0099	0.0074	0.0084	0.0056	0.0067	0.0073	0.0082	0.0105
	0.0083	0.0100	0.0071	0.0068	0.0067	0.0066	0.0077	0.0094
Average	0.0091	0.0087	0.0078	0.0062	0.0067	0.0070	0.0080	0.0100
Rep. 33	0.0098	0.0081	0.0064	0.0076	0.0074	0.0074	0.0081	0.0094
	0.0100	0.0102	0.0100	0.0054	0.0073	0.0081	0.0077	0.0110
Average	0.0099	0.0092	0.0082	0.0065	0.0074	0.0076	0.0079	0.0103
Avg. Filter 1	0.0091	0.0064	0.0058	0.0055	0.0061	0.0065	0.0069	0.0084
Avg. Filter 2	0.0092	0.0069	0.006	0.0058	0.0065	0.0069	0.0073	0.0099
Avg. Filter 3	0.0095	0.0082	0.0071	0.0068	0.0071	0.0074	0.0075	0.1130

Figure A12. Biofilm thickness calculated (μm).

	PART 1(180 mg/m^2-d)				PART 2 (12 Lpm)			
	6 Lpm	8 Lpm	10 Lpm	12 Lpm	225 mg/m^2-d	270 mg/m^2-d	315 mg/m^2-d	360 mg/m^2-d
Filter 1	73.2	52.3	46.6	44.2	49.1	52.3	55.5	67.6

Filter 2	74.0	55.5	48.3	46.6	52.3	55.5	58.7	79.7
Filter 3	76.4	66.0	57.1	54.7	57.1	59.5	60.3	90.1

Table A 13. Bed expanding level (cm) at various flow rates (Lpm) tested in the study.

Flow rate, Lpm	0	6	8	10	12
System 1					
Filter D1	79.0	110.5	122.5	137.0	155.5
Filter D2	55.1	72.5	79.0	86.5	94.5
Filter D3	40.2	47.5	52.5	55.5	57.5
System 2					
Filter D1	79.0	109.0	12.0	136.5	157.0
Filter D2	55.1	71.0	78.0	85.5	93.5
Filter D3	40.2	48.5	51.5	55.0	57.0
System 3					
Filter D1	79.0	109.0	123.5	138.5	154.0
Filter D2	55.1	71.5	80.0	87.5	94.5
Filter D3	40.2	47.5	51.5	55.5	57.5

Table A 14. Average bed expanding level (cm) at various flow rates tested in the study.

Flow rate, Lpm	0	6	8	10	12
Filter D1	79.0	109.6	122.3	137.3	155.0
Filter D2	55.1	71.6	79.0	86.5	94.0
Filter D3	40.2	47.8	51.8	55.3	57.3

Table A 15. Fluidization levels (cm) at various water velocities (cm) tested in the study.

Flow rate, Lpm	0	6	8	10	12
Fluidiz. level D1,cm	79.0	109.6	122.3	137.3	155
Water velocity,cm	0.0	0.4	0.53	0.67	0.8
Fluidiz. level D2, cm	55.1	71.6	79.0	86.5	94.0
Water velocity, cm	0.0	0.6	0.8	1.0	1.2
Fluidiz. level D3, cm	40.2	47.8	51.8	55.3	57.3
Water velocity, cm	0.0	0.8	1.0	1.3	1.6

Table A 16. Volumes of expanded bed (L) at different velocities (cm) tested in the study.

Flow rate, Lpm	0	6	8	10	12
Water velocity,cm	0.00	0.40	0.53	0.67	0.80
Vol. expansion D1	10.00	13.87	15.48	17.38	19.62
Water velocity, cm	0.00	0.60	0.80	1.00	1.20
Vol. expansion D2	10.00	12.98	14.32	15.68	17.04
Water velocity, cm	0.00	0.80	1.06	1.33	1.60
Vol. expansion D3	10.00	11.88	12.88	13.74	14.25

VITA

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