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Chapter · January 2013

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Wastewater Treatment: Biological

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Abstract

Rapid urbanization and indiscriminate use of natural resources have placed the environment under increasing stress, and different measures are being implemented to prevent further deterioration. For example, treatment of our wastes and efficient reuse of our resources are prerequisites to further sustainable existence. As such, various waste treatment technologies have developed with the goal of minimizing negative impacts of wastes on the environment while also potentially recovering value from the wastes. Although many technologies exist, biological processes compare very favorably with non-biological processes because of their sustainability potential, including energy production and resource recovery. Further, carbon, nitrogen, and phosphorus are the main constituents of most wastes, and removal of such elements from waste effluents can reduce environmental stress and minimize ecosystem deterioration. This summary describes typical aerobic and anaerobic biological treatment methods, including activated sludge processes, upflow anaerobic sludge blanket reactors and other anaerobic systems, and biological nitrogen and phosphorus removal systems, which can be used to treat different types of wastes. An emphasis is placed on methods that also have the capacity to generate potential energy as combustible biogas or nutrients from wastes.

INTRODUCTION

Human activities and population growth have placed the environment under increasing stress. Furthermore, indiscriminate use of natural resources is accompanied by increased local and global pollution levels, which are reflected in imbalances in our ecosystems. The generation of large quantities of wastewater with a high organic content and toxicants is one obvious product of excessive consumption. It has been known for many years that environmental discharges of high loads of organic matter can result in oxygen depletion in receiving waters due to stimulated microbial activity. This oxygen depletion and the presence of trace toxicants found in wastes also negatively influence ecosystems, including reduced biodiversity and environmental health. Therefore, negative environmental impacts have driven our need to understand the effect of pollution on water bodies and develop proper measures to reduce discharges, including treatment processes.

Different technologies are available to treat wastes. However, biological wastewater treatment methods are most valuable because their economic benefits are high, especially when coupled with waste stabilization and resource recovery. The optimal treatment processes depend on the waste type and treatment goals. Wastewater generally originates from two sources: 1) domestic wastewater from gray water, toilets, and other domestic activities; and 2) industrial wastewater, generated by industries during the normal course of activity, which often rely on the local sewerage systems for waste processing. Therefore, the composition of wastewater, including quantity and constituents, varies considerably from place to place, depending on suite of sources, social behavior, the type and number of industries within a catchment, climatic conditions, water consumption, and the nature of the wastewater collection system. Given this variety, wastewater treatment processes must be innately versatile, but also sometimes must be tailored to the specific waste and conditions. The purpose of this entry is to describe different biological treatment methods and then discuss their relative capacities to treat different wastes on the basis of waste characteristics and the desire for resource recovery.

WASTEWATER TREATMENT OPTIONS

Special handling and treatment of wastes have been performed for thousands of years in response to their perceived importance, although approaches have changed as perceptions have changed over history. In 4th century B.C. in Greece, the *Athenian Constitution* written by Aristotle[1] proscribed provisions for the appropriate handling of sewage. Concern was based on aesthetics, probably odors, because relationships between domestic wastes and health were not yet known. It was not until the mid-1800s that links between wastes and human health became more apparent, which led to a progression of waste management approaches and technologies to address health concerns.

Treatment technologies evolved slowly over time, including physical, chemical, and biological approaches,

many of which are still used in different sectors. Physical methods are based on the application of physical forces, such as screening, mixing, flocculation, sedimentation, flotation, filtration, and gas transfer. Alternately, chemical processes treat contaminants by adding chemicals or by stimulating specific chemical reactions. Precipitation, adsorption, and disinfection are common examples of chemical treatment methods. Physical and chemical methods are often combined, especially in industrial treatment scenarios. In contrast to physiochemical processes, biological processes remove organic contaminants (e.g., biodegradable organic material) largely through microbiological activity. Commonly used biological treatment methods include aerobic treatment in ponds, lagoons, trickling filters, and activated sludge plants,[2] and anaerobic treatment[3,4] in similar reactor systems. Processes that combine anaerobic and aerobic unit operations are also common.[5]

The best overall treatment approach depends on the source and nature of waste, such as production rates, constituents, and relative concentrations. As such, optimal process trains and designs should be as simple as possible in design and operation, while being efficient in removing key pollutants and minimizing energy consumption and negative by-products. More complex operations are only used when absolutely necessary.

Within a typical treatment plant, each type of treatment has a different purpose. For example, the main objective of biological treatment is to treat soluble organic matter in the wastes, which often requires physical pretreatment to remove solids before biological treatment.[2] For domestic wastewater, the main objective is to reduce the organic content and, in growing numbers of cases, secondary nutrients (nitrogen, N; phosphorus, P). For industrial wastewaters, the objective is usually to remove or reduce the concentration of organic compounds, especially specific toxicants that can be present in some wastes, which is why chemical processes are also included in industrial treatment systems. However, biological processes are almost always used when possible.

Biological degradation of organics is accomplished through the combined activity of microorganisms, including bacteria, fungi, algae, protozoa, and rotifers. To maintain the ecological balance in the receiving water, regulatory authorities have set standards for the maximum amount of the undesirable compounds present in the discharge water. In a typical wastewater treatment plant, the following steps are carried out to achieve the desired quality of the effluent before it can be safely discharged into the receiving water.

Pretreatment/Preliminary Treatment

Pretreatment is primarily used to protect pumping equipment and promote the success of subsequent treatment steps. Pretreatment devices such as screen and/or grit removal systems are designed and implemented to remove the larger suspended or floating solids, or heavy matter that

can damage pumps. Sometimes, froth flotation is also used to remove excessive oils or grease in the wastes.

Primary Treatment

Most of the settleable solids are removed from the wastewater by simple sedimentation, a purely physical process. In this process, the horizontal velocity of the water through the settle is maintained at a level that provides solids adequate time to settle and floatable material be removed from the surface. Therefore, primary treatment steps consist of settling tanks, clarifiers, or flotation tanks, which send separated solids to digestion units and supernatant to subsequent, typically microbiological, treatment units.

Secondary Treatment

Secondary treatment uses microbial communities, under varying growth conditions, to biochemically decompose organic compounds in the waste that have passed from primary treatment units. An array of reactors are employed for biological treatment, which include suspended biomass, biofilm, fixed-film reactors, and pond or lagoon systems.

Secondary Clarification

Most biological treatment processes produce excess biomass through the conversion of waste carbon to new cells. As such, before the final treatment steps, such as disinfection or nutrient removal, solids must be separated from the secondary treatment effluents. This is usually by settling, but membranes are also employed. The separated solids are either recycled back to the head of the process train or sent to digesters for solids reduction and processing, depending on the type of the digester system.

Tertiary/Advanced Treatment

Advanced or tertiary treatment consists of processes that are designed to achieve higher effluent quality than attainable by conventional secondary treatment methods. These include polishing steps such as activated carbon adsorption, ion exchange, reverse osmosis, electrodialysis, chemical oxidation, and nutrient removal. Although not technically a tertiary process, final effluent disinfection is often performed after secondary or tertiary treatment using chlorination, ultraviolet methods, ozonation, and other methods designed specifically to kill residual organisms in the wastewater after all previous treatment steps.

BIOLOGICAL TREATMENT OPTIONS

Biological processes are classified according to the primary metabolic pathways present in the dominant different microorganisms active in the treatment system. As per the availability and utilization of oxygen, the biological processes are classified as aerobic, anoxic, and anaerobic.

Aerobic Processes

Treatment processes that occur in the presence of molecular oxygen (O_2) and use aerobic respiration to generate cellular energy are called aerobic processes. They are most metabolically active, but also generate more residual solids as cell mass.

Anoxic Processes

These are processes that occur in the absence of free molecular oxygen (O_2) and generate energy through anaerobic respiration. Microorganisms use combined oxygen from inorganic material in the waste (e.g., nitrate) as their terminal electron acceptor. Anoxic processes are common biological nitrogen removal systems through denitrification.[2]

Anaerobic Processes

These are the processes that occur in the absence of free or combined oxygen, and result in sulfate reduction and methanogenesis. They usually produce biogas (i.e., methane) as a useful by-product and tend to generate lower amounts of biosolids through treatment.

Apart from a classification based on microbial metabolism and/or oxygen utilization, biological wastewater treatment processes also can be classified based on the growth conditions in the reactor (see Fig. 1). In this case, the two main categories are suspended growth and attached growth processes.

Suspended Growth Processes

In these processes, the microorganisms, which are responsible for the conversion of waste organic matter to simpler compounds and biomass, are maintained in suspension within the liquid phase. However, there are different types of aerobic and anaerobic suspended growth processes. Aerobic processes include activated sludge, aerated lagoons, and sequencing batch reactors, whereas anaerobic processes include bag digesters, plug-flow digesters, stirred-tank reactors, and baffled reactors with organisms primarily in the liquid phase.

Attached Growth Process

In these processes, the microorganisms responsible for degrading the waste are attached to surfaces (e.g., stones, inert packing materials), or are self-immobilized on flocs or granules in the system. Attached growth processes can be aerobic or anaerobic. Aerobic attached growth processes include trickling tilters, roughing filters, rotating biological contactors, and packed-bed reactors. Anaerobic systems
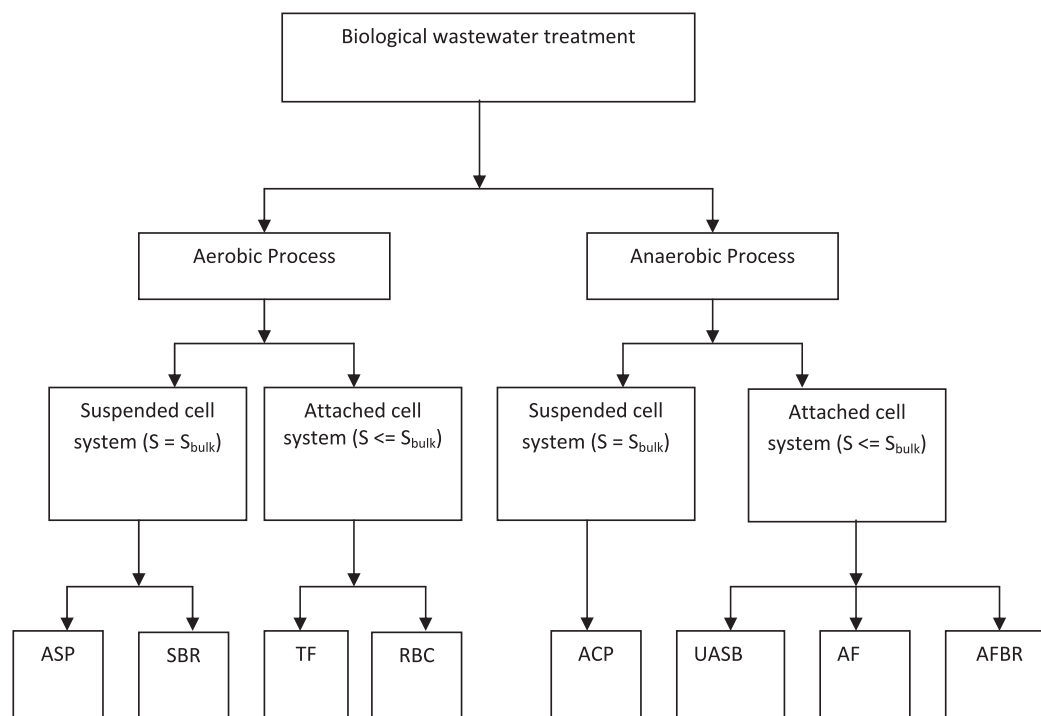
Fig. 1 Different biological treatment processes. S, substrate concentration available to microorganisms; S_{bulk}, substrate concentration in the bulk of the liquid; ASP, activated sludge process; SBR, sequencing batch reactor; TF, trickling filter; RBC, rotating biological contactor; ACP, anaerobic contact process; AF, anaerobic filter; UASB, upflow anaerobic sludge blanket; AFBR, anaerobic fluidized bed reactor.

include upflow packed-bed reactors, down-flow packed-bed reactors, anaerobic rotating biological contactors, anaerobic fluidized bed reactors, upflow anaerobic sludge blanket (UASB) reactors, and various hybrid anaerobic reactors (HAR). UASBs are widely used reactors for the anaerobic treatment of industrial and domestic wastewater.

AEROBIC BIOLOGICAL WASTE TREATMENT PROCESSES

Typical aerobic waste treatment systems provide a location where microbes are exposed to molecular oxygen (O_2) to oxidize complex organics present in the waste, producing carbon dioxide, simple organics, and new cell biomass. The activated sludge process (ASP) is very well known and the most widely used biological treatment process in developed countries.

Activated Sludge Process

Classic ASPs are aerobic suspended cell systems. Mineralization of waste organic compounds is accompanied by the formation of new microbial biomass and sometimes the removal of inorganic compounds, such as ammonia and phosphorus, depending on the particular process design. Activated sludge processes were first conceived in the early 1900s with the word "activated" referring to solids that catalyze the degradation of the waste. It was subsequently discovered that the "activation" part of the sludge was a complex mixture of microorganisms. The liquid in activated sludge systems is called the "mixed liquor," which includes both wastewater and the resident organisms.

There have been several incarnations of the ASP. The most common designs use conventional, step aeration, and continuous-flow stirred-tank reactors.[2] A conventional ASP consists of standard pretreatment steps, an aeration tank, and a secondary clarifier, an example of which is shown in Fig. 2. The aeration tank can be aerated by sub-

surface or surface aerators designed to supply adequate dissolved oxygen to the water for the microorganisms to thrive. The wastewater flows through the tank and resident microorganisms consume organic matter in the wastewater. The aeration tank effluent flows to the clarifier where the microorganisms are removed. The clarifier supernatant is then transferred to disinfection or treatment units, and then ultimately discharged to the receiving water. Biosolids from the settler are recycled back to the head of the treatment system or sent to digesters for further processing.

Aeration Tanks

Aeration tanks are usually designed uncovered, open to the atmosphere. Air is supplied to the microorganisms by two primary methods: mechanical aerators or diffusers. Mechanical aerators, such as surface aerators and brush aerators, aerate the surface of the water mechanically and promote diffusion of oxygen to water from the atmosphere. The concentration of dissolved oxygen in the liquid can be controlled by adjusting the speed of the rotors. Both mechanical aerators and diffusers are the largest energy consumers in aerobic biological wastewater treatment processes. Diffusers bubble air directly into the tank at depth and are usually preferred because of higher oxygen transfer efficiencies.

As previously indicated, aeration provides O_2 to the microorganisms and also serves to mix the liquor in the tank. Although complete mixing is desired, there are usually "dead zones" in the tank where anaerobic/anoxic conditions develop in poorly mixed areas. It is desirable to keep these zones to a minimum to minimize undesired odors and also problems with sludge bulking, which can reduce settling efficiency in secondary clarifiers.

Secondary Clarifiers

Clarifiers are used to separate the biomass and other solids coming out of the aeration tank by means of gravity set-
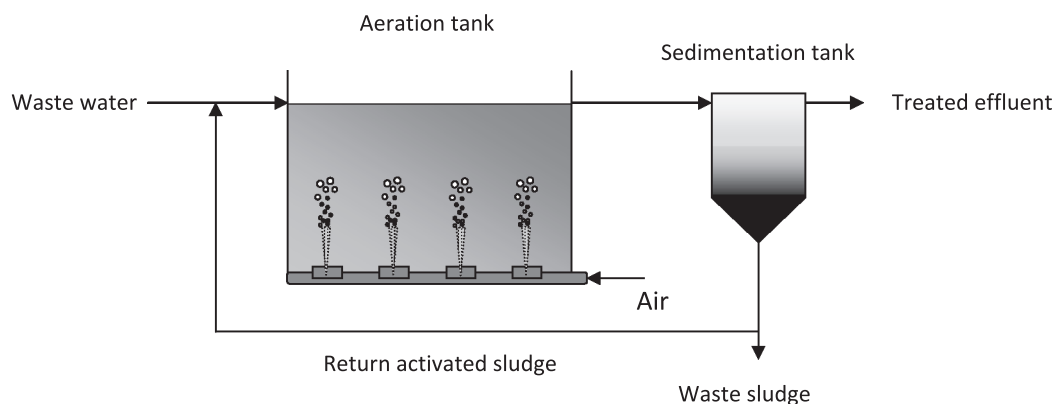

Fig. 2   Activated sludge process.

tling. The flow rate of the liquid is maintained in such a way that the upflow velocity of the liquid is less than the settling velocity of the biosolids present in the liquid. As noted, some of the settled biosolids are returned back to the aeration tank to increase the solids' contact time with the wastes and also maintain the desired biomass levels in the aeration tank.

Important Operating Parameters in Activated Sludge Systems

Key operating parameters and typical values for activated sludge systems are provided in Table 1. All parameters ultimately are used to guide and pseudo-control biosolids levels, and they profoundly affect process performance. The total suspended solids in the aeration tank are known as mixed-liquor suspended solids (MLSS). This term refers to the amount of solids in a certain volume of the water (usually milligram of solids per liter). The actual biomass fraction of the solids is estimated as the solids that can be volatilized at 550°C. The volatile fraction is known as mixed-liquor volatile suspended solids (MLVSS). Therefore, MLVSS is frequently used as a proxy for the active biomass treating the waste. MLVSS ranges from about 70% to 90% of the MLSS concentration in most activated sludge systems.[6]

Solid Retention Time

The most important design parameter in activated sludge systems is the mean cell residence time of cells in the reactor, also known as the sludge age or solid retention time (SRT). The SRT can be controlled by manipulating the rate at which excess sludge is wasted and is influenced by hydraulic flow conditions through the reactor. It is the ratio of the total solids in the system and the total solids leaving the system.

Table 1 Typical design parameters for ASP.

| Process components or variables	Typical values	Reference
Aeration tank		[2]
Depth (m)	5–8	
Width (m)	7–12	
SRT (day)	5–15	[2]
MLSS (kg/m^3)	1500–4000	[9]
SVI (kg/m^3)	40–150	[7]
F/M	0.2–0.4	[6]
Organic loading rate (kg COD/m^{3}day)	20–60	[6]
Oxygen requirement (kg/kg COD removed)	1.4–1.6	[9]

$$SRT = VX/(QX_e + Q_wX_w)$$

where SRT is the mean cell residence time (day); V is the volume of aeration basin (e.g., L); X is the mixed liquor suspended solids concentration (mg/L); Q is the volumetric flow rate (e.g., L/day); X_e is the effluent suspended solids concentration (mg/L); Q_w is the waste sludge flow rate (e.g., L/day); and X_w is the waste sludge suspended solids concentration (mg/L).

Sludge Volume Index

The sludge volume index (SVI) is another key parameter and used to describe the settling characteristics of the sludge. The SVI is expressed as the volume occupied by 1 g of sludge (mL/g) after 30 min of settling time. Well-settled sludge normally yields a clear separation between the water and the sludge. However, if the sludge has any problems, such as bulking, pinpoint floc formation of tiny, poorly settling floc, or ashing, the interface between the sludge and the water may not be seen clearly. Such conditions usually result from problems in the aeration tank and cause reduced effluent quality because of poor settling in the clarifier.

Dissolved Oxygen Concentration

Microorganisms in an activated sludge system require adequate oxygen to oxidize organics in the waste. The basic oxidation reaction for organics degradation can be approximated as (stoichiometry not provided)

$$CHON + O_2 + \text{microorganisms} \rightarrow$$
$$CO_2 + H_2O + NH_3 + \text{more microorganisms}$$
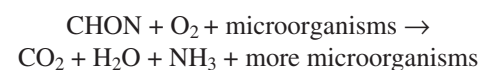
Organics are consumed by microorganisms, and new microbial cells are synthesized with ratio of organisms produced relative to the organics consumed being the sludge yield. As noted, oxygen is supplied by mechanical aerators or diffusers in the aeration tank. Required oxygen levels in the system depend on the process, but the design goal is to minimize oxygen addition due to energy costs. The dissolved oxygen concentration can be controlled by either adjusting the speed of the air pump or throttling the air pipes. Air pumps are more widely used to aerate the wastewater because of their lower operational and maintenance costs.

Food-to-Microorganism Ratio

The food-to-microorganism ratio (F/M) is a good indicator for designing and regulating the operation of the aeration tank.[7] The F/M ratio is expressed as the amount of organic biodegradable material [milligrams of 5-day biological oxygen demand (BOD$_5$)] available

for the amount of microorganisms present (mg MLVSS) per day.

$$F/M = (QS_o)/X$$

where F/M is the food-to-microorganism ratio (day^{-1}); S_o is the influent BOD_5 concentration (mg/L); X is the MLVSS concentration (mg/L); and Q is the volumetric flow rate (L/day).

The targeted F/M ratio for any treatment system varies depending on the design of the system, and values can range widely. However, since influent BOD cannot be controlled, MLVSS is typically modulated by varying the return activated sludge rate from the secondary clarifier, the goal being to maintain an optimum F/M ratio for specific activated sludge design.

Organic Loading Rate

The amount of organic matter in wastewater is commonly measured by BOD_5, chemical oxygen demand (COD), or the total organic carbon content.[8,9] If there are excess organics in the influent or inadequate organisms in the aeration tank, incomplete treatment will result.

Common Microorganisms in Activated Sludge Systems

Activated sludge is a complex mixture of broadly differing microorganisms.[10] Major categories are as follows: bacteria, fungi, algae, protozoa (e.g., flagellates, ciliates, and rotifers), and viruses. Viruses and pathogenic bacteria are often present in wastewater, which is the primary reason for having post-biological disinfection steps in treatment plants.

ATTACHED GROWTH PROCESSES

Attached growth processes, such as trickling filters (Fig. 3), can achieve similar treatment objectives as activated sludge systems. Conversion processes in these systems are typically mass transport limited: microorganisms in the outer layers of the biofilm contribute most to the overall substrate removal. The support material in trickling filters is chosen to provide sufficiently large pore spaces to allow air through the trickling filter regardless of biofilm growth and water trickling down the filter. Wastewater is distributed using rotary arms at the top and then trickles down the filter. Trickling filters are mainly used for the oxidation of carbon and ammonia, but can also achieve
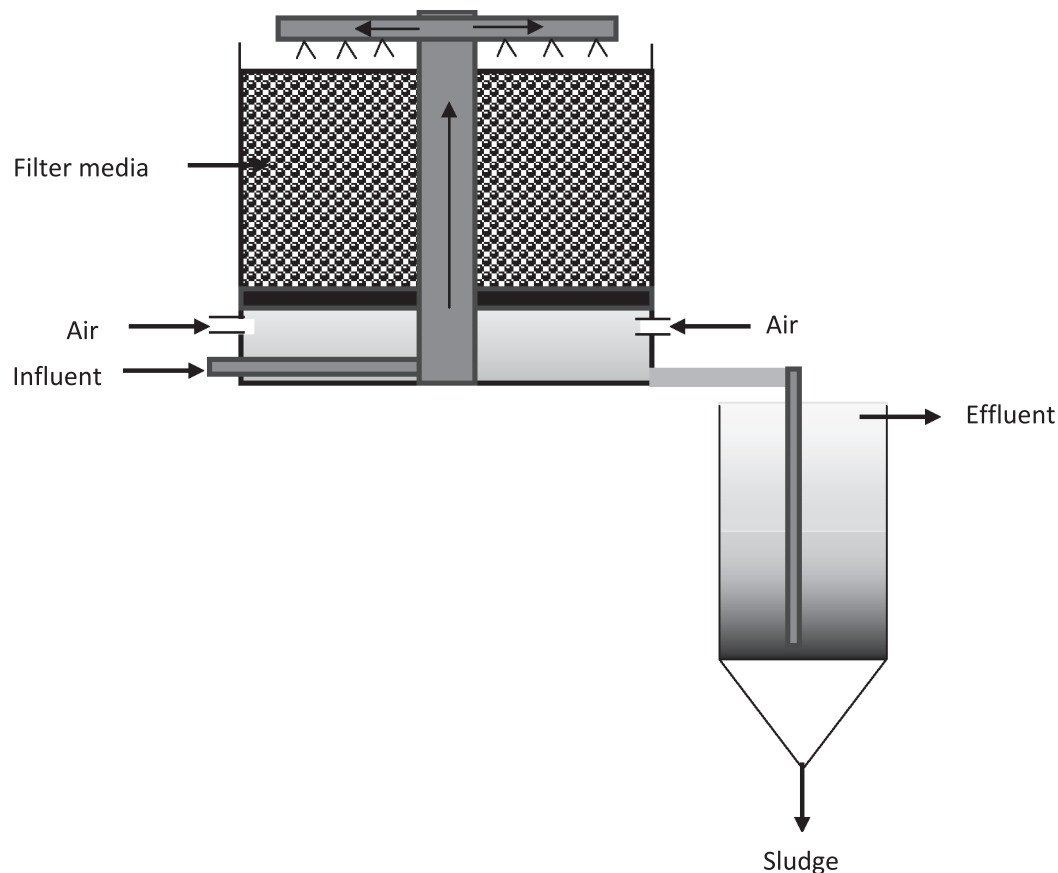
Fig. 3 Aerobic trickling filter.

denitrification when convection of air through the system is optimized.[11]

ANAEROBIC WASTEWATER TREATMENT PROCESSES

Anaerobic treatment technologies are widely practiced in different industries on the basis of their requirement and suitability. The processes have some advantages and disadvantages in treating different wastes, and few of them are summarized in Table 2. Under anaerobic conditions, organic matter is degraded through the sequential and syntrophic metabolic interactions of various trophic groups of prokaryotes, including fermenters, acetogens, methanogens, and sulfate-reducing bacteria (SRB).[12,13] Metabolic interactions between these microbial groups lead to the transformation of complex organic compounds to simple compounds such as methane, carbon dioxide, hydrogen sulfide, and ammonia.[14] The digestion process is essentially accomplished in four major reaction stages involving different microorganisms in each stage.[15,16]

Stage 1: Hydrolysis—The organic waste material mainly consists of carbohydrates, proteins, and lipids. Complex and large substances are broken down into simpler compounds by the activity of the microbes and the extracellular enzymes released by these microbes. The hydrolysis or solubilization is mainly done by hydrolytic microbes such as *Bacteroides*, *Bifidobacterium*, *Clostridium*, and *Lactobacillus*. These organisms hydrolyze complex organic molecules (cellulose, lignin, proteins, lipids) into soluble monomers such as amino acids, glucose, fatty acids, and glycerol. These hydrolysis products are used by the fermentative acidogenic bacteria in the next stage.[14,17]

Stage 2: Acidogenesis—Fermentative acidogenic bacteria convert simple organic materials such as sugars, amino acids, and long-chain fatty acids into short-chain organic acids such as formic, acetic, propionic, butyric, valeric, isobutyric, isovaleric, lactic, and succinic acids; alcohols and ketones (ethanol, methanol, glycerol, and acetone); carbon dioxide; and hydrogen. Generally, acidogenic bacteria have high growth rates and are the most abundant bacteria in any anaerobic digester.[18] The high activity of these organisms implies that acidogenesis is never the rate-limiting step in the anaerobic digestion process.[19] The volatile acids produced in this stage are further processed by microorganisms characteristic for the acetogenesis stage.

Stage 3: Acetogenesis—In this stage, acetogenic bacteria, also known as obligate hydrogen-producing acetogens, convert organic acids and alcohols into acetate, hydrogen, and carbon dioxide, which are subsequently used by methanogens and SRB. There is a strong symbiotic relationship between acetogenic bacteria and methanogens. Methanogens and SRB use hydrogen, which helps achieve the low hydrogen pressure conditions required for acetogenic conversions.[20]

Stage 4: Methanogenesis—It is the final stage of anaerobic digestion where methanogenic archaea convert the acetate, methanol, methylamines, formate, and hydrogen produced in the earlier stages into methane. The growth rate of methanogens is very low, and therefore, in most cases, this step is considered as the rate-limiting step of the anaerobic process, although there are also examples where hydrolysis is rate limiting.[21]

UASB Reactors

The most common and widely used anaerobic reactor is the UASB reactor.[22] It is an attached, self-immobilized cell system, which consists of a bottom layer of packed sludge bed (sludge blanket) and an upper liquid layer, as shown in Fig. 4.[23]

Wastewater flows upward through a sludge bed consisting of bacterial aggregates floating blanket, and the

Table 2 Advantages and disadvantages of anaerobic wastewater treatment.

Advantages
High efficiency: Good removal efficiency can be achieved in the system, even at high loading rates and low temperature.
Simplicity: The construction and operation of these reactors are relatively simple.
Flexibility: Anaerobic treatment can easily be applied on either a very large or a very small scale.
Low energy consumption: As far as no heating of the influent is needed to reach the working temperature and all plant operations can be done by gravity, the energy consumption of the reactor is almost negligible.
Energy recovery: Energy is produced during the process in the form of methane.
Low sludge production: Sludge production is low, well stabilized, and has good dewatering property.
Low nutrient and chemical requirement: Especially in the case of sewage, an adequate and stable pH can be maintained without addition of chemicals.

Disadvantages
Low pathogen and nutrient removal: Pathogens and nutrients are partially removed and hence post-treatment is needed.
Long start-up: Due to low growth rate of methanogenic organisms, the start-up takes longer time.
Possible bad odor: Hydrogen sulfide is produced. Proper handling of biogas is required to avoid bad smell.
Necessity of post-treatment: Post-treatment of the anaerobic effluent is generally required to reach the discharge standards for organic matter and pathogen.

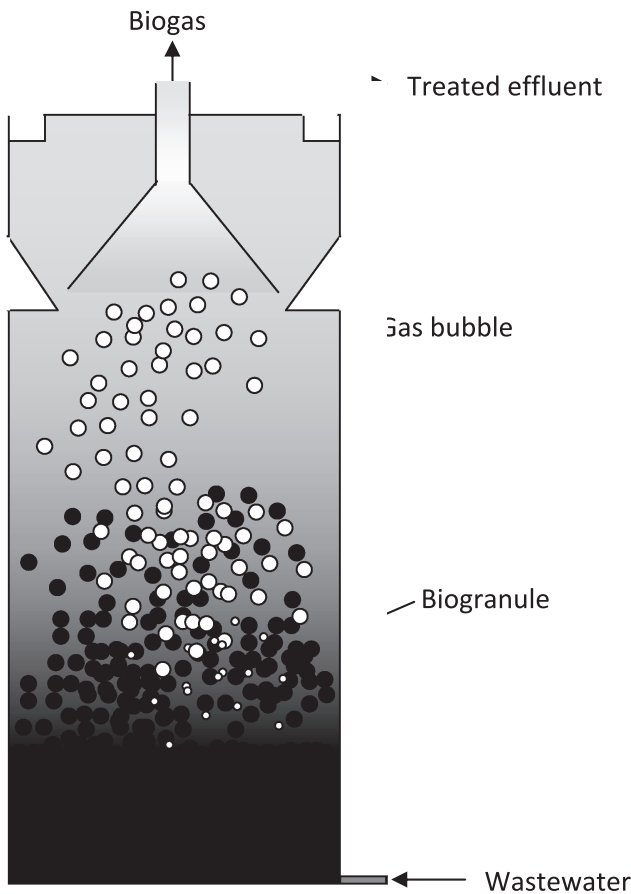**Source:** Data from Seghezzo et al.[22]

Fig. 4 Upflow anaerobic sludge blanket reactor.

microbes present in the sludge bed convert the complex organic materials to methane, carbon dioxide, and hydrogen.[24] The granular sludge (1–5 mm in diameter) has high biomass content (MLVSS) and specific activity, and good settling properties. The upward flow of the liquid inside the reactor is obtained by means of effluent recirculation. Because of the high density of biomass present in the self-immobilized granular sludge, the reactor is able to support a high SRT, which is diverse from the hydraulic retention time (HRT) and require no support material. The major drawback of the UASB is the requirement of high HRT to achieve desired biodegradation. Maintenance of high HRT demands huge reactor volume. These problems are overcome by using HAR where the advantages of AFBR are coupled with UASB operation by maintaining a high upflow velocity (4–8 m/hr) inside the reactor.[25] With higher upflow velocity, better mass transfer is obtained in the reactor, which reflects on the higher degradation with less HRT operation. The main purpose of these reactors is to achieve better degradation of waste and increase the production of biogas (methane) in a substantially reduced-size anaerobic reactor.

Important Operating Parameters in Anaerobic Reactors

Different operating parameters such as pH, temperature, HRT, and nutrients, among others, and their disturbances can manifest in case of industrial wastewaters treatment in anaerobic reactors, even under normal operational conditions.[26,27] Some of these factors are discussed below.

pH

The optimum degradation is achieved when the pH value of wastewater in the digester is maintained between 6.5 and 7.5. In the initial period of fermentation, as large quantities of organic acids are produced by acidogens and acetogens, a drop in pH occurs inside the digester. This low pH condition inhibits methanogens and subsequently reduces methane production. As the digestion proceed, the pH increases owing to the conversion of organic nitrogen to NH_4. When the methane production level is stabilized, the pH range remains buffered between 7.2 and 7.8.[28,29]

Waste Composition

To attain optimum degradation, wastewaters have to be nutritionally balanced in terms of carbon (C), nitrogen (N), phosphorous (P), and sulfur (S). The C/N/P ratio of 700:5:1 is recommended for efficient anaerobic digestion.[30] A fairly high concentration of acetate is required to prevent SRB outcompeting methanogens for acetate and hydrogen.[31]

Temperature

Methanogens are inactive at extremely high and low temperatures.[32] Few psychrophilic methanogens have been discovered, which can grow at a temperature range of 4–6°C.[33] Most of the methanogens can grow well from 25°C to 65°C temperatures.[34] The optimum temperature for the growth of the mesophilic methanogens is 35–37°C.[34] When the ambient temperature goes down to 10°C, gas production virtually stops. Satisfactory gas production takes place in the mesophilic range, from 30°C to 40°C.

Loading Rate

High organic loading rate may lead to acid accumulation and reduction of methane production. Similarly, if the plant is underfed, the gas production will also be low.[15]

Retention Time

The retention time depends on the growth rate of the microbial population and reactor configuration (attached

cell or suspended cell system), waste strength, and waste composition.

Toxicity

The presence of toxicants in the wastewater, such as oxygen (lethal to obligate anaerobes), ammonia, chlorinated hydrocarbons, aromatic hydrocarbons, heavy metals, and long-chain fatty acids, among several others, may also result in occasional failures of anaerobic digesters.[15] The presence of trace amount of metals (e.g., nickel, cobalt, molybdenum) also stimulates the growth of microbes. Excess volatile fatty acid (VFA) concentrations are reported to inhibit the growth of several microbial species.[35] The undissociated forms of VFA can diffuse across the cell membrane and dissociate intracellularly, which results in reduction in growth rate.[35,36] The 50% inhibition of acetoclastic methanogenesis in granular sludge was observed at a concentration of 13,000, 3,500, and 15,000 mg/L of acetate, propionate, and butyrate, respectively.[37] Small amounts of sulfide, a vital sulfur source, are beneficial for methanogens.[38] Acetoclastic methanogens are the most sensitive in terms of sulfide inhibition. Fifty percent inhibition was observed at total sulfide concentrations of 220–980 mg/L over the pH range 6.5–8.0.[39]

Granule Deterioration

Lipids present in the wastewater creates problem by forming long-chain fatty acids during hydrolysis in the anaerobic reactor. Long-chain fatty acid imparts toxic effect to acetogenic and methanogenic microbes. It also becomes adsorbed onto the sludge, inducing sludge flotation and resulting in washout.[40] Some long-chain fatty acids also act as surfactant at neutral pH and obstruct the floc formation by lowering the surface tension between water and the hydrophobic bacteria and promote their washout.[41] Addition of polyelectrolytes (calcium salts) may prevent inhibition to some extent, but it does not prevent flotation.[42]

BIOLOGICAL REMOVAL OF NITROGEN

The conventional biological nitrogen removal is a two-step process, nitrification followed by denitrification. The process is slow due to low microbial activity and yield. Nitrification involves a chemolithoautotrophic oxidation of ammonia to nitrate under strict aerobic conditions. This oxidation is a result of two sequential oxidative stages: ammonia to nitrite (ammonia oxidation) and nitrite to nitrate (nitrite oxidation). Different microorganisms involved in these stages use molecular oxygen as an electron acceptor and carbon dioxide as carbon source. The oxidation of ammonia to nitrite is performed by nitrifier microorganisms such as *Nitrosomonas*, *Nitrosococcus*, *Nitrosopira*,

Nitrosovibrio, and *Nitrosolobus.* In the nitrite oxidation stage, *Nitrobacter*, *Nitrospira*, *Nitrospina*, *Nitrococcus*, and *Nitrocystis* are known to be involved in the production of nitrate.[10,43] Ammonia uptake rate varies according to reactor configuration, substrate type, and influent ammonium concentration. Denitrification is the second stage of the nitrogen removal process. It is a heterotrophic bioconversion process carried out by the heterotrophic denitrifiers under anoxic conditions. The oxidized nitrogen compounds (NO_2^- and NO_3^-) are reduced to nitrogen gas by the denitrifiers that use nitrite and/or nitrate as terminal electron acceptors and organic matter as carbon and energy source. *Pseudomonas*, *Alcaligenes*, *Paracoccus*, *Thiobacillus*, and *Halobacterium* are commonly found in dentrification systems.[44]

Few advanced processes, including partial nitrification, anaerobic ammonium oxidation (Anammox) and autotrophic nitrogen removal (Canon) are also being practiced in different treatment plants according to the characteristics of the wastewater. A combined system of partial nitrification and Anammox is advantageous as no extra carbon addition is needed, a negligible amount of sludge is produced, and less energy and oxygen are required compared with the conventional two-stage process.[45]

Sharon Process

The Sharon (single-reactor high-activity ammonium removal over nitrite) process is used for removal of ammonia through nitrite formation.[45,46] In this process, both autotrophic nitrification and heterotrophic denitrification take place in a single reactor with intermittent aeration. The denitrification in the Sharon process is achieved by adding methanol as a carbon source. Although the process is not suitable for all wastewaters due to a high temperature dependency, the Sharon process is suitable for removing nitrogen from waste streams having high ammonia concentrations (>0.5 g/L).

Anaerobic Ammonium Oxidation

Anaerobic ammonium oxidation (Anammox) is a highly exergonic, lithoautotrophic biological conversion process where ammonia becomes converted to nitrogen by the activity of a group of planctomycete bacteria.[47] These microorganisms use CO_2 as the sole carbon source and have a capability to oxidize ammonia to gaseous nitrogen by using nitrite as the electron acceptor in an anoxic condition.

Combined Nitrogen Removal

Ammonia-rich wastewater can be treated by Anammox, which requires nitrite as precursor. Thus, before feeding into the Anammox process, ammonia has to be preoxidized

to nitrite. Thus, a partial Sharon process can be used before the Anammox process to improve the nitrogen removal efficiency. Partial nitritation (conversion of 55%–60% of ammonium to nitrite) is achieved in the Sharon process without heterotrophic denitrification. Nitrite-rich waste is then treated in an Anammox reactor. In the partial Sharon–Anammox digester, overall 83% ammoniacal nitrogen removal can be obtained from the waste stream has a total nitrogen load of 0.8 kg N/m^3/day.[48]

Canon Process

The Canon (completely autotrophic nitrogen removal over nitrite) process is also the combination of partial nitritation and Anammox processes. In this process, two groups of aerobic and anaerobic microorganisms (e.g., *Nitrosomonas* and planctomycetes) perform two sequential reactions in a single and aerated reactor. The nitrifiers consume oxygen and oxidize ammonia to nitrite. Consumption of oxygen creates an anoxic condition the Anammox process needed. The performance of the Canon process is very much dependent on operational parameters such as dissolved oxygen, biofilm thickness, nitrogen-surface load, and temperature.[49]

BIOLOGICAL PHOSPHORUS REMOVAL

The removal of phosphorus from the wastewater by the biological means is known as biological removal of phosphorus. The groups of microorganisms that are largely responsible for phosphorus removal are known as the polyphosphate-accumulating organisms (PAOs). These organisms are able to store phosphate as intracellular polyphosphate, leading to phosphorus removal from the bulk liquid phase through PAO cell removal in the waste activated sludge. Enhanced biological phosphorus removal can be achieved through the ASP by recirculating sludge through anaerobic and aerobic conditions.[50] Unlike most other microorganisms, PAOs can take up carbon sources such as VFAs under anaerobic conditions, and store them intracellularly as carbon polymers, namely poly-β-hydroxyalkanoates (PHAs). The energy for this biotransformation is mainly generated by the cleavage of polyphosphate and release of phosphate from the cell. Reducing power is also required for PHA formation, which is produced largely through the glycolysis of internally stored glycogen.[51] The principal advantages of biological phosphorous removal are reduced chemical costs and less sludge production as compared with chemical precipitation.

The different types of bacteria used in biological phosphorus removal are *Acinetobacter*, *Pseudomonas*, *Microlunatus phosphovorus*, *Aeromonas*, and *Lampropedia. Acinetobacter calcoaceticus* has a very high capacity to intracellularly accumulate polyphosphate from various activated sludges. It can accumulate phosphate of an amount of 0.9%–1.9% of dry cell weight.

CONCLUSION

Biological treatment processes have a proven track record of dealing adequately with various kinds of wastes generated by human activities. They mimic natural processes occurring in streams and rivers. Waste treatment processes are increasingly engineered in such a way that they perform this task efficiently with a minimal input of energy. Traditionally, treatment has relied on technological approaches designed to mimic aerobic processes occurring in the water column of streams and rivers. To become truly sustainable, however, we must move away from energy-consuming aerobic processes and switch to anaerobic treatment processes, again mimicking natural processes, but now those occurring in the anaerobic sediments of the aforementioned streams and rivers. For example, there is a new focus in the water industry to integrate these two processes into systems where the waste is initially digested in an anaerobic step followed by an aerobic polishing step. Only by integrating these two processes, and variants thereof such as partial nitrification and Anammox wastewater treatment, will waste treatment become truly energy efficient and sustainable. Finally, it should be noted that anaerobic digestion to methane is not the only sustainable option. Great strides are now being made in microbial fuel cell technology within waste treatment with chemical energy from wastes being captured as electricity. All told, we are finally beginning to see again that wastes are not problems to be solved but are valuable resources, and new technologies continue to be developed to capture this capacity.

REFERENCES

1. Van de Kraats, J. Editorial. Eur. Water Pollut. Control **1997**, *7*, 3–4.
2. Tchobanoglous, G.; Burton, F.L.; Stensel, H.D.; Metcalf and Eddy, Inc. Fundamentals of biological treatment. In *Wastewater Engineering: Treatment, Disposal, Reuse*, 6th Ed.; McGraw-Hill: New York, 2003.
3. Lettinga, G. Anaerobic digestion and wastewater treatment systems. Antonie van Leeuwenhoek **1995**, *67*, 3–28.
4. Lettinga, G. Sustainable integrated biological wastewater treatment. Water Sci. Technol. **1996**, *33*, 85–98.
5. Jewell, W.J. Resource-recovery wastewater treatments with biological systems. Proceedings of the Workshop on Sustainable Municipal Wastewater Treatment Systems, ETC-WASTE, Leusden, the Netherlands, 1996; 67–101.
6. Qasim, S.R. *Wastewater Treatment Plants: Planning, Design and Operation*; Holt, Rinehart and Winston: New York, 1985.
7. WPCF, Natural Systems for Wastewater Treatment. *Manual for Practice*, prepared by Task Force on Natural Systems,

Sherwood C. Reed, chairman, Water Pollution Control Federation, Alexandria VA, 1990.

8. American Public Health Association. *Standard Methods for the Examination of Water and Wastewater,* 17th Ed.; American Public Health Association: Washington, DC, 1989.

9. Water Environment Federation. *Design of Municipal Wastewater Treatment Plants*, 4th Ed.; Water Environment Federation: Alexandria, VA, 1998; Manual of Practice No. 8.

10. Rittmann, B.E.; McCarty, P.L. *Environmental Biotechnology: Principles and Applications*; McGraw-Hill: New York, 2001.

11. Henze, M.; van Loosdrecht, M.C.M.; Ekama, G.A.; Brdjanovic, D., Eds. *Biological Wastewater Treatment. Principles, Modelling and Design*; IAW Publishing: London, 2008.

12. Zehnder, A.J.B., Ed. *Biology of Anaerobic Microorganisms*. John Wiley and Sons: New York, 1988.

13. Colleran, E.; Finnegan, S.; Lens, P. Anaerobic treatment of sulphate-containing waste streams. Antonie van Leeuwenhoek **1995**, *67* (1), 29–46.

14. Polprasert, C. *Organic Wastes Recycling*; Wiley: Chichester, 1989.

15. Stafford, D.A.; Wheatley, B.I. *Anaerobic Digestion*; Applied Science Pub. Ltd.: London, 1979.

16. Bitton, G. *Wastewater Microbiology*, 4th Ed.; Wiley-Blackwell: New York, 1994.

17. Speece, R.E. Anaerobic biotechnology for industrial wastewater treatment. Environ. Sci. Technol. **1983**, *17*, 416A–427A.

18. Zeikus, J.G. Microbial populations in digesters. In *Anaerobic Digestion;* Stafford, D.A., Wheatley, B.I., Hughes, D.E., Eds.; Applied Science Pub. Ltd.: London, U.K., 1980; 61–89.

19. Gujer, W.; Zehnder, A.J.B. Conversion processes in anaerobic digestion. Water Sci. Technol. **1983**, *15*, 127–167.

20. Dolfing J. Acetogenesis. In *Biology of Anaerobic Microorganisms*; Zehnder A.J.B., Ed.; John Wiley and Sons: New York, 1988; 417–468.

21. Gavala, H.N.; Angelidaki, I.; Ahring, B.K. Kinetics and modelling of anaerobic digestion process. Adv. Biochem. Eng. Biotechnol. **2003**, *81*, 57–93.

22. Seghezzo, L.; Zeeman, G.; van Lier, J.B.; Hamelers, H.V.M.; Lettinga, G. A review: The anaerobic treatment of sewage in UASB and EGSB reactors. Bioresour. Technol. **1998**, *65*, 175–190.

23. Lettinga, G.; van Velsen, A.F.M.; Hobma, S.W.; de Zeeuw, W.; Klapwijk, A. Use of up-flow sludge blanket (USB) reactor concept for biological wastewater treatment, especially for anaerobic treatment. Biotechnol. Bioeng. **1980**, *22*, 699–734.

24. Schink, B. Principles and limits of anaerobic degradation: Environmental and technological aspects. In *Biology of Anaerobic Microorganisms*; Zehnder, A.J.B., Ed.; John Wiley and Sons: New York, 1988; 771–846.

25. Ahammad, Sk. Z.; Gomes, J.; Sreekrishnan, T.R. A comparative study of two high cell density methanogenic bioreactors. Asia Pac. J. Chem. Eng. **2011**, *6*, 95–100.

26. Dolfing J. Granulation in UASB reactors. Water Sci. Technol. **1986**, *18* (12), 15–25.

27. Punal, A.; Lema, J.M. Anaerobic treatment of wastewater from a fish-canning factory in a full-scale upflow anaerobic sludge blanket (UASB) reactor. Water Sci. Technol. **1999**, *40* (8), 57–62.

28. Gerardi, M.H. *The Microbiology of Anaerobic Digesters*; John Wiley and Sons, Inc., Hoboken, New Jersey, USA, 2001.

29. Kim, In S.; Hwang, Moon H.; Jang, Nam J.; Hyun, Seong H.; Lee, S.T. Effect of low pH on the activity of hydrogen utilizing methanogen in bio-hydrogen process. Int. J. Hydrogen Energy **2004**, *29*, 1133–1140.

30. Sahm, H. Anaerobic wastewater treatment. Adv. Biochem. Eng. Biotechnol. **1984**, *29*, 84–115.

31. Lawrence, A.W.; McCarty, P.L.; Guerin, F.J.A. The effects of sulfides on anaerobic treatment. Air Water Int. J. **1966**, *110*, 2207–2210.

32. McHugh, S.; Carton, M.; Collins, G.; O'Flaherty, V. Reactor performance and microbial community dynamics during anaerobic biological treatment of waste waters at 16–37°C. FEMS Microbiol. Ecol. **2004**, *48*, 369–378.

33. Nozhevnikova, A.N.; Zepp, K.; Vazquez, F.; Zehnder, A.J.B.; Holliger, C. Evidence for the existence of psychrophilic methanogenic communities in anoxic sediments of deep lakes. Appl. Environ. Microbiol. **2003**, *69* (3), 1832–1835.

34. Bergey D.H.; Holt, J.G.; Krieg, N.R.; Sneath, P.H.A. *Bergey's Manual of Determinative Bacteriology*, 9th Ed.; Lippincott Williams and Wilkins: Philadelphia, PA, 1994.

35. van den Heuvel, J.C.; Beeftink, H.H.; Verschuren, P.G. Inhibition of the acidogenic dissimilation of glucose in anaerobic continuous cultures by free butyric-acid. Appl. Microbiol. Biotechnol. **1988**, *29* (1), 89–94.

36. Gyure, R.A.; Konopka, A.; Brooks, A.; Doemel, W. Microbial sulfate reduction in acidic (pH-3) strip-mine lakes. FEMS Microbiol. Ecol. **1990**, *73* (3), 193–201.

37. Dogan, T.; Ince, O.; Oz, N.A.; Ince, B.K. Inhibition of volatile fatty acid production in granular sludge from a UASB reactor. J. Environ. Sci. Health A **2005**, *40* (3), 633–644.

38. Daniels, L.; Belay, N.; Rajagopal, B.S. Assimilatory reduction of sulfate and sulfite by methanogenic bacteria. Appl. Environ. Microbiol. **1986**, *51* (4), 703–709.

39. O'Flaherty, V.; Colohan, S.; Mulkerrins, D.; Colleran, E. Effect of sulphate addition on volatile fatty acid and ethanol degradation in an anaerobic hybrid reactor. II: Microbial interactions and toxic effects. Bioresour. Technol. **1999**, *68* (2), 109–120.

40. Hwu, C.S.; van Beek, B.; van Lier, J.B.; Lettinga, G. Thermophilic high-rate anaerobic treatment of wastewater containing long-chain fatty acids: Effect of washed out biomass recirculation. Biotechnol. Lett. **1997**, *19* (5), 453–456.

41. Daffonchio, D.; Thaveesri, J.; Verstraete, W. Contact angle measurement and cell hydrophobicity of granular sludge from upflow anaerobic sludge bed reactors. Appl. Environ. Microbiol. **1995**, *61*, 3676–3680.

42. Hanaki, K.; Matsuo, T.; Nagase, M. Mechanisms of inhibition caused by long chain fatty acids in anaerobic digestion process. Biotechnol. Bioeng. **1981**, *23*, 1591–1560.

43. Teske, A.; Alm, E.; Regan, J.M.; Toze, S.; Rittmann, B.E.; Stahl, D.A. Evolutionary relationship among ammonia- and nitrite-oxidizing bacteria. J. Bacteriol. **1994**, *176*, 6623–6630.

44. Zumft W.G. The denitrifying prokaryotes. In *The Prokaryotes. A Handbook on the Biology of Bacteria: Ecophysiology*

Isolation Identification Applications; Balows, A, Truper, H.G., Dworkin, M., Harder, W., Schleifer, K.H., Eds.; 2nd Ed.; Springer-Verlag: New York, 1992; Vol. 1, 554–582.

45. Jetten, M.S.M.; Schmid, M.; Schmidt, I.; Wubben, M.; Van Dongen, U.; Abma, W. Improved nitrogen removal by application of new nitrogen-cycle bacteria. Rev. Environ. Sci. Biotechnol. **2002**, *1*, 51–63.

46. Hellinga C.; Schellen A.A.J.C; Mulder J.W.; van Loosdrecht, M.C.M. The Sharon process: An innovative method for nitrogen removal from ammonium-rich wastewater. Water Sci. Technol. **1998**, *37*, 135–142.

47. Jetten, M.S.M.; Wagner, M.; Fuerst, J.; van Loosdrecht, M.C.M.; Kuenen, J.G.; Strous, M. Microbiology and application of the anaerobic ammonium oxidation (anammox) process. Curr. Opin. Biotechnol. **2001**, *12*, 283–288.

48. Jetten, M.S.M.; Horn, S.J.; Van Loosdrecht, M.C.M. Towards a more sustainable municipal wastewater treatment system. Water Sci. Technol. **1997**, *35*, 171–180.

49. van Loosdrecht M.C.M. *Recent Development on Biological Wastewater Nitrogen Removal Technologies*, In Proceedings of the International Conference on Wastewater Treatment for Nutrient Removal and Reuse (ICWNR'04); Bangkok, Thailand, 2004.

50. Barnard J.L. Biological nutrient removal without addition of chemicals. Water Res. **1975**, *9* (5–6), 485–490.

51. Mino, T.; van Loosdrecht, M.C.M.; Heijnen, J.J. Microbiology and biochemistry of the enhanced biological phosphate removal process. Water Res. **1998**, *32* (11), 3193–3207.