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Review

Biofilm Fixed Film Systems

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Abstract: The work reviewed here was published between 2008 and 2010 and describes research that involved aerobic and anoxic biofilm treatment of water pollutants. Biofilm denitrification systems are covered when appropriate. References catalogued here are divided on the basis of fundamental research area or reactor types. Fundamental research into biofilms is presented in two sections, Biofilm Measurement and Characterization and Growth and Modeling. The reactor types covered are: trickling filters, rotating biological contactors, fluidized bed bioreactors, submerged bed biofilm reactors, biological granular activated carbon, membrane bioreactors, and immobilized cell reactors. Innovative reactors, not easily classified, are then presented, followed by a section on biofilms on sand, soil and sediment.

Keywords: biofilm; wastewater treatment systems; fixed film models; trickling filters; biotowers; rotating biological contactors; biomembrane processes; submerged fixed film; xenobiotics; nutrient removal; nitrification; denitrification; biological phosphorus removal; extracellular polymeric substances

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1. Introduction

The scope of research in the area of biofilm fixed film systems continues to expand beyond the traditional trickling filters, biotowers, and rotating biological contactors (RBCs) into biofilm measurement and characterization methods, growth and modeling, new biofilm growth media, innovative bioreactors (including various membrane bioreactors and hybrid reactors), fixed-film xenobiotics removal, bioelectricy generation, and the roles of biofilms to remove nutrients and recalcitrant contaminants in the natural environment. Biofilm fixed film systems will continue to have relevance in the treatment of wastewater as technological advances, such as membrane bioreactors and their hybrids, evolve. Natural biofilm attenuation, accumulation and destruction of nutrients, pharmaceuticals and personal care products (PPCPs), and recalcitrant contaminants may open up new applications of biofilm systems.

2. Biofilm Measurement and Characterization

2.1. Sensors and Microsensors

Downing and Nerenberg [1,2] used microsensors to measure nitrogen forms produced by biofilms on aerated submerged membranes. They also used fluorescence *in situ* hybridization (FISH) tests on biofilm to reveal three distinct biofilm regions: ammonia-oxidizing bacteria (AOB) and nitrite oxidizing bacteria (NOB) near the membrane, strictly AOB at intermediate biofilm depth and AOB and heterotrophs near the outer biofilm/bulk liquid interface. McLamore *et al.* [3] used noninvasive, microsensor techniques to quantify real time changes in oxygen and proton flux for Nitrosomonas europaea and Pseudomonas aeruginosa biofilms following exposure to environmental toxins in membrane-aerated bioreactors.

2.2. Biofilm Attachment and Detachment

Biofilm formation and adherence properties of 13 bacterial strains commonly found in wastewater treatment systems were studied by Andersson *et al.* [4]. Four different culture media were used and it was found that the medium composition strongly affected biofilm formation. Adherence properties of pure and multi-strain biofilms were assessed. Strongest biofilm formation was observed when mixtures of all 13 bacteria were grown together. Bacterial biofilm development in tertiary treatment processes was characterized by molecular biological methods by Shoji *et al.* [5]. Low assimilable organic carbon hindered heterotrophic bacteria and favored autotrophs and oligotrophs. Ammonia load affected the two dominant Nitrospirae-related (nitrite oxidizing) and Acidobacteria-related (oligotrophic) bacteria species and their ratio in biofilm more than other operational conditions. Roeselers *et al.* [6] reported that a matrix of substances secreted by phototrophs and heterotrophs enhances the attachment of biofilm community.

Jechalke, *et al.* [7] studied biofilm development on coconut fibers and polypropylene textiles for enhancing biodegradation of low-concentration methyl tert-butyl ether (MTBE), benzene, and ammonium from groundwater in aerated treatment ponds. Coconut fibers were more effective biofilm support media than polypropylene textiles for recruitment and development of biofilms for MTBE degradation. Benzene metabolizing bacteria biofilms did not exhibit a preference for one support medium over the other. Confocal laser scanning microscopy (CLSM) and denaturing gradient gel electrophoresis (DGGE) techniques were used to study the microbial community and structure in the biofilm.

2.3. Microscopy

Guzzon *et al.* [8] performed elemental analysis by energy filtering transmission electron microscopy to show subcellular localization of phosphorus and confirm the accumulation in phototrophic microorganisms in biofilms grown in high light conditions.

Tian *et al.* [9] conducted research on integrative membrane coagulation adsorption bioreactors (MCABR) for the purpose of removal of organic matter, including biodegradable dissolved organic carbon (BDOC), assimilable organic carbon (AOC), and disinfection byproducts. Biofilm on the membrane provided additional rejection of dissolved organic matter, and the biofouling of the membrane was observed using scanning electron microscopy (SEM) in conjunction with CLSM. Biofouling of membranes by river waters containing BDOC was also studied by Marconnet *et al*. [10]. Attenuated total reflection Fourier transform infrared (ATR-FTIR) spectroscopy and CLSM were used to determine the composition and organization of the biofilm fouling on the membrane.

2.4. Novel Techniques

Increased popularity of attached-growth wastewater treatment systems (e.g., biological aerated filtration processes-BAF and various hybrids of membrane biological reactors-MBR) has created the need for a rapid and reliable method of characterizing biofilms. Spettmann *et al.* [11] used fluorescently labeled foulants filtered through and deposited on a polyethersulfone ultrafiltration membrane to study the fouling from inorganic salt precipitants, polysaccharides, organic and inorganic particles, and microbial biofilms. CLSM in conjunction with image analysis allowed three-dimensional visualization of the three-dimensional distribution of fluorescently labeled foulants in multi-layered deposits and cleaning or deposit removal efficiency evaluation. Fluorochrome stains, CLSM, and an image analysis program were similarly used by Bjerkey and Fiksdal [12] to study biofilm structure on curved membrane surfaces, such as hollow fiber membranes. Thickness, volume of biomass, porosity, and roughness of biofilms were calculated.

Delatolla *et al.* [13] described a simple, rapid, and reliable technical procedure that enabled biofilm samples attached to polystyrene beads to be characterized in terms of the biofilm mass and nitrogen content and proposed a protocol that demonstrated 99.9% removal of the biofilm from polystyrene beads. The application of molecular techniques to the study of wastewater treatment systems by Wojnowska-Baryla *et al.* [14] suggested that microbial groups may be organized in various spatial structures such as activated sludge flocs, biofilm or granules and represented by single coherent phylogenic groups such as ammonia-oxidizing bacteria (AOB) and polyphosphate-accumulating organisms (PAO). The microbial community structure of biomembrane in biological contact oxidation packing was analyzed by He *et al.* [15] to assess growth of the biomembrane and mechanisms of the water purification process using 16S rDNA and amoA gene based amplification and denaturing gradient gel electrophoresis (PCR-DGGE).

2.5. Extracellular Polymeric Substances (EPS)

During the past decade, biofilm reactors have been successfully applied for production of many value added products, often because of EPS. Advances in biofilm reactors were investigated by Cheng *et al.* [16], and biofilm reactors with novel applications and designs were summarized in a review.

Phototrophic biofilm samples from a wastewater treatment plant were studied by Di Pippo *et al.* [17] in microcosm experiments under varying irradiances, temperatures and flow regimes to assess the effects of environmental variables and phototrophic biomass on capsular exopolysaccharides (CPS). The results suggested that CPS have a stable spatial conformation and a complex monosaccharide composition. They noted the potential of cyanobacteria and diatoms in removal of residual nutrients and noxious cations.

Avella *et al.* [18] examined three paper mill wastewater treatment plant (WWTP) sludge flocs using size exclusion chromatography and CLSM observations and identified that a sludge with good settling characteristics involved an important EPS production in the presence of nitrification and phosphate nutrient. The other two sludges had poor settling properties and the EPS production was weak. Although these sludges were from activated sludge WWTPs, this suggests that EPS production may have importance to a wide range of WWTP processes.

2.6. Metal and Radionucleotide Sorption

Lin *et al.* [19] derived a biodegradation model for anaerobic fixed-biofilm reactor simultaneous removal of phenol with chromium (VI) reduction. The model, based on diffusive mass transfer and double Monod kinetics, was tested against a laboratory column reactor and showed close agreement.

2.7. Ammonia Removal

Nitrification processes have served as an important basis for the development of today's understanding and mathematical models for many wastewater treatment processes (activated sludge, biofilm reactors) and self-purification processes in rivers, in the view of Gujer [20]. Redundancy analysis demonstration of the relevance of the temperature to ammonia oxidizing was investigated by Park *et al.* [21] and temperature was more significant than salt concentration effects on AOB compositions and dynamics.

2.8. Microbial Community Structure

In order to monitor and control engineered microbial structure in wastewater treatment systems, it is necessary to understand the relationships between the microbial community structure and the process performance. The review by Wojnowska-Baryla *et al.* [14] focused on bacterial communities in wastewater treatment processes, the quantity of microorganisms and structure of microbial consortia in wastewater treatment bioreactors. The study by Weber *et al.* [22] on the diversity of fungi in aerobic sewage granules by gene sequence analysis suggested that fungal community composition in granules depended on the wastewater type and the phase of granule development.

Potential of biofilm-based biofuel production was investigated by Wang and Chen [23]. Biofilm advantages include cell-associated hydrolytic enzymes concentration at the biofilm-substrate interface to increase reaction rates, a layered multiple-species microbial structure in which may sequentially convert complex substrates and coferment hexose and pentose as hydrolysates diffuse outward, and the possibility of fungal-bacterial symbioses allowing delignification and saccharification.

Environmental factors shaping the ecological niche of ammonia-oxiding archaea (AOA) were studied by Ergurder *et al.* [24]. They proposed that AOA might be important, even predominant, biological actors within the nitrogen cycle in low nutrient, low pH and sulfide-containing environments. Bacterial biofilm communities in tertiary wastewater treatment processes were characterized by Shoji *et al.* [5]. Clone library analysis showed that Nitrospirae-related (nitrite-oxydizing bacteria) and Acidobacteria-related (probably oligotrophic bacteria) groups were dominant. Santoro *et al.* [25] studied the relative abundance of AOA and beta-AOB across a subterranean estuary environment, subject to salinity, oxygen and dissolved organic nitrogen gradients. The beta-AOB was dramatically lower in fresh water stations compared with the saline stations while AOA abundance remained nearly constant across the study site. These results offered some new insight into the ecology of AOA and beta-AOB by elucidating conditions that may favor the numerical dominance of beta-AOB over AOA in coastal sediments.

Electro-active biofilms (EAB) were studied by Erable *et al.* [26]. EABs, from soils, seawater, freshwater sediments, and sewage components, coated onto electrodes are popular in the field of microbial fuel cell technology, bioremediation, biosynthesis, biosensor design, *etc*. The review on electricity from microorganisms by Debabov [27] discussed the molecular mechanisms of electron transfer to the environment. Ren *et al.* [28] studied the electricity production and microbial biofilm ecology in cellulose-fed microbial fuel cells. In that study, electric generation and the microbial ecology of cellulose-fed MFCs were analyzed using defined co-culture of Clostridium cellulolyticum and Geobacter sulfurreducens.

Effect of oxygen gradient on the activity and microbial community structure of nitrifying, membrane-aerated biofilm (MAB) was investigated by Downing and Nerenberg [1]. Biofilm Nitrobacter were dominant at membrane dissolved oxygen (DO) concentrations of 2 g/m^3 and anoxic bulk liquid, and Nitrospira spp. were dominant at membrane DO concentrations less than 2 g/m^3. The biofilm model predicted the greatest nitrite formation and lowest ammonium oxidation flux when the membrane DO was 2 g/m^3 and bulk liquid was anoxic and the lowest nitrite formation and highest ammonium oxidation flux when membrane DO and bulk liquid DO were 8 g/m^3 and 2 g/m^3, respectively.

You *et al.* [29] studied nitrogen removal by AOA, reporting that AOA thrive in many environments, including wastewater treatment systems, and that AOA may outnumber AOB.

2.9. Intercellular Communication

Influence of microbial interactions and polysaccharide compositions on nutrient removal activity in multi-species biofilms, formed by strains found in wastewater treatment systems, was studied by Andersson *et al.* [30]. In this report, relationship between biofilm formation, denitrification activity, phosphorus removal and the composition of EPS, exopolysaccharides and bacterial community was investigated using biofilm of denitrification and phosphorus removing strains of microbes. Denitrification activity in biofilms increased with the amount of biofilm, while phosphorus removal depended on bacterial growth rate.

2.10. Others

Guzzon *et al.* [8] studied phosphorus removal using cultured phototropic biofilm from wastewater sedimentation, which confirmed phosphorus accumulation in phototropic microorganisms in biofilms grown in high light conditions.

3. Growth and Modeling

Over the last decade new technologies are emerging for wastewater treatment. One of most recent alternatives is the couple of moving bed biofilm reactors (MBBR) and conventional activated sludge process referred to as hybrid MBBR or HMBBR. The paper by Trapani *et al.* [31] presented first results of a respirometric technique study aimed at estimating the kinematic heterotrophic constants in the HMBBR pilot plant.

Wichern *et al.* [32] studied optimization of a sequencing batch reactor (SBR) operation treating dairy wastewater with aerobic granular sludge. The report said that under different operational conditions average nitrification rates up to 5 $gN/(m^3h)$ and denitrification rates up to 3.7 $gN/(m^3h)$ could be achieved.

A model of integrated fixed-film activated sludge (IFAS) and moving-bed biofilm reactor (MBBR) systems was developed by Boltz *et al.* [33]. The model was based on theoretical considerations that include: simultaneous diffusion and Monod-type reaction kinetics inside the biofilm; competition between aerobic autotrophic nitrifiers, non-methanol-degrading facultative heterotrophs, methanol-degrading heterotrophs; slowly biodegradable chemical oxygen demand; and inert biomass for substrate (when appropriate) and space inside the biofilm; and biofilm and suspended biomass compartments, which compete for both the electron donor and electron acceptor. The model assumed identical reaction kinetics for suspended biomass and biofilm bacteria.

Later Boltz *et al.* [34] reported on the state of the art mathematical modeling of biofilms and biofilm reactors for engineering design. Boltz and Daigger [35] also studied uncertainty in bulk liquid hydrodynamics and biofilm dynamics in biofilm reactor design. They considered the uncertainties of liquid hydrodynamics on biofilm thickness control, surface area, and development and the biofilm dynamics influence on biofilm structure, thickness and function. From a substrate transformation perspective, the mass transfer by diffusion limitation in biofilm reactors controls, while the suspended growth systems are kinetically or biomass limited.

The kinetics of biodegradation of phenolic wastewater in a biofilm reactor was investigated by Lin and Hsien [36]. A laboratory scale column reactor was employed to validate the model, based on diffusive mass transfer and Haldane kinetics. Removal efficiency was 94% to 96.5% for different hydraulic retention time at a steady state conditions. Polyurethane foam sponge cubes were used by Lin [37] as a biofilm support medium in modeling of biodegradation of textile wastewater. Biofilm and suspended biomass reached a maximal value in the steady state when the substrate flux reached a constant value and remained maximal. Approximately 33% of the substrate concentration (as COD) was converted to CO_2 during biodegradation in the fixed-biofilm reactor (FBR) test.

3.1. Computational Method

Sen and Randall [38] combined the integrated fixed-film activated sludge (IFAS), and moving-bed biofilm reactor (MBBR) systems in a model. The model embeds a biofilm model into a multicell activated sludge model. The biofilm flux rates for organics, nutrients, and biomass could be computed by two methods—a semi-empirical model of the biofilm that is relatively simpler, or a diffusional model that is computationally intensive. Later Sen and Randall [39] developed a diffusional model of the biofilm that can be applied in lieu of a semi-empirical model to upgrade an activated sludge system to an integrated fixed-film activated sludge (IFAS) or moving-bed biofilm reactor (MBBR) system. The model has been developed to operate with up to 12 cells (reactors) in series, with biofilm media incorporated to one or more of the zone cells, except the anaerobic zone cells.

Optimal control of film casting processes was investigated by Selvanayagam *et al.* [40]. The control goal was to establish an even thickness profile and minimal cost.

4. Trickling Filters

The impact of organic carbon on the performance of a high rate nitrifying trickling filter designed for pretreatment of potable water was discussed by van den Akker *et al.* [41,42]. When organic load increased beyond 0.75 to 2.1 g $sBOD_5/m^2d$, a linear decline in nitrification from 70% down to 15% was observed in 8 to10 d. In addition, van den Akker *et al.* [43] reported on the application of high rate nitrifying trickling filters to remove low concentrations (approximately 3 mg/L) of ammonia from reclaimed municipal wastewater. Nitrification was impeded by high organic carbon loads and aquatic snails. Nitrifying microorganisms in fixed bed biofilm reactors fed with different nitrite and ammonia concentrations were reported by ter Haseborg *et al.* [44]. Increased abundance of Nitrobacter occurred during high nitrite concentration periods.

Mondal and Warith [45] reported using shredded tire chips as packing media in trickling filter systems for landfill leachate treatment. Matthews *et al.* [46] evaluated treatment of landfill leachate using passive aeration trickling filters, reporting on the effects of leachate characteristics and temperature on rates and process dynamics. Ziolko *et al.* [47] reported about 49% effectiveness of conventional trickling filters at reducing copper concentrations in wastewaters. Little effect was noted on dissolved copper forms.

5. Horizontal Flow Bioreactor

Nitrogen dynamics and removal in a horizontal flow biofilm reactor (HFBR), step-feed wastewater treatment process was investigated by Clifford *et al.* [48]. They reported significant simultaneous nitrification and denitrification in the nitrifying zone. The results can be used to optimize HFBR reactor design and provide an alternative, low maintenance, economically efficient system for carbon and nitrogen removal for low flow wastewater discharges. Dairy wash water treatment using step-feed HFBR systems was investigated by Rodgers *et al.* [49]. Removals up to 99.7% for BOD_5, 96.7% for total COD, and 98.7% for nitrogen were reported.

6. Rotating Biological Contactors

Fixed biomass technologies, such as rotating biological contactors (RBC) or biodiscs, have been applied for wastewater treatment both in large and medium-small-sized agglomerations. The results obtained by Coello *et al.* [50] confirmed the potential of the microbial activity measurements studied for an accurate characterization of biofilms and biomass activity estimation in fundamental research and for the practical operation and control of fixed biomass wastewater treatment systems.

A polyurethane rotating disc system for post treatment of anaerobically pretreated sewage was investigated by Tawfik and Klapwijk [51]. The performance of polyurethane discs *versus* polystyrene rotating discs for the treatment of an upflow anaerobic sludge blanket reactor effluent fed with domestic water was investigated. Polyurethane discs provided superior ammonia removal at equal organic loading rate (OLR). Experimental results clearly showed a strong and immediate detrimental effect of imposing high OLRs of 26 gCOD/m^2d on the nitrification process in the nitrifying RBC unit.

7. Fluidized Bed and Air Lift Bioreactors

7.1. Anoxic/Aerobic Reactors

High-strength nitrogenous wastewater treatment in biofilm and granule anammox processes was studied by Kim *et al.* [52]. The biofilm reactor showed high NH_4^+-N and NO_2^--N removal efficiencies of over 88% and 94%, respectively, until total nitrogen concentration reached 800 mg N/L. Better nitrogen removal performance, NH_4^+-N and NO_2^--N removal efficiencies over 90% up to total nitrogen concentration of 1,400 mg N/L, was achieved with the granule reactor than the biofilm reactor.

Biofilm and membrane filtration processes for reclamation and reuse of rural wastewater was investigated by Hyun and Lee [53]. The performances of a three-stage process of anaerobic-oxic-anoxic biofilm filtration (AOBF) and membrane filtration (MF) processes were assessed for the potential for reclamation and reuse of blended wastewater containing domestic wastewater, black water, and landfill leachate. The effluent of the AOBF/MF system met the reuse standard for industrial and agricultural water, while effluent of AOBF met the standard for wastewater treatment plant effluent quality.

Biological removal of nitrogen from wastewater was reviewed and reported by Zhu *et al.* [54]. The comprehensive review included consideration of SND, OLAND, SHARON, CANON, and ANAMMOX processes and their advantages and disadvantages. A two-stage bioaugmented anoxic-oxic (A/O) biofilm process treating petrochemical wastewater under different DO concentrations was investigated by Guo *et al.* [55]. Polyurethane foam cube biofilm medium was used and dissolved oxygen (DO) concentration was studied as a crucial environmental factor on the system performance. Lower DO concentration took less time to start-up, obtained higher pollutants removal efficiency and had greater resistance to shock loadings compared to the system with higher DO level.

7.2. Airlift Reactor

Walters *et al.* [56] researched a biofilm airlift suspended (BAS) reactor for autotrophic nitrification and denitrification. Guo *et al.* [57] developed a novel airlift combined anoxic-oxic biofilm reactor for nitrogen removal and studied the influence of carbon to nitrogen ratio and aeration rate on nitrification and denitrification.

8. Submerged Bed Biofilm Reactors

A submerged reactor was tested by Bravo and Spyra [58] as a modification to the conventional trickling-flow configuration. This modified fixed-film reactor was effective when high loadings of diesel were present as an emulsion. Wirthensohn *et al.* [59] investigated physical and biological treatment steps for the remediation of groundwater from a former MGP site in a pilot plant. The scope of the study was to test the effectiveness of different process steps, which included an aerated sedimentation basin, a submerged fixed film reactor (SFFR), a multi-media filter, and an activated carbon filter. The treatment system was effective in reducing the various organic and inorganic pollutants in the pumped groundwater.

8.1. Hybrid Reactor

Downing and Nerenberg [2,60] investigated total nitrogen removal in a hybrid, membrane-aerated activated sludge process in which a nitrifying biofilm grows on the membrane and the bulk liquid is anoxic thereby promoting nitrate/nitrite reduction by low bulk liquid BOD. Shortcut nitrogen removal was confirmed using microsensor measurements, showing that nitrite was the dominant form of oxidized nitrogen produced by the biofilm. Nitrification rates decreased during short-term spikes in bulk liquid BOD concentrations. The hybrid membrane biofilm process (HMBP) obtained full denitrification when sufficient BOD was available in the influent.

An up-flow anaerobic sludge blanket (UASB) and down-flow hanging sponge (DHS) system was investigated for removal COD, BOD_5, ammonia, and fecal coliform from domestic water by Tawfik *et al.* [61]. The combined system achieved reduction in TSS and improvement in fecal coliform removal. Nitrification occurred mainly in the lower DHS zone.

8.2. Sequencing Batch Biofilm Reactor (SBBR)

Kritsunankul and Wantawin [62] investigated the possibility of using a biofilm process for partial nutrient removal from digested swine wastewater containing low ratios of chemical oxygen demand (COD) to nitrogen and phosphorus. Biomass from the reactor consisted of denitrifying polyphosphate accumulating organisms (DNPAO) that can use nitrite as an electron acceptor, based on activity tests. Organic carbon in the digested swine wastewater was utilized effectively through the denitrifying phosphorus uptake process.

Yang *et al.* [63] researched an integrated biofilm reactor, using polyacrylonitrile balls, and an anthracite gravitational filtration bed in a sequencing batch reactor (SBR) to aerobically treat a municipal wastewater. Seasonal influence on COD and nitrogen removal by the biofilm reactor was significant. Nitrogen and phosphorus removals were limited by COD levels in the wastewater. The filtration process removed considerable COD, nitrogen, phosphorus, and turbidity, and all secondary treatment standards in China were met except phosphorus.

Gonzalez *et al.* [64] evaluated a combined strategy of a photo-Fenton pretreatment followed by an aerobic SBBR for total C and N removal from a synthetic wastewater containing exclusively 200 mg/L of the antibiotic Sulfamethoxazole (SMX). The SBBR was reported to be an efficient way to treat wastewaters contaminated with biorecalcitrant pharmaceuticals, such as the SMX, and could denitrify with the inclusion of an anoxic stage in SBBR operation.

Zhou *et al.* [65] developed a novel 2-sludge, 3-stage process using a combination of granular sludge SBR and biofilm to achieve biological removal of nitrogen and phosphorus from nutrient-rich wastewater. The system used alternating anaerobic/anoxic granular sludge SBR conditions, supplemented by a short aerobic phase and followed by an aerobic biofilm SBR to produce an effluent suitable for land irrigation. Wantawin *et al.* [66] employed lab-scale sequencing batch biofilm reactors (SBBRs) inoculated with normal nitrifying sludge to study the potential of an oxygen-limited autotrophic nitrification-denitrification process initiated with typical nitrifying sludge for treating a synthetic ammonia wastewater devoid of organic carbon in one step. Occurrence of aerobic ammonia oxidizing bacteria (AAOB) and competition between aerobic nitrite oxidizing bacteria (ANOB) and anaerobic ammonia-oxidizing bacteria (anammox) were examined.

Andrade do Canto *et al.* [67] investigated the biological removal of ammonium nitrogen from synthetic wastewater by the simultaneous nitrification/denitrification (SND) process, using a sequencing batch biofilm reactor (SBBR). This process was potentially viable in post-treatment of wastewater containing ammonium nitrogen. Cherchi *et al.* [68] evaluated and compared MicroC (Environmental Operating Solutions, Bourne, Massachusetts), methanol and acetate carbon sources in terms of their denitrification rates and kinetics, effect on overall nitrogen removal performance and microbial community structure of carbon-specific denitrifying enrichments. Denitrification and kinetics were determined from biomass obtained from laboratory scale sequencing batch reactors or full-scale plants. The feasibility of the use of MicroC for denitrification processes was demonstrated. Zhang *et al.* [69] reported that biological nitrogen removal can be enhanced by simultaneous nitrification and denitrification (SND) via nitrite with a sequencing batch biofilm reactor (SBBR) at certain temperatures.

8.3. Moving Medium Biofilm Reactors

Addition of sodium carbonate was found to have a beneficial effect on pH control, nitrification, and ammonia oxidizing bacteria (AOB) ecology in a full-scale powdered activated carbon treatment (PACT) and a pilot-scale moving bed biofilm reactor (MBBR) treating petrochemical wastewater with high strength of ammonia Whang *et al.* [70].

Bill *et al.* [71] did research to evaluate the effectiveness of four different electron donors, specifically methanol, ethanol, glycerol and sulfide in post-denitrifying bench-scale MBBRs. Maximum denitrificaion rate measurements from profile testing suggested that sulfide, ethanol and glycerol substrates exhibited rates greater than methanol. Di Trapani *et al.* [72] compared the traditional activated sludge system system (AS) and a hybrid moving bed biofilm reactor (HMBBR). The HMBBR system obtained better organic matter removal as well as ammonium removal than the conventional AS system.

Tawfik *et al.* [73] evaluated the performance of a laboratory-scale sewage treatment system composed of an up-flow anaerobic sludge blanket (UASB) reactor and a moving bed biofilm reactor (MBBR) at temperatures of 22 °C to 35 °C and at different hydraulic retention times (HRT). Ammonia removal was a function of organic loading rate (OLR). A 13.3-h HRT was recommended for the mixed system. Joao *et al.* [74] did research in which industrial wastewater treated by the activated sludge process, was fed to moving-bed reactors and its salinity was increased. Residual substances present in the saline treated industrial wastewater had a strong inhibitory effect on the nitrification process.

8.4. Granules

Yilmaz *et al.* [75] investigated the biological removal of nitrogen and phosphorus from nutrient-rich abattoir wastewater using granular sludge in SBRs. The result showed that good nutrient removal was achieved by the presence of large anoxic zones in the center of the granules. Diffusion transfer in the granules, rather than sludge settleability, controlled minimum HRT. Beliavski *et al.* [76] did research on denitrification of brines originating from membrane treatment of ground water in an upflow sludge blanket (USB) reactor, a biofilm reactor without carrier. Ethanol and acetic acid denitrification electrons donors were compared. Acetic acid was found to be the more suitable electron donor substrate for brine denitrification and produced excellent sludge settling characteristics.

8.5. Photosynthetic Biological Sulfide Removal (BSR) Biofilm System

Several systems using phototrophic bacteria, often green sulfur bacteria, have been proposed for removal of malodorous, corrosive and toxic sulfide from liquid wastes. Timothy *et al.* [77] discussed supply of light and other system issues as well as efficiency of light use by different phototrophic bacteria in BSR reactors. Li *et al.* [78] investigated sulfide removal by the simultaneous autotrophic and heterotrophic desulfurization-denitrification process, demonstrating the possibility to remove sulfide and organic carbon in the presence of nitrate and nitrite. Roeselers *et al.* [6] provided a review of the actual and potential applications of phototrophic biofilms in wastewater treatment, bioremediation and other areas.

9. Biological Granular Activated Carbon (BAC)

Pasukphun *et al.* [79] investigated the decolorization of textile wastewater in anaerobic/aerobic biological activated carbon (A/A BAC) system. The combination of mixed culture (MC) and biological activated carbon (BAC) in the system promotes decolorization and dye intermediate removal. The feasibility of using a granular activated carbon-biofilm configured packed column system in the decolorization of azo dye acid orange 7- containing wastewater was investigated by Ong *et al.* [80]. The decolorization rate increased along with the increasing of initial acid orange 7 concentrations until it reached an optimum level.

10. Membrane Bioreactors (MBR)

10.1. Nutrient Removal

Liang *et al.* [81] studied nitrogen removal for a submerged membrane bioreactor with mixed liquor recirculation (MLE/MBR) and a membrane bioreactor with integrated fixed biofilm medium (IFMBR). The MLE/MBR exhibited higher nitrifying bacteria diversity and nitrifying activity. Both reactors exhibited fouling. The recirculation of mixed liquor from the aerobic zone to the anoxic zone in the MBR resulted in higher microbial activities of heterotrophic and autotrophic bacteria in the MBR compared to those from IFMBR. Metabolic selection via alternating anoxic/aerobic processes has the potential of having higher bacterial activities and improved nutrient removal in MBR systems.

Modin *et al.* [82] found that methane-to-nitrate consumption efficiency in an aerobic methane oxidation coupled to denitrification (AME-D) process can be improved by using a membrane biofilm reactor. Feng *et al.* [83] reported that membrane aeration bioreactors could achieve a suitable NH_4^+/NO_2^- ratio (1:1 to 1:1.3) and reduced level of dissolved oxygen, providing cost effective and ideal pretreatment for anaerobic ammonium oxidation (ANAMMOX). A membrane bioreactor (MBR), seeded with enriched ammonia-oxidizing bacteria (AOB), maintained a high nitrite ratio and nitrification efficiency at HRT of 24 h and a subsequent anaerobic packed-bed biofilm reactor (PBBR) showed satisfactory denitrification efficiency and low nitrite and nitrate concentration according to Zhang *et al.* [84].

10.2. Fouling

Biomass characteristics of membrane bioreactors studied by Liang *et al.* [81] found that the membrane fouling due to bacterial growth was evident in both the reactors. Membrane biofilm reactors (MBfR) utilize membrane fibers for bubble-less transfer of gas by diffusion and provide a surface for biofilm development. Hwang *et al.* [85] carried out nitrification and subsequent autotrophic denitrification in an MBfR with pure oxygen and hydrogen supply, respectively, in order to remove nitrogen without the use of heterotrophic bacteria. Biomass accumulation and scouring could be balanced in autotrophic denitrification and long-term stable operation can be maintained for these reactors.

10.3. Submerged MBR

Kimura *et al.* [86] studied the feasibility of nitrogen removal from municipal wastewater by simultaneous nitrification and denitrification in a baffled membrane bioreactor (BMBR). It was found that denitrification was the limiting step in removal of nitrogen in the BMBR. Total organic carbon (TOC), total phosphorus (T-P) and total nitrogen (T-N) removals were 85%, 97% and 77%, respectively.

10.4. Applications and Other

Performance and microbial ecology of an anaerobic/aerobic sequencing batch reactor (SBR) and an aerobic membrane bioreactor (MBR) treating thin film transistor liquid crystal display (TFT-LCD) wastewater was in investigated by Wu *et al.* [87]. Both reactors achieved about 99% degradation efficiencies for three important difficult-to-biodegrade, nitrogen-bearing organic compounds in the waste, but their microbial ecologies were very different. Whang *et al.* [88] conducted a study of long-term performance of an aerobic membrane bioreactor (MBR), treating TFT-LCD wastewater. Experimental results suggested inhibition of nitrifying bacteria at concentrations of 250 and 50 mg/L, respectively, for monoethanolamine and tetra-methyl ammonium hydroxide. Dimethyl sulfoxide was noninhibitory at concentrations up to 100 mg/L.

Syron and Casey [89] provided a comparative performance rate analysis of the membrane aerated biofilm reactor (MABR) in terms of its application for carbonaceous pollutant removal, nitrification/denitrification and xenobiotic biotreatment and discussed scale-up challenges. Li *et al.* [90] also provided a recent patent and literature review of MABR processes, including characteristics, membranes, modular design, operation parameters, and potential applications. In addition, Li *et al.* [91] evaluated MABR treatment of wastewater containing acetonitrile, reporting that acetonitrile degrading bacteria secrete more EPS, thereby enhancing membrane biofilm attachment and development. Sahu *et al.* [92] performed hydrogenotrophic wastewater denitrification in a hollow fiber membrane bioreactor (HFMB) using hydrogen gas in the fiber lumen. Dissolved oxygen in the bulk liquid did not adversely affect overall nitrogen removal.

Yang *et al.* [93] developed a new type of moving bed membrane bioreactor (MBMBR) by using carriers instead of activated sludge in a membrane bioreactor (MBR) and investigated the removal efficiency for synthetic wastewater and characteristics of simultaneous nitrification and denitrification (SND) performance. Good organics removal and SND performance was achieved during the 67-day experimental period. Yang *et al.* [94,95] compared a conventional membrane bioreactor (CMBR) and a MBMBR, focusing on organic carbon and nitrogen removal and on membrane fouling. Although the MBMBR organic carbon and nitrogen removal was far superior, the MBMBR exhibited more filamentous bacteria in the suspended solids and three times the rate of fouling of the CMBR.

Chen *et al.* [96] also developed a hybrid biological nitrogen removal system, consisting of an aerobic tank and an anoxic tank with an intermediate sludge settler connected to an MBR, which provided both nitrification and denitrification. Hwang *et al.* [97] investigated total nitrogen removal in a two-step membrane biofilm reactor (MBfR) system incorporating sequential nitrification and hydrogen driven autotrophic denitrification in order to achieve nitrogen removal by autotrophic bacteria alone. This investigation showed that, with an appropriate biofilm control plan, stable long-term operation of a fully autotrophic MBfR system for total nitrogen removal was possible without major membrane cleaning procedures. Hwang *et al.* [98] concluded that MBfR biomass accumulation and scouring can be balanced in autotrophic denitrification using gas sparging control.

Celmar-Repin *et al.* [99] reviewed the research on MBfR equipment, with varied types of porous and non-porous membrane material and membrane module configuration for autotrophic nitrogen removal. The authors also discussed the challenges ahead before MBfR can reliably be used on full-scale treatment plants.

10.5. Fixed Film and Xenobiotics

The effectiveness of bioaugmentation and transfer of plasmid to mixed microbial populations in pilot and laboratory scale SBBRs treating synthetic wastewater containing benzyl alcohol (BA) as a model xenobiotic was studied by Venkata *et al.* [100]. Engineering principles and requirements for high-rate biotreatment with membrane-aerated biofilm were investigated by Syron and Casey [89]. This paper provides a comparative performance rate analysis of the MABR in terms of its application for carbonaceous pollutant removal, nitrification/denitrification and xenobiotic biotreatment.

Real time analysis of Escherichia coli biofilms was investigated by Peitzsch *et al.* [101], who found that microbial communities grow more stably when they are associated with surfaces or organized in aggregates. This advantage of biofilms is technically exploited for the degradation of xenobiotics or in biocatalysis, where the fixed biomass has the added advantage of easier separation of excreted products. Efficient dye decolorization and production of dye decolorizing enzymes by a liquid and solid hybrid culture was researched by Shimokawa *et al.* [102], demonstrating that Thanatephorus cucumeris Dec 1, a basidiomycete, is a promising decomposer of several xenobiotics. Air-membrane surface bioreactor culture (AMS culture), a hybrid between solid-state and submerged culture, was used for Dec 1 growth.

An automatic biodetector of water toxicity as a tool for examination of phenol and cyanide contaminated water was investigated by Woznica *et al.* [103]. Magnitudes of toxic effect were proportional to concentration, whereas kinetics of the response is an indicator of the mechanism of toxicity.

A review of analytical methods and removal processes of the endocrine disruptor, 17-ethinylestradiol, was presented by Clouzot *et al.* [104]. The AS, MBRs, biofilm reactors, and SBR technologies appeared to have potential for improved endocrine disruptor removal. Modeling of 1,2-dichloroethane biodegradation by Klebsiella oxytoca va 8391 immobilized on granulated activated carbon was done by Mileva *et al.* [105]. Biodegradation potential of the genus Rhodococcus was investigated by Martinkova *et al.* [106]. The large genomes, catabolic pathway versatility, hydrophobic compound uptake and metabolism capability, biofilm formation qualities, persistence in hostile environments, and availability of genetic engineering tools in Rhodococci make them suitable industrial microorganisms for biotransformations and the biodegradation of many organic compounds.

11. Immobilized Cell Reactors (MBR)

The immobilized cell fluidized bed reactor and contact oxidation biofilm reactor are two common treatment choices for high strength ammonia wastewaters. Qiao *et al.* [107] thoroughly studied nitrification performance of the two reactors, concluding that the immobilized cell fluidized bed reactor had a shorter acclimation period and offered advantages over the contact oxidation biofilm reactor. Paslawski *et al.* [108] introduced a model for biodegradation of naphthenic acid in an immobilized cell reactor.

12. Innovative Reactors and Systems

Katuri and Scott [109] reported the performance of a prototype up-flow single-chambered microbial fuel cell (MFC) for electrical power generation using brewery wastewater as fuel. In the experiment a stable current density of approximately 2,270 mA/m^2 was generated. Venkata *et al.* [110] evaluated the effect of anodic biofilm growth and extent of its coverage on the anodic surface of a single-chambered, mediatorless MFC for bioelectricity generation using designed synthetic wastewater and chemical wastewater as substrates and anaerobic mixed consortia as biocatalyst. Anodic biofilm formation enhancement of extracellular electron transfer in the absence of mediators was shown. Venkata *et al.* [111]

also evaluated biochemical functioning of a single-chambered microbial fuel cell (MFC) using glass wool as the proton exchange membrane operated with selectively-enriched, acidogenic mixed culture for bioelectricity production and wastewater treatment. Higher current density was observed at acidophilic conditions. Venkata *et al.* [112] also studied the effect of catholyte on bioelectricity production from wastewater treatment in dual-chambered MFC, using selectively-enriched, mixed microflora.

Venkata *et al.* [113] evaluated the function of MFC as a bio-electrochemical treatment system in concurrence with power generation at high loading conditions (18.6 g COD/L; 56.8 g TDS/L). Marked improvement in power output was observed at applied higher substrate loading rate for extended periods without process inhibition. Mohanakrishna *et al.* [114] did research in evaluating open-air cathode MFC as a bio-electrochemical treatment system for distillery wastewater during bioelectricity generation. In addition to marked improvement in electricity generation at higher substrate loading, efficient treatment of distillery wastewater and multiple pollutants was demonstrated.

Gapes and Keller [115] did research on the impact of oxygen mass transfer on nitrification reactions in suspended carrier reactor biofilms. The result showed that heterogeneous biofilms grown under high ammonium loadings had much greater area-specific rates of nitrification than the gel-like biofilms sourced from low loaded systems.

Kim *et al.* [116] discussed the effects of integrated fixed film activated sludge (IFAS) media on activated sludge (AS) settling in biological nutrient removal systems. They found the suspended solids density in conventional AS to be slightly greater than suspended solids density in IFAS. The stability and capacity of AS are enhanced by IFAS as system loadings increased, according to Sriwiriyarat *et al.* [117]. Carbon removal and nitrification were compared for varying HRT and solids retention time (SRT). Attached biomass in IFAS suppressed the growth of filamentous microorganisms. Sriwiriyarat *et al.* [118] also investigated the effects of dissolved oxygen (DO) on biological nitrogen removal in IFAS systems at various chemical oxygen demand (COD carbon) to nitrogen ratios (C/N). Optimal DO concentrations were found to be about 6 mg/L for nitrogen removal at low C/N and 2 mg/L at high C/N, but COD removal was relatively unaffected by C/N. It was suggested that IFAS is not beneficial at C/N of 10 or higher.

The effects of influent substrate concentration load, hydraulic load and total nitrogen load on total nitrogen removal rate were compared in an up-flow ANAMMOX bio-filter reactor by Tian *et al.* [119]. The results showed that the maximum nitrogen removal rate in the ANAMMOX reactor increased linearly as the increasing of influent substrate concentration load, hydraulic load and total nitrogen load, respectively.

13. Biofilm on Sand, Soil and Sediments

Campos *et al.* [120] did a study in which two arsenite-oxidizing strains were isolated from volcanic rocks obtained from the Camarones Valley, Atacama Desert, Chile. Strains were isolated from biofilms and identified by 16s ARNr sequences analysis and arsenic oxidizing genes were detected by RT-PCR. The arsenic oxidation ability was assayed with silver nitrate and HPLC-HG-AAS. This study showed the way to further studies aimed at implementing biological systems to treat arsenic rich wastewater. Gorbushina and Broughton [121] studied the microbiology of the atmosphere-rock interface and also tried to explore how biological interactions and physical stresses modulate a sophisticated microbial ecosystem.

Newton and Wilson [122] did research on the recirculating gravel filter (RGF) in which pre-settled wastewater was recirculated through a gravel filter bed, while a biofilm on the filter media oxidized the organic matter and ammonia. Effluent from the RGF process had equivalent or lower effluent concentrations of BOD_5, TSS and ammonia nitrogen than other wastewater treatment processes typically employed in small communities.

13.1. Wetlands

Zhang *et al.* [123] commented that the concentrations of nutrients (N and P) in the wastewater and loading rate to constructed wetlands may influence the nutrient removal from secondary-treated municipal wastewater by wetland plants. High N concentrations may hamper Schoenoplectus validus growth. Pollard [124] aimed to apply the thymidine assay to quantify bacterial growth without disturbing the biofilm on the surfaces of emergent Schoenoplectus validus of a constructed wetland. He concluded that in the constructed wetlands of this study, biofilm-bacterial specific growth rates, compared to those of natural ecosystems, could be markedly improved through changes in wetland design that increased bacterial respiration while minimizing biofilm growth.

Iasur-Kruh *et al.* [125] investigated the assembly and function of microbial populations in vertical-flow constructed wetland microcosms designed to improve the quality of wastewater after activated sludge treatment. The performance of 3-year-old wetland ponds was investigated. Improvements in water parameters such as coliform level, ammonia concentration, BOD and TSS were observed. Yin *et al.* [126] installed three lab-scale horizontal subsurface flow constructed wetlands (HSSF CWs) to demonstrate the use of CWs as a viable low-cost treatment option to purify polluted scenery water. Bacterial compositions, present in 3 CWs, were characterized. The sequence analysis showed ammonia-oxidizing bacteria (AOB) in CWs were uncultivable and the population of AOB had a higher percentage of *Nitrosomonas*-like sequences from wetlands, while no *Nitrosospira*-like sequences were found. Tuszynska and Obarska-Pempkowiak [127] analyzed the operation of three hybrid CW systems composed with vertical flow (VF-CW) and horizontal flow (HF-CW) constructed wetlands. The analyses were carried out in two wetlands (CWs) located in northern Poland and one in Germany. The analysis found that increase of organic matter concentration in filter material of first beds in plants caused clogging and decreased removal efficiency.

Hijosa-Valsero *et al.* [128] operated seven mesocosm-scale CWs of different configurations for nine months to assess their ability to remove pharmaceuticals and personal care products (PPCPs) from urban wastewaters. They found out that the presence of plants favored the removal of some PPCPs. The performance of the mesocosm studied was compound-dependant. Soilless CWs showing the highest removal efficiency for ketoprofen, ibuprofen and carbamazepine, while free-water CWs with effluent leaving through the bottom of the tank performed well for the degradation of ketoprofen, salicylic acid, galaxolide and tonalide. Subsurface horizontal flow CWs were efficient for the removal of caffeine.

Zhao *et al.* [129] investigated the developing process of clogging caused by biofilm growth or organic particle accumulation instead of total organic matter accumulation in two groups of lab-scale vertical flow constructed wetlands (VFCWs), which were fed with glucose (dissolved organic matter) and starch (particulate organic matter) influent. Growth of biofilms within the substratum pores certainly caused remarkable reduction of effective porosity, especially for the strong organic wastewater, whereas its influence on infiltration rate was negligible. Imfeld *et al.* [130] summarized the recent progress made towards understanding how the various mechanisms attributed to organic chemicals removal interact to form a functioning wetland.

13.2. Riverine Sediment

Bonnineau *et al.* [131] reported that fluvial biofilms are a pertinent tool in assessing beta-blocker toxicity. They conducted acute toxicity tests of the effect of metoprolol, propranolol and atenolol on fluvial biofilms, observing the alteration of biofilm algae (photosynthesis efficiency) and biofilm bacteria (peptidase activity and mortality).

Triclosan, a common bactericide, survives several degradation steps of wastewater treatment. Ricart *et al.* [132] examined the short-term effects of triclosan on biofilm algae and bacteria, finding that the no effect concentration was 0.21 μg/L and that environmentally relevant concentrations caused increased algae and, to a greater extent, bacteria mortality and photosynthesis efficiency inhibition. Relevance was suggested beyond the aquatic habitat environment, including wastewater treatment plant processes.

Writer, *et al.* [133] reported that steroidal hormones and alkylphenols are attenuated by and accumulated or concentrated in biofilms on stream sediments. Implications were: (1) that this serves as a mechanism for these contaminants to enter the food chain when aquatic organisms consume the biofilm; and (2) that wastewater treatment plant biofilm processes may represent viable means of reducing the presence of these pollutants in wastewater treatment plant discharges.

14. Discussion

Fixed-film biological systems have been used widely in the treatment of wastewater, particularly in the attainment of secondary effluent standards and nitrification. Biofilm measurement and characterization advances in the past three years, including ATR-FTIR, CLSM, image analysis, and microsensor techniques, are helping to unlock the three dimensional understanding of biofilm community structure and composition, and the role of EPS in attachment and detachment mechanisms and in nutrient removal. Phototropic and electro-active biofilms studies are opening new potentials for nutrient control with biofilms and for microbial fuel cell technology.

Models of integrated fixed-film activated sludge and moving-bed biofilm reactors represent the state of the art in recent modeling efforts. These models address substrate transformation in diffusion-limited mass transfer of biofilms and kinetically or biomass-limited mass transfer of suspended growth systems. Modeling challenges remain in the uncertainties of bulk liquid hydrodynamics on biofilm thickness control, surface area, and development and of biofilm dynamics influence on biofilm structure, thickness and function. Haldane kinetics has been used in modeling of wastewaters possessing toxicity.

Biofilm growth support media research continues to investigate alternative materials, including biological granular activated carbon, shredded tire chips, coconut fiber, and polyurethane for organic carbon and nitrogen removal systems, Anoxic/oxic bioreactors, submerged bed biofilm reactors and hybrids, and membrane bioreactors and hybrids are areas of state of the art research in carbonaceous pollutant removal, nitrification/denitrification and xenobiotic control. Membrane biofouling remains a significant challenge to stable operation in some of these bioreactors and hybrids.

Biofilm research in the natural environment of sand, soil, sediments and wetland vegetation has revealed the potential that biofilms can have capabilities to polish secondary treated wastewaters and to attenuate and concentrate certain contaminants in the biofilm. Biofilms may have the capability to remove PPCPs, steroidal hormones, alkylphenols, and metals from secondary effluent and natural waters containing low levels of these contaminants. At the same time, the tendency to concentrate the contaminants in biofilm in the natural environment can serve the negative function of providing a mechanism of entry of the contaminants into the aquatic organism food chain.

15. Conclusions

Biofilm systems continue to draw significant research attention. While this review is not an all-encompassing documentation for the last three years, it does provide an opportunity to reflect on what biofilm and hybrid biofilm systems may still have to offer the wastewater and environmental research and engineering community. Future research will likely extend the focus of biofilm systems application in the areas of nutrient control, removal of traces of PPCPs, steroidal hormones, metals and other contaminants, and microbial fuel cell technology.

References

1. Downing, L.S.; Nerenberg, R. Effect of oxygen gradient on the activity and microbiol community structure of a nitrifying, membrane aerated biofilm. *Biotecnol. Bioeng*. **2008**, *101*, 1193-1204.
2. Downing, L.S.; Nerenberg, R. Total nitrogen removal in a hybrid, membrane-aerated activated sludge process. *Water Res.* **2008**, *42*, 3697-3708.
3. McLamore, E.S.; Zhang, W.; Porterfield, D.M.; Banks, M.K. Membrane-Aerated biofilm proton and oxygen flux during chemical toxin exposure. *Environ. Sci. Technol.* **2010**, *44*, 7050-7057.
4. Andersson, S.; Kuttuva, R.G.; Land, C.J.; Dhalhammar, G. Biofilm formation and interaction of bacterial strains commonly found in wastewater treatment systems. *FEMS Microbiol. Lett.* **2008**, *283*, 83-90.
5. Shoji, T., Ochi, S.; Ozaki, M. Characterization of bacterial biofilm communities in tertiary treatment processes for wastewater reclamation and reuse. *Water Sci. Technol.* **2008**, *58*, 1023-1030.
6. Roeselers, G.; Loosdrecht, M.C.; Muyzer, G. Phototrophic biofilms and their potential applications. *J. Appl. Phycol.* **2008**, *20*, 227-235.
7. Jechalke, S.; Vogt, C.; Reiche, N.; Franchini, A.; Borsdorf, H.; Neu, T.; Richnow, H. Aerated treatment pond technology with biofilm promoting mats for the bioremediation of benzene, MTBE and ammonium contaminated groundwater. *Water Res.* **2010**, *44*, 1785-1796.
8. Guzzon, A.; Bohn, A.; Diociauti, M.; Albertano, P. Cultured phototrophic biofilms for phosphorus removal in wastewater treatment. *Water Res.* **2008**, *42*, 4357-4367.

9. Tian, J.; Chen, Z.; Nan, J.; Liang, H.; Li, G. Integrative membrane coagulation adsorption bioreactor (MCABR) for enhanced organic matter removal in drinking water treatment. *J. Membr. Sci*. **2010**, *352*, 205-212.
10. Marconnet, C.; Houari, A.; Galas, L.; Vaudry, H.; Heim, V.; Di Martino, P. Biodegradable dissolved organic carbon concentration of feed water and NF membrane biofouling: A pilot train study. *Desalination* **2009**, *242*, 228-235.
11. Spettmann, D.; Eppmann, S.; Flemming, H.; Wingender, J. Visualization of membrane cleaning using confocal laser scanning microscopy. *Desalination* **2008**, *224*, 195-200.
12. Bjerkey, A.; Fiksdal, L. Characterization of biofouling on hollow fiber membranes using confocal laser scanning microscopy and image analysis. *Desalination* **2009**, *245*, 474-484.
13. Delatolla, R.; Berk, D.; Tufenkji, N. Rapid and reliable quantification of biofilm weight and nitrogen content of biofilm attached to polystyrene beads. *Water Res*. **2008**, *42*, 3082-3088.
14. Wojnowska-Baryla, I.; Cydzik-Kwiatkowska, A.; Zielinska, M. The application of molecular techniques to the study of wastewater treatment systems. *Methods Mol. Biol*. **2010**, *599*, 157-183.
15. He, S.; Li, J.; Xu, Y. Microbial Community Structure of Biological Contact Oxidation Process Used In Landscape River. In Proceedings of 3rd International Conference on Bioinformatics and Biomedical Engineering, Beijing, China, 11–13 June 2009; Article number: 5163050.
16. Cheng, K.C.; Demicri, A.; Catchmark, J.M. Advances biofilm reactors for production of value added products. *Appl. Microbiol. Biotechnol*. **2010**, *87*, 445-456.
17. Di Pippo, F.; Bohn, A.; Congestri, R.; De Philippis, R.; Albertano, P. Capsular polysaccharides of cultured phototrophic biofilms. *Biofouling* **2009**, *25*, 495-504.
18. Avella, A.C.; Gomer, T.; Yvon, J.; Chappe, P.; Guinot-Thomas, P.; de Donato, P. A combined approach for a better understanding of wastewater treatment plants operation: Statistical analysis of monitoring database and sludge physico-chemical characterization. *Water Res.* **2011**, *45*, 981-992.
19. Lin, Y.H.; Wu, C.L.; Hsu, C.H.; Li, H.L. Biodegradation of phenol with chromium (6) reduction in anaerobic fixed biofilm process—kinetic model and reactor performance. *J. Hazard Mater*. **2009**, *172*, 1394-1401.
20. Gujer, W. Nitrification and me—A subjective review. *Water Res*. **2010**, *44*, 1-19.
21. Park, H.D.; Lee, S.Y.; Hwang, S. Redundancy analysis demonstration of the relevance of the temperature to ammonia oxidizing bacterial community composition in a full scale nitrifying bioreactor treating saline waste water. *J. Microbiol. Biotechnol*. **2009**, *19*, 346-350.
22. Weber, S.D.; Hofmann, A.; Pilhofer, M. The diversity of fungi in aerobic sewage granules assessed By 18s Rrna gene and its sequence analyses. *FEMS Microbiol. Ecol.* **2009**, *68*, 246-254.
23. Wang, Z.W.; Chen, S. Potential of biofilm-based biofuel production. *Appl. Microbiol. Biotechnol*. **2009**, *83*, 1-18.
24. Ergurder, T.H.; Boon, N.; Wittebolle, L.; Marzorati, M.; Verstraete, W. Environmental factors shaping the ecological niches of ammonia oxidizing archaea. *FEMS Microbiol. Rev.* **2009**, *33*, 855-869.
25. Santoro, A.E.; Francis, C.A.; De Sieyes, N.R.; Boehm, A.B. Shifts in the relative abundance of ammonia oxidizing bacteria and archaea across physicochemical gradient in subterranean estuary. *Environ. Microbiol.* **2008**, *10*, 1068-1079.

26. Erable, B.; Dunteanu, N.M.; Dumas, C.; Scott, K. Application of electro active biofilms. *Biofouling* **2010**, *26*, 57-71.
27. Debabov, V.G. Electricity from microorganisms. *Mikrobiologia* **2008**, *77*, 149-157.
28. Ren, Z.; Steinberg, L.M.; Regan, J.M. Electricity production and microbiol biofilm characterization in cellulose fed microbial fuel cells. *Water Sci. Technol*. **2008**, *58*, 617-622.
29. You, J.; Das, A.; Dolan, E.M.; Hu, Z. Ammonia oxidizing archaea involved in nitrogen removal. *Water Res*. **2009**, *43*, 1801-1809.
30. Andersson, S.; Dalhammar, G.; Kuttuva, R.G. Influence of microbial interactions and eps/polysaccharide composition on nutrient removal activity in biofilms formed by strains found in wastewater treatment systems. *Microbiol. Res.* **2010**, *166*, 449-457.
31. Trapani, D.D.; Mannina, G.; Torregrossa, M.; Viviani, G. Quantification of kinetic parameters for heterotrophic bacteria via respirometry in a hybrid reactor. *Water Sci. Technol.* **2010**, *61*, 1757-1766.
32. Wichern, M.; Lubken, M.; Horn, H. Optimizing sequencing batch reactor (SBR) reactor operation for treatment of dairy wastewater with aerobic granular sludge. *Water Sci. Technol.* **2008**, *58*, 1199-1206.
33. Boltz, J.P.; Johnson, B.R.; Daigger, G.T.; Sandino, J. Modeling integrated fixed-film activated sludge and moving-bed biofilm reactor systems I: Mathematical treatment and model development. *Water Environ. Res.* **2009**, *81*, 555-575.
34. Boltz, J.P.; Morgenroth, E.; Sen, D. Mathematical modelling of biofilms and biofilm reactors for engineering design. *Water Sci. Technol*. **2009**, *62*, 1821-1836.
35. Boltz, J.P.; Daigger, G.T. Uncertainty in bulk liquid hydrodynamics and biofilm dynamics creates uncertainties in biofilm reactor design. *Water Sci. Technol.* **2010**, *61*, 307-316.
36. Lin, Y.H.; Hsien, T.Y. Kinematics of biodegradation of phenolic wastewater in a biofilm reactor. *Water Sci. Technol.* **2009**, *59*, 1703-1711.
37. Lin, Y.H. Modeling the performance of biodegradation of textile wastewater using polyurethane foam sponge cube as a supporting medium. *Water Sci. Technol.* **2010**, *62*, 2801-2810.
38. Sen, D.; Randall, C.W. Improved computational model (Aquifas) for activated sludge, integrated fixed-film activated sludge, and moving-bed biofilm reactor systems, part I: Semi-Empirical model development. *Water Environ. Res.* **2008**, *80*, 439-453.
39. Sen, D.; Randall, C.W. Improved computational model (Aquifas) for activated sludge, integrated fixed-film activated sludge, and moving-bed biofilm reactor systems, part II: Multilayer biofilm diffusional model. *Water Environ. Res*. **2008**, *80*, 624-632.
40. Selvanayagam, K.; Götz, T.; Sundar, S.; Vetrivel, V. Optimal control of film casting processes. *Int. J. Numer. Methods Fluids* **2008**, *59*, 1111-1124.
41. van den Akker, B.; Holmes, M.; Cromar, N.; Fallowfield, H. Application of high rate nitrifying trickling filters for potable water treatment. *Water Res.* **2008**, *42*, 4514-4524.
42. van den Akker, B.; Holmes, M.; Cromar, N.; Fallowfield, H. The impact of organic carbon on the performance of a high rate nitrifying trickling filter designed to pre-treat potable water. *Water Sci. Technol.* **2010**, *61*, 1875-1883.

43. van den Akker, B.; Holmes, M.; Short, M.D.; Cromarm, N.J.; Fallowfield, H.J. Application of high rate nitrifying trickling filters to remove low concentrations of ammonia from reclaimed municipal wastewater. *Water Sci. Technol.* **2010**, *65*, 2425-2432.
44. ter Haseborg, E.; Zamora, T.M.; Fröhlich, J.; Frimmel, F.H. Nitrifying microorganisms in fixed-bed biofilm reactors fed with different nitrite and ammonia concentrations. *Bioresour. Technol.* **2010**, *101*, 1701-1706.
45. Mondal, B.; Warith, M.A. Use of shredded tire chips and tire crumbs as packing media in trickling filter systems for landfill leachate treatment. *Environ. Technol.* **2008**, *29*, 827-836.
46. Matthews, R.; Winson, M.; Scullion, J. Treating landfill leachate using passive aeration trickling filters; effects of leachate characteristics and temperature on rates and process dynamics. *Sci. Total Environ.* **2009**, *407*, 2557-2564.
47. Ziolko, D.; Halla, D.; Lester, J.N.; Scrimshaw, M.D. The effectiveness of conventional trickling filter treatment plants at reducing concentration of copper in wastewaters. *Sci. Total Environ.* **2009**, *407*, 6235-6241.
48. Clifford, E.; Nielsen, M.; Sorensen, K.; Rodgers, M. Nitrogen dynamics and removal in a horizontal flow biofilm reactor for wastewater treatment. *Water Res*. **2010**, *44*, 3819-3828.
49. Rodgers, M.; De Paoar, D.; Clifford, E. Dairy waste water treatment using a horizontal flow biofilm system. *J. Environ Manag*. **2008**, *86*, 114-120.
50. Coello, M.D.; Rodrigues-Barroso, M.R.; Anagon, C.A.; Quiroga, J.M. Use of microbial activity measurements for monitoring RBC biofilms. *Environ. Monit. Assess.* **2010**, *169*, 451-455.
51. Tawfik, A.; Klapwijk, A. Polyurethane rotating disc system for post treatment of anaerobically pretreated sewage was investigated. *J. Environ. Manag*. **2010**, *91*, 1183-1192.
52. Kim, I.; Lee, H.H.; Chung, Y.C.; Jung, J.Y. High-Strength nitrogenous wastewater treatment in biofilm and granule anammox processes. *Water Sci. Technol*. **2009**, *60*, 2365-2371.
53. Hyun, K.S.; Lee, S.J. Biofilm/Membrane filtration for reclamation and reuse of rural wastewaters. *Water Sci. Technol.* **2009**, *59*, 2145-2152.
54. Zhu, G.; Peng, Y.; Li, B.; Guo, J.; Yang, Q.; Wang, S. Biological removal of nitrogen from wastewater. *Rev. Environ. Contam. Toxicol*. **2008**, *192*, 159-195.
55. Guo, J.; Ma, F.; Chang, C.C. Start-Up of a two-stage bioaugmented anoxic-oxic (a/o) biofilm process treating petrochemical wastewater under different do concentrations. *Bioresuor. Technol*. **2009**, *100*, 3483-3488.
56. Walters, E.; Hille, A.; He, M.; Ochmann, C.; Horn, H. Simultaneous nitrification/denitrification in a biofilm airlift suspension (BAS) reactor with biodegradable carrier material. *Water Res.* **2009**, *43*, 4461-4466.
57. Guo, H.; Zhou, J.; Zhang, X. Nitrogen removal in an airlift combined A/O biofilm reactor. *Dalian Ligong Daxue Xuebao/J. Dalian Univ. Technol*. **2009**, *49*, 193.
58. Bravo, V.; Spyra, W.; Antaño-López, R. Biodegradation of high concentrations of benzene and diesel in a fixed-film reactor. *Water Air Soil Pollut.* **2009**, *204*, 351-361.
59. Wirthensohn, T.; Schoeberl, P.; Ghosh, U.; Fuchs, W. Pilot plant experiences using physical and biological treatment steps for the remediation of groundwater from a former MGP site. *J. Hazard Mater*. **2009**, *163*, 43-52.

60. Downing, L.S.; Nerenberg, R. Sustainable nitrogen removal from wastewater with the hybrid membrane biofilm process (HMBP): Bench-Scale studies. *Water Sci. Technol.* **2008**, *58*, 1715-1720.
61. Tawfik, A.; El-Gohary, F.; Ohashi, A.; Harada, H. Optimization of the performance of an integrated anaerobic aerobic system for domestic wastewater treatment. *Water Sci. Technol.* **2008**, *58*, 185-194.
62. Kritsunankul, C.; Wantawin, C. Partial nutrient removal under insufficient organic carbon from digested swine wastewater in sequencing batch biofilm reactor. *J. Environ. Sci. Health* **2008**, *43*, 1085-1092.
63. Yang, K.; He, J.; Dougherty, M.; Yang, X.; Li, L. Municipal wastewater treatment through an aerobic biofilm sbr integrated with a submerged filtration bed. *Water Sci. Technol.* **2009**, *59*, 917-926.
64. Gonzalez, O.; Esplugas, M.; Sans, C.; Torres, A.; Esplugas, S. Performance of a sequencing batch biofilm reactor for the treatment of pre-oxidized sulfamethoxazole solutions. *Water Res.* **2009**, *43*, 2149-2158.
65. Zhou, Y.; Pijuan, M.; Yuan, Z. Development of a 2-Sludge, 3-Stage system for nitrogen and phosphorus removal from nutrient-rich wastewater using granular sludge and biofilms. *Water Res.* **2008**, *42*, 3207-3217.
66. Wantawin, C.; Juateea, J.; Noophan, P.L.; Munakata-Marr, J. Autotrophic nitrogen removal in sequencing batch biofilm reactors at different oxygen supply modes. *Water Sci. Technol.* **2008**, *58*, 1889-1894.
67. Andrade do Canto, C.; Rodrigues, J.; Ratusznei, S.; Zaiat, M.; Foresti, E. Feasibility of nitrification/denitrification in a sequencing batch biofilm reactor with liquid circulation applied to post-treatment. *Bioresour. Technol.* **2008**, *99*, 644-654.
68. Cherchi, C.; Onnis-Hayden, A.; El-Shawabkeh, I.; Gu, A. Implication of using different carbon sources for denitrification in wastewater treatments. *Water Environ. Res.* **2009**, *81*, 788-799.
69. Zhang, L.; Wei, C.; Zhang, K.; Zhang, C.; Fang, Q.; Li, S. Effects of temperature on simultaneous nitrification and denitrification via nitrite in a sequencing batch biofilm reactor. *Bioprocess Biosys. Eng*. **2009**, *32*, 175-182.
70. Whang, L.M.; Yang, K.H.; Yang, Y.F.; Han, Y.L.; Chen, Y.J.; Cheng, S.S. Microbial ecology and performance of ammonia oxidizing bacteria (AOB) in biological processes treating petrochemical wastewater with high strength of ammonia: Effect of Na(2)Co(3) addition. *Water Sci. Technol.* **2008**, *59*, 223-231.
71. Bill, K.; Bott, C.; Murthy, S. Evaluation of alternative electron donors for denitrifying moving bed biofilm reactor (MBBRs). *Water Sci. Technol.* **2009**, *60*, 2647-2657.
72. Di Trapani, D.; Mannina, G.; Torregrossa, M.; Viviani, G. Comparison between hybrid moving bed biofilm reactor and activated sludge system: A pilot plant experiment. *Water Sci. Technol.* **2010**, *61*, 891-902.
73. Tawfik, A.; El-Gohary, F.; Temmink, H. Treatment of domestic wastewater in an up-flow anaerobic sludge blanket reactor followed by moving bed biofilm reactor. *Bioprocess Biosyst. Eng.* **2010**, *33*, 267-276.
74. João, B.; Marcia, D.; Geraldo, L.S. Nitrification of industrial and domestic saline wastewaters in moving bed biofilm reactor and sequencing batch reactor. *J. Chem. Technol. Biotechnol.* **2008**, *83*, 6.

75. Yilmaz, G.; Lemaire, R.; Keller, J.; Yuan, Z. Simultaneous nitrification, denitrification and phosphorus removal from nutrient-rich industrial wastewater using granular sludge. *Biotechnol. Bioeng.* **2008**, *100*, 529-541.
76. Beliavski, M.; Meerovich, I.; Tarre, S.; Green, M. Biological denitrification of brines from membrane treatment process using an upflow sludge blanket (USB) reactor. *Water Sci. Technol.* **2010**, *61*, 911-917.
77. Timothy, J.; Ulrike, K.; Jürg, K. Using anoxygenic photosynthetic bacteria for the removal of sulfide from wastewater. *Adv. Photosynth. Respir*. **2008**, *27*, 437-460.
78. Li, W.; Zhao, Q.-L.; Liu, H. Sulfide removal by simultaneous autotrophic and heterotrophic desulfurization–denitrification process. *J. Hazard Mater*. **2008**, *162*, 848-853.
79. Pasukphun, N.; Vinitnantharat, S.; Gheewala, S. Investigation of decolorization of textile wastewater in an anaerobic/aerobic biological activated carbon system (A/A BAC). *Pak. J. Biol. Sci.* **2010**, *13*, 316-324.
80. Ong, S.; Toorisaka, E.; Hirata, M.; Hano, T. Combination of adsorption and biodegradation processes for textile effluent treatment using a granular activated carbon-biofilm configured packed column system. *J. Environ. Sci. (China)* **2008**, *20*, 952-956.
81. Liang, Z.; Das, A.; Beerman, D.; Hu, Z. Biomass characteristics of two types of submerged membrane bioreactors for nitrogen removal from wastewater. *Water Res*. **2010**, *44*, 3313-3320.
82. Modin, O.; Fukushi, K.; Nakajima, F.; Yamamoto, K. A membrane biofilm reactor achieves aerobic methane oxidation coupled to denitrification (AME-D) with high efficiency. *Water Sci. Technol.* **2008**, *58*, 83-87.
83. Feng, Y.; Tseng, S.; Hsia, T.; Ho, C.; Chou, W. Aerated membrane-attached biofilm reactor as an effective tool for partial nitrification in pretreatment of anaerobic ammonium oxidation (ANAMMOX) process. *J. Chem. Technol. Biotechnol.* **2008**, *83*, 6-11.
84. Zhang, Y.; Zhou, J.; Zhang, J.; Yuan, S. An innovative membrane bioreactor and packed-bed biofilm reactor combined system for shortcut nitrification-denitrification. *J. Environ. Sci.* **2009**, *21*, 568-574.
85. Hwang, J.H.; Cicek, N.; Oleszkiewicz, J.A. Achieving biofilm control in a membrane biofilm reactor removing total nitrogen. *Water Res*. **2010**, *44*, 2283-2289.
86. Kimura, K.; Nishisako, R.; Miyoshi, T.; Shimada, R.; Watanabe, Y. Baffled membrane bioreactor (BMBR) for efficient nutrient removal from municipal wastewater. *Water Res.* **2008**, *42*, 625-632.
87. Wu, Y.J.; Whang, L.M.; Huang, S.J.; Lei, C.N.; Cheng, S.S. Evaluation of performance and microbial ecology of sequencing batch reactor and membrane bioreactor treating thin film transistor liquid crystal display waste water. *Water Sci. Technol.* **2008**, *58*, 1085-1093.
88. Whang, L.; Yang, Y.; Huang, S.; Cheng, S. Microbial ecology and performance of nitrifying bacteria in an aerobic membrane bioreactor treating thin-film transistor liquid crystal display wastewater. *Water Sci. Technol.* **2008**, *58*, 2365-2371.
89. Syron, E.; Casey, E. Membrane-Aerated biofilms for high rate biotreatment: Performance appraisal, engineering principles, scale-up, and development requirements. *Environ. Sci. Technol.* **2008**, *42*, 1833-1844.
90. Li, T.; Liu, J.; Bai, R. Membrane aerated biofilm reactors: A brief current review. *Recent Pat. Biotechnol.* **2008**, *2*, 88-93.

91. Li, T.; Liu, J.; Bai, R.; Wong, F.S. Membrane-aerated biofilm reactor for the treatment of acetonitrile wastewater. *Environ. Sci. Technol.* **2008**, *42*, 2099-2104.
92. Sahu, A.K.; Sengupta, S.; Ergas, S.J. Onsite wastewater denitrification using a hydrogenotrophic hollow-fiber membrane bioreactor. *Water Environ. Res.* **2009**, *81*, 680-686.
93. Yang, S.; Yang, F.L.; Fu, Z. Characteristics of simultaneous nitrification and denitrification in moving bed membrane bioreactor. *Huan Jing Ke Xue* **2009**, *30*, 803-808.
94. Yang, S.; Yang, F.L.; Fu, Z.; Lei, R. Comparison between a moving bed membrane bioreactor and a conventional membrane bioreactor on membrane fouling. *Bioresour. Technol.* **2009**, *100*, 6655-6657.
95. Yang, S.; Yang, F.L.; Fu, Z.; Lei, R. Comparison between a moving bed membrane bioreactor and a conventional membrane bioreactor on organic carbon and nitrogen removal. *Bioresour. Technol.* **2009**, *100*, 2369-2374.
96. Chen, W.; Sun, F.; Wang, X.; Li, X. A membrane bioreactor for an innovative biological nitrogen removal process. *Water Sci. Technol.* **2010**, *61*, 671-676.
97. Hwang, J.H.; Cicek, N.; Oleszkiewicz, J. Long term operation of membrane biofilm reactors for nitrogen removal with autotrophic bacteria. *Water Sci. Technol.* **2009**, *60*, 2405-2412.
98. Hwang, J.H.; Cicek, N.; Oleszkiewicz, J. Achieving biofilm control in a membrane biofilm reactor removing total nitrogen. *Water Res.* **2010**, *44*, 2283-2291.
99. Celmer-Repin, D.; Hwang, J.; Cicek, N.; Oleszkiewicz, J. Autotrophic nitrogen-removing biofilms on porous and non-porous membranes. *Environ. Technol.* **2010**, *31*, 1391-1401.
100. Venkata, S.; Falkentoft, C.; Venkata, Y.; Sturm, B.S.; Wattiau, P.; Wilderer, P.A.; Wuertz, S.; Hausner, M. Bioaugmentation of microbial communities in laboratory and pilot scale sequencing batch biofilm reactors using the tol plasmid. *Bioresour. Technol.* **2009**, *100*, 1746-1753.
101. Peitzsch, M.; Kiesel, B.; Harms, H.; Maskow, T. Real time analysis of escherichia coli biofilms using calorimetry. *Chem. Eng. Process. Process Intensif.* **2008**, *47*, 1000-1006.
102. Shimokawa, T.; Hirai, M.; Shoda, M.; Sugano, Y. Hybrid culture efficient dye decolorization and production of dye decolorizing enzymes by the basidiomycete thanatephorus cucumeris Dec 1 in a liquid and solid hybrid culture. *J. Biosci. Bioeng.* **2008**, *106*, 481-487.
103. Woznica, A.; Nowak, A.; Karczewski, J.; Klis, C.; Bernas, T. Automatic biodetector of water toxicity (ABTOW) as a tool for examination of phenol and cyanide contaminated water. *Chemosphere* **2010**, *81*, 767-772.
104. Clouzot, L.; Marrot, B.; Doumenq, P.; Roche, N. 17-ethinylestradiol: An endocrine disrupter of great concern. *Environ. Prog.* **2008**, *27*, 383-396.
105. Mileva, A.; Sapundzhiev, T.; Beschkov, V. Modeling 1,2-Dichloroethane biodegradation by klebsiella oxytoca Va 8391 immobilized on granulated activated carbon. *Bioprocess Biosyst. Eng.* **2008**, *31*, 75-85.
106. Martínková, L.; Uhnáková, B.; Pátek, M.; Nesvera, J.; Kren, V. Biodegradation potential of the genus rhodococcus. *Environ. Int.* **2008**, *35*, 162-177.
107. Qiao, X.L.; Chen, Q.X.; Zhang, Z.J. Comparative study of nitrification performances of immobilized cell fluidized bed reactor and contact oxidation biofilm reactor in treating high strength ammonia wastewater. *J. Chem. Technol. Biotechnol.* **2008**, *83*, 84-90.

108. Paslawski, C.; Nemati, M.; Hill, A.; Headley, V. Model for biodegradation of a naphthenic acid in an immobilized cell reactor. *Can. J. Chem. Eng.* **2009**, *87*, 507-513.
109. Katuri, K.; Scott, K. Electricity generation from the treatment of wastewater with a hybrid up-flow microbial fuel cell. *Biotechnol. Bioeng.* **2010**, *107*, 52-58.
110. Venkata, M.S.; Veer, R.S.; Sarma, P.N. Influence of anodic biofilm growth on bioelectricity production in single chambered mediatorless microbial fuel cell using mixed anaerobic consortia. *Biosens. Bioelectron.* **2009**, *24*, 41-47.
111. Venkata, M.S.; Veer, R.S.; Sarma, P.N. Biochemical evaluation of bioelectricity production process from anaerobic wastewater treatment in a single chambered microbial fuel cell (MFC) employing glass wool membrane. *Biosens. Bioelectron.* **2008**, *23*, 1326-1332.
112. Venkata, M.S.; Veer, R.S.; Sarma, P.N. Bioelectricity production from wastewater treatment in dual chambered microbial fuel cell (MFC) using selectively enriched mixed microflora: Effect of catholyte. *Bioresour. Technol.* **2008**, *99*, 596-603.
113. Venkata, M.S.; Raghuvulu, S.; Peri, D.; Sarma, P. Integrated function of microbial fuel cell (mfc) as bio-electrochemical treatment system associated with bioelectricity generation under higher substrate load. *Biosens. Bioelectron.* **2009**, *24*, 2021-2027.
114. Mohanakrishna, G.; Venkata, M.; Sarma, P. Bio-Electrochemical treatment of distillery wastewater in microbial fuel cell facilitating decolorization and desalination along with power generation. *J. Hazard Mater.* **2010**, *177*, 487-494.
115. Gapes, D.; Keller, P. Impact of oxygen mass transfer on nitrification reactions in suspended carrier reactor biofilms. *Process Biochem.* **2008**, *44*, 43-53.
116. Kim, H.S.; Gellner, J.W.; Boltz, J.P.; Freudenberg, R.G.; Gunsch, C.K.; Schuler, A.J. Effects of integrated fixed film activated sludge media on activated sludge settling in biological nutrient removal systems. *Water Res.* **2010**, *44*, 1553-1561.
117. Sriwiriyarat, T.; Pittayakool, K.; Fongsatitkul, P.; Chinwetkitvanich, S. Stability and capacity enhancements of activated sluge process by IFAS technology. *J. Environ. Sci. Health A* **2008**, *43*, 1318-1324.
118. Sriwiriyarat, T.; Ungkurarate, W.; Fongsatitkul, P.; Chinwetkitvanich, S. Effects of dissolved oxygen on biological nitrogen removal in integrated fixed film activated sludge (IFAS) wastewater treatment process. *J. Environ. Sci. Health A* **2008**, *43*, 518-527.
119. Tian, Z.; Zeng, P.; Song, Y.; Li, D.; Zhang, J. Nitrogen Removal Potential and Biofilm Characteristics in the Anaerobic Ammonium Oxidation ('Anammox') Biofilter Reactor. In Proceedings of 4th International Conference on Bioinformatics and Biomedical Engineering, Chengdu, China, 18–20June 2010; Article number: 5514910.
120. Campos, V.L.; Escalante, G.; Yanez, J.; Zaror, C.A.; Mondaca, M.A. Isolation of arsenic-oxidizing bacteria from a natural biofilm associated to volcanic rocks of atacama desert, Chile. *J. Basic Microbiol.* **2009**, *49*, 93-97.
121. Gorbushina, A.; Broughton, W. Microbiology of the atmospheric-rock interface: How biological interactions and physical stresses modulate a sophisticated microbial ecosystem. *Annu. Rev. Microbiol.* **2009**, *63*, 431-450.
122. Newton, C.; Wilson, J. Recirculating gravel filters: High-performance treatment at low cost for two small communities. *Water Sci. Technol.* **2008**, *58*, 1245-1251.

123. Zhang, Z.; Rengel, Z.; Meney, K. Interactive effects of nitrogen and phosphorus loadings on nutrient removal from simulated wastewater using schoenoplectus validus in wetland microcosms. *Chemosphere* **2008**, *72*, 1823-1828.
124. Pollard, P. Bacterial activity in plant (schoenoplectus validus) biofilms of constructed wetlands. *Water Res.* **2010**, *44*, 5939-5948.
125. Iasur-Kruh, L.; Hadar, Y.; Milstein, D.; Gasith, A.; Minz, D. Microbial population and activity in wetland microcosms constructed for improving treated municipal wastewater. *Microb. Ecol.* **2010**, *59*, 700-709.
126. Yin, J.; Jiang, L.; Wen, Y.; Yao, Z.; Zhou, Q. Treatment of polluted landscape lake water and community analysis of ammonia-oxidizing bacteria in constructed wetland. *J Environ. Sci. Health A* **2009**, *44*, 722-731.
127. Tuszynska, A.; Obarska-Pempkowiak, H. Dependence between quality and removal effectiveness of organic matter in hybrid constructed wetlands. *Bioresour. Technol.* **2008**, *99*, 6010-6016.
128. Hijosa-Valsero, M.; Matamoros, V.; Sidrach-Cardona, R.; Martin-Villacorta, J. Becares, E.; Bayona, J. Comprehensive assessment of the design configuration of constructed wetlands for the removal of pharmacueticals and personal care products from urban wastewaters. *Water Res.* **2010**, *44*, 3669-3678.
129. Zhao, L.; Zhu, W.; Tong, W. Clogging processes caused by biofilm growth and organic particle accumulation in lab-scale vertical flow constructed wetlands. *J Environ. Sci. (China)* **2009**, *21*, 750-757.
130. Imfeld, G.; Braeckevelt, M.; Kuschk, P.; Richnow, H. Monitoring and assessing processes of organic chemicals removal in constructed wetlands. *Chemosphere* **2009**, *74*, 349-362.
131. Bonnineau, C.; Guasch, H.; Proia, L.; Ricart, M.; Geiszinger, A.; Romani, A.M.; Sabater, S. Fluvial biofilms: A pertinent tool to assess beta-blockers toxicity. *Aquat. Toxicol.* **2010**, *96*, 225-233.
132. Ricart, M.; Guasch, H.; Alberch, M.; Barceló, D.; Bonnineau, C.; Geiszinger, A.; Farré, M.; Ferrer, J.; Ricciardi, F.; Romani, A.M.; *et al.* Triclosan persistence through wastewater treatment plants and its potential toxic effects on river biolfilms. *Aquat. Toxicol.* **2010**, *100*, 346-353.
133. Writer, J.H.; Barbert, L.B.; Ryan, J.N.; Bradley, P.M. Biodegradation and attenuation of steroidal hormones and alkylphenols by stream biofilms and sediments. *Environ. Sci. Technol.* **2011**, *45*, 4370-4376.