

## Microbial Roles and Dynamics in Wastewater Treatment Systems: An Overview

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### ABSTRACT

*The quality of wastewater effluents is responsible for the degradation of receiving water bodies, such as lakes, rivers, streams. The two main processes for the removal of impurities from wastewater influents are chemical and biological treatment but due to some drawbacks of the chemical treatment, biological treatment is now employed. Microorganisms are of major importance in industrial wastewater treatment, agricultural and aquaculture. They reside in the sediment and other substrates, and in the water of aquaculture facilities. Microorganisms may have positive or negative effects on the outcome of aquaculture operations. Positive microbial activities include elimination of toxic materials such as ammonia, nitrite, and hydrogen sulfide, degradation of uneaten feed, and nutrition of aquatic animals such as shrimp, fishes. These and other functions make microorganisms the key players in the health and sustainability of aquaculture. The role of the different microbial groups present in the waste water treatment systems with particular importance of bacteria and protozoa in the removal process of nitrogen and phosphorus indicate that the biological treatment system is very effective in the wastewater treatment systems.*

**Keywords:** Wastewater Treatment Systems, Microorganisms, protozoa, bacteriophages.

### INTRODUCTION

The major microbial populations found in wastewater treatment systems are bacteria, protozoa, viruses, fungi, algae and helminthes. The presence of most of these organisms in water leads to spread of diseases<sup>1</sup>. The majority of waterborne microorganisms that cause human disease come from animal and human fecal wastes. These contain a wide variety of viruses, bacteria, and protozoa that may get washed into drinking water supplies or receiving water bodies<sup>2</sup>. Microbial pathogens are considered to be critical factors contributing to numerous waterborne outbreaks. Many microbial pathogens in wastewater can cause chronic diseases with costly long-term effects, such as degenerative heart disease and stomach ulcer. The density and diversity of these pollutants can vary depending on the intensity and prevalence of infection in the sewer community. The detection, isolation and identification of the different types of microbial pollutants in wastewater are always difficult, expensive and time consuming. To avoid this, indicator organisms are always used to determine the relative risk of the possible presence of a particular pathogen in wastewater<sup>3</sup>.

Viruses are among the most important and potentially most hazardous pollutants in wastewater. They are generally more resistant to treatment, more infectious, more difficult to detect and require smaller doses to cause infections<sup>4,5</sup>. Because of the difficulty in detecting viruses, due to their low numbers, bacterial viruses (bacteriophages) have been examined for use in faecal pollution and the effectiveness of treatment

processes to remove enteric viruses. Bacteria are the most common microbial pollutants in wastewater. They cause a wide range of infections, such as diarrhea, dysentery, skin and tissue infections, etc. Disease-causing bacteria found in water include different types of bacteria, such as *E. coli* O157:H7; *Listeria* sp., *Salmonella* sp., *Leptosporosis* sp., etc (CDC, 1997). The major pathogenic protozoans associated with wastewater are *Giardia* sp. and *Cryptosporidium* sp. They are more prevalent in wastewater than in any other environmental source<sup>6,4</sup>.

The two major chemical pollutants in wastewater are nitrogen and phosphorus. Although there are other chemical pollutants, such as heavy metals, detergents and pesticides, however nitrogen and phosphorus are the most frequent limiting nutrients in eutrophication<sup>7,8</sup>. The processes for the removal of impurities in wastewater can be divided into two: chemical and biological. Chemical removal is a method of wastewater treatment in which chemicals are added to form particles which settle and remove contaminants. The treated water is then decanted and appropriately disposed of or reused after the resultant sludge is dewatered to reduce the volume. The most common techniques in chemical treatment are coagulation/flocculation, chlorination, chloramination, ozonation and ultraviolet light (UV)<sup>9,10</sup>. All biological-treatment processes take advantage of the ability of microorganisms to use diverse wastewater constituents to provide the energy for microbial metabolism and the building blocks for cell synthesis. This metabolic activity can remove contaminants that are as varied as raw materials and by-products<sup>11</sup>.

The aim of this paper was to review the various wastewater treatment systems and examine the roles and dynamics of the various microorganisms that are found in wastewater treatment systems.

### **Biological Wastewater Treatment Systems**

The fundamental reason for the treatment of wastewater is to circumvent the effect of pollution of water sources and protect public health through the safeguarding of water sources against the spread of diseases. This is carried out through a variety of treatment systems, which could be onsite treatment systems or offsite treatment systems. This section is therefore aimed at describing the offsite (activated sludge, trickling filters, stabilization ponds, constructed wetlands, membrane bioreactors) wastewater treatment systems<sup>12</sup>.

#### **Activated sludge**

The activated sludge is a process that has to do with high concentration of microorganisms, basically bacteria, protozoa and fungi, which are present as loose clumped mass of fine particles that are kept in suspension by stirring, with the aim of removing organic matter from wastewater<sup>13</sup>. An activated sludge can also be referred to as sewage that contains active microorganisms which helps in the breakdown of organic matter. It is the most versatile and effective of all the wastewater treatment systems. In an activated sludge system, the microorganisms present in the aeration tank can breakdown the organic matter present in the wastewater. The microorganisms are composed of 70-90 % organic matter and 10-30 % inorganic matter. The types of cells vary, depending on the chemical conditions and the specific characteristics of the organisms in the biological mass<sup>14</sup>. After the mixed liquor is discharged from the tank, a clarifier (also referred to as a settling or sedimentation tank) separates, by gravity the suspended solids from the treated wastewater. The concentrated biological solids are then recycled back to the aeration tank, to maintain a concentrated population of microorganisms to treat the wastewater. Because microorganisms are continuously produced in the system, a way must be provided to dissipate the excess biological solids produced. Wasted solids from the aeration tank are lower in concentration than those from the clarifier; therefore, a higher volume of sludge has to be handled. Depending on the design and operation of the process, it is possible to maximize or minimize the production of solids<sup>15</sup>.

A typical activated process consists of an aeration tank, a means of transferring oxygen to the microorganisms present in the aeration tank, a means of stirring the mixture of fluid dispersed in the aeration tank, a means of separating the microorganisms from the treated water and a system of recycling some of the microorganisms back to the reactor<sup>16</sup>. The activity of the microorganisms helps in the oxidation of the sewage organic matter into carbon dioxide and water. During the early stage of treatment, the large floating materials in wastewater are first screened out, before the sewage is allowed to pass

through the settling chamber, which helps in the removal of sand and other materials, while the floating debris is shredded and ground. In the aeration tank of the system, air is passed through the effluent for primary treatment. It is indicated that an activated sludge is an efficient system that has the ability to remove 75-90% of the biological oxygen demand (BOD) from sewage<sup>17,18</sup>.

In the activated sludge, the carbonaceous organic matter of the wastewater provides the energy source for the production of new cells. The microbes convert the carbonaceous organic matter of the wastewater into cell tissue and oxidize end products that include CO<sub>2</sub>, SO<sub>4</sub>, NO<sub>3</sub> and PO<sub>4</sub><sup>19</sup>. The activated sludge system is perhaps the most widely-used process for the reduction of the concentration of dissolved, particulate and colloidal organic pollutants in wastewater. The basic design parameters for the process are fairly well known and adequate conservation design standards based on the empirical data that have evolved over the years<sup>14</sup>.

Most activated sludge processes are used to degrade carbonaceous biochemical oxygen demand (BOD). It is also possible to design and operate the process to oxidize ammonia (nitrification) and reduce nitrate to nitrogen gas. Many activated sludge plants are now designed to achieve nitrification, while other systems' modification include phosphorus removal and biological nitrification<sup>20</sup>. Activated sludge plant designs are based on the length of time the sludge is kept in the system, typically termed solid retention time (SRT) or on the amount of food provided to the microorganisms in the aeration tank (the food-to-microorganism ratio (F:M) and the hydraulic detention time<sup>10</sup>. The constant aeration, agitation and recirculation in an activated sludge system, create an ideal environment for the numerous microorganisms present, while inhibiting the growth of larger organisms. Bacteria, fungi, protozoa, rotifers and nematodes are commonly found in activated sludge, though all may not exist in any single system. Despite the presence of other microorganisms, the bacteria are typically considered to be the significant organisms consuming the organic matter in wastewater. Algae, because of their need for light rarely exist in mixed liquor<sup>10</sup>. The overall reactions occurring in the activated sludge system are determined by the composite metabolism of all the microorganisms in the activated sludge<sup>15</sup>. The metabolic process consists of the separate, yet simultaneously occurring reactions of synthesis and respiration. Synthesis is the use of a portion of the waste matter (food) for the production of new cells (protoplasm), while respiration is the coupled release of energy through the conversion of food material to lower energy-containing compounds, typically carbon dioxide, water and possibly the various oxidized products forms of nitrogen. The precise nature of the products formed depends to some extent on process design, including reaction time, temperature and process loading of the system<sup>15</sup>.

### ***Trickling filter***

A trickling filter is a commonly used method of secondary wastewater treatment. It is made up of a filter bed that contains a highly permeable media (gravel or plastic material etc), which has a layer of microorganisms on the surface that leads to the formation of a slime layer. In a trickling filter system, the microorganisms are attached to the media in the bed and form a biofilm over it. As the wastewater passes through the media, the microorganisms consume and remove contaminants from the wastewater<sup>22</sup>.

In a trickling filter, the sewage is sprayed over the permeable media (bed of rocks, molded plastic, gravel and ceramics etc). The media must be large enough so that air will be able to pass through to the bottom but small enough to maximize the surface area available for microbial activity. A biofilm of aerobic microorganisms grows on the media because air passes through the media, the aerobic microorganisms in the slime layer can oxidize the organic matter trickling over the surface into carbon dioxide and water. This treatment system removes 80 - 85% of the BOD so they are less efficient than activated sludge systems. They are easier to operate and do not have problem with toxic sewage. Trickling filters consist of a septic tank, a clarifier and an application system. The septic tank helps with the removal of the solids present in the wastewater; the clarifier enables the biological materials to settle out of the wastewater and the application system assists in the distribution of the treated wastewater to the proper site. Prior treatment, wastewater that is passed to a trickling filter must first be pretreated to remove solid and greasy

materials, so as to prevent them from covering the thin layer of microorganisms present and to prevent the solid and greasy materials from killing them.

Trickling filters can be classified as high rate or low rate according to hydraulic or organic loading. Low rate filters has to do with simple treatment that produces a consistent effluent quality. The low rate trickling system should be able to remove 80-85% of applied BOD. High rate filters are usually characterized by higher hydraulic and organic loadings than the low rate filters. Recirculation takes place in high rate filters while for low rate filters, it does not occur. Recirculation is the process by which filter effluent is returned and reapplied on the filter. This recycling of the wastewater increases the application of the waste with microorganisms thereby making the effluent to undergo proper treatment<sup>23</sup>.

### ***Membrane bioreactor***

A membrane bioreactor is a combination of the biological degradation process of an activated sludge with a direct solid-liquid separation by membrane filtration through the use of micro or ultrafiltration membrane technology. The system allows for the complete physical retention of bacterial flocs and all suspended solids within the bioreactor. The advantages of a membrane bioreactor over other treatment systems is its high effluent quality, good disinfection capability, higher volumetric loading and less sludge production. A membrane bioreactor (MBRs) is a biological wastewater treatment process that uses membrane to replace the gravitational settling of the conventional activated sludge process for the solid-liquid separation of sludge suspension. Membrane bioreactors are used to treat biologically active wastewater feeds from municipal or industrial sources Two MBR configurations could exist, internal/submerged and external/sidestream. In the submerged, the membranes are immersed in and integral to the biological reactor while in the external/sidestream, the membranes are a separate unit process that requires intermediate pumping steps<sup>79,80</sup>.

The membranes in membrane bioreactor system are made up of polymers or inorganic substances. They are made of several small pores, which can only be seen with the help of a microscope. Because of their small pore size, only very tiny particles and water is allowed to pass through the membrane<sup>24</sup>. Although several configurations of membrane are known, the common ones that are applied in membrane bioreactor are the hollow fiber, flat sheet and tubular membranes. The hollow fiber and flat sheet membranes are usually dipped in water while the tubular membrane is usually placed outside the bioreactor<sup>25</sup>. The two major problems in hollow fibers are sludging and braiding. Both braiding and sludging can affect the membrane surface area so mechanical cleaning should also be carried out. Braiding is caused by the presence of hairs and cellulose fibers that ring around the membrane surface. Braiding can be avoided through careful screening of influents or recirculated liquid. On the other hand, sludging is caused by the accumulation of thick soft wet mud of industrial wastes. This is avoided by ensuring a sufficient flow of water in the medium<sup>25,26</sup>.

In the membrane bioreactor, the rate of passing of water through the membrane is called membrane flux. In some operations, the membrane flux likely to decrease leading, which may lead to membrane fouling, which is a great problem that increases operation cost and may require membrane cleaning. Membrane fouling may be caused by the accumulation of particles on the membrane surface, which leads to resistance in the membrane filtration. To control membrane fouling control, aerators are installed beneath the membrane<sup>27</sup>. In a membrane filtration, periodic relaxation, and back flushing for the removal of the foul layer from the membrane surface are some of the operations that are carried out.

The MBR process competes with other biological wastewater treatment systems, such as the conventional activated sludge. Although conventional biological processes perform well in meeting normal discharge standards and are cost effective, they can struggle to meet treatment standards for discharge into sensitive environments. Also, conventional processes are indicated not to be cost effective for wastewater reuse, unless ultrafiltration or microfiltration membranes are used as a post treatment<sup>28</sup>.

### ***Stabilization pond***

A wastewater stabilization pond is one of the most important natural methods for wastewater treatment. It is usually a shallow man-made pond that consists of single or several series of anaerobic, facultative or

maturation ponds<sup>81</sup>. In this system, the treatment of the wastewater starts from the anaerobic pond, which is designed for the removal of suspended solids and some other organic matter present. In the second stage, also known as the facultative pond, the remaining organic matter is removed through the activity of algae and the heterotrophic bacteria e.g *Arthrobacter* sp., *Rhodococcus* sp., *Pseudomonas* sp.<sup>82</sup>. During the last stage, treatment is achieved through the maturation pond, whose main function is the removal of pathogens and nutrients. The system is said to be a cost effective wastewater treatment system for the removal of pathogenic microorganisms. The treatment of the wastewater is gained through the use of natural disinfection mechanisms. Stabilization ponds are suitable for tropical and subtropical countries because of the intensity of sunlight and the temperature which aids the effectiveness of the removal processes<sup>29</sup>.

Anaerobic ponds are the smallest of the series of stabilization ponds processing, they are about 2-5m deep and receive high organic matter. This high organic matter produces strict anaerobic conditions meaning that dissolved oxygen is not present in the pond. The function of the anaerobic pond can be likened to the open septic tanks. A well designed anaerobic pond can gain around 60% organic matter removal at 20°C<sup>78</sup>. This pond is very efficient and also requires small area and it produces a very rich nutrient sludge which can be used for other purposes. In a warm condition, the BOD removal can be as high as 60-85% within a short period of time<sup>30</sup>

Facultative ponds are of two types; a primary facultative pond which has to do with the receiving of raw wastewater and the secondary facultative ponds which receives the settled water from the first stage (anaerobic pond). They are designed for the removal of organic matter based on the low organic surface load to permit the development of algal population; by this algae generate oxygen needed for them to remove soluble organic matter. Because of the algal population the water gets a dark green colouration but can also turn red or pink due to the presence of purple sulphide oxidizing photosynthetic activity. The change in colour in facultative ponds is an indicator that removal process is going on in the pond. At times, it could be due to some Red algae, e.g. species of Rhodophyta. Wind velocity is of great importance because it helps in the mixing of the pond water and this indicates that there is a uniform distribution of organic matter, dissolved oxygen, bacteria and algae<sup>29,31</sup>.

### **Constructed wetland**

Constructed wetlands are excavated basins with irregular perimeters and undulating bottom contours into which wetland vegetation is purposely placed to enhance pollutant removal from stormwater runoff. Stormwater enters a constructed wetland through a fore bay where the larger solids and coarse organic materials settle out. The stormwater discharged from the fore bay passes through emergent vegetation that filters organic materials and soluble nutrients. The vegetation can also remove some dissolved nutrients. Constructed wetlands can also be designed to reduce peak stormwater flows<sup>32,33,22</sup>.

The use of constructed wetlands can be looked at in two ways. First, a constructed wetland may be used primarily to maximize pollutant removal from stormwater runoff and also help to control stormwater flows. It may also be used primarily to control stormwater flows, with increased pollutant removal capabilities. The secondary benefits of constructed wetlands include preservation and restoration of the natural balance between surface waters and ground waters, increased wildlife habitats, and higher property values, if the same area were turned into a rectangular stormwater basin<sup>22</sup>.

The system takes advantage of many of the processes that exists in natural wetlands. Constructed wetlands treatment systems can be divided into two; subsurface flow and free water surface systems<sup>83</sup>. In the free water surface system, pollutants are removed from the wastewater by the decomposing microorganisms (mostly bacteria and fungi) living on the surface of the aquatic plants and soils<sup>34</sup>. During decomposition, oxygen is utilized by microorganisms attached to the aquatic plants below the level of the water. Apart from helping in the decomposition process, the aquatic plants also play an important role in the uptake of nutrients, such as nitrogen, phosphorus and other compounds from the wastewater. When the plants die and decompose, some of the nitrogen and phosphorus is released back into the water<sup>77</sup>. Many of these systems have been prepared and run to improve water quality, but also to provide high quality wetland habitat for migratory birds. Many of the systems are used for wastewater treatment, reuse and disposal systems<sup>35</sup>.

In the subsurface flow, the system is designed to create subsurface flow through a permeable medium, keeping the water that is treated below the surface. This helps in avoiding the development of odours and other problems<sup>36</sup>. The media, which is typically soil, sand, gravel or crushed rock usually, affects the hydraulics of the system. In the free water surface, the system is designed to replicate the natural wetlands with water flowing through at shallow depths. The two types of wetlands treatment systems are constructed in basins or channels with a natural or constructed subsurface barrier to limit leaching<sup>37,74</sup>.

### **Roles and Dynamics of Microorganisms in Wastewater Treatment Systems**

The major microbial populations found in wastewater treatment systems are bacteria, protozoa, viruses, fungi, algae and helminthes. The presence of most of these organisms in water leads to spread of diseases<sup>84</sup>.

#### **Bacteria**

In wastewater treatment systems, bacteria play vital role in the conversion of organic matter present to less complex compounds. In terms of size, bacteria range from 0.2- 2.0  $\mu\text{m}$  in diameter and are responsible for most of the wastewater treatment in septic tanks<sup>39</sup>. Though not all bacteria are harmful, a number of them cause water-related diseases in human and animals. Some of these diseases include cholera, dysentery, typhoid fever, salmonellosis and gastroenteritis<sup>40</sup>. Bacteria can be found entangled in flocs as in the case of activated sludge, where some play important role in biological treatment while some like the filamentous bacteria can cause serious problem in settling and foaming<sup>40,41</sup>. Waterborne gastroenteritis of unknown cause is frequently reported, with susceptible agent being bacteria. Certain strains of *Pseudomonas* and *Escherichia coli* which may affect the newborn are potential sources of this disease. These strains of microbes have also been implicated in gastrointestinal disease outbreaks<sup>42</sup>.

Bacteria are of the greatest numerical importance in wastewater treatment systems. The majority is facultative living in either the presence or absence of oxygen<sup>6,19,43</sup>. Although both heterotrophic and autotrophic bacteria are found in wastewater treatment systems the predominant ones are the heterotrophic bacteria. Generally, heterotrophic bacteria obtain their energy from the carbonaceous organic matter in wastewater effluent. The energy obtained is used for the synthesis of new cells and also for the release of energy through the conversion of organic matter and water. Some important bacteria genera that are found in wastewater treatment systems are *Achromobacter*, *Alcaligenes*, *Arthrobacter*, *Citromonas*, *Flavobacterium*, *Pseudomonas*, *Zoogloe* and *Acinetobacter*<sup>44,45,46</sup>.

In wastewater treatment systems, bacteria are responsible for the stabilization of influent wastes. The majority of the bacteria are known to form floc particles. The floc particles are clusters of bacteria that break down waste. Also, the floc particles also serve as sites on which waste can be absorbed and broken down. Filamentous bacteria form, which trichomes or filaments provide a backbone for the floc particles, allowing the particles to grow in size and withstand the shearing action in the treatment process. When filamentous bacteria are present in excessive numbers or length, they often cause solid/liquid separation or settleability problems<sup>3,10</sup>.

Furthermore, bacteria are the most common microbial pollutants in wastewater. The presence of pathogenic bacteria can be indicated using the tests for total and faecal coliforms<sup>46,47</sup>. Conventionally, the detection of faecal coliform is generally accepted as a reliable indicator of faecal contamination<sup>43</sup>. *Escherichia coli* is also regarded as a good and reliable indicator for faecal pollution from animal and human sources since it is known not to last for long periods outside the faecal environment<sup>85</sup>. The tests for total and faecal coliforms can be carried out, using either the traditional or enzymatic methods. Traditionally, the tests for total and faecal coliforms are carried out using the multiple-tube fermentation or by membrane filtration techniques. While the multiple-tube fermentation technique is used for medium or highly contaminated waters, the membrane filtration technique is used for low or very low contaminated waters.

#### **Protozoa**

Protozoa are microscopic, unicellular organisms that are also found in the wastewater treatment systems. They perform many beneficial functions in the treatment process, including the clarification of the

secondary effluent through the removal of bacteria, flocculation of suspended material and as bio-indicators of the health of the sludge. The protozoa that inhabit the wastewater treatment systems are capable of movement in at least one stage of their development<sup>48,49</sup>. They are 10 times bigger than bacteria; they are unicellular organisms with membrane enclosed organelles. Protozoa prey on pathogenic bacteria which make it have an advantage in wastewater<sup>50</sup>. They can be classified into five groups depending on their mode of locomotion, which are the free swimming ciliates, crawling ciliates, stalked and sessile ciliates, flagellates and amoeboid<sup>51</sup>.

In wastewater treatment systems, protozoa are useful biological indicators of the condition of the systems<sup>52</sup>. Although some protozoa are able to survive up to 12 h in the absence of oxygen, they are generally known as obligate aerobes, hence prove to be excellent indicators of an aerobic environment. Additionally, they serve as indicators of a toxic environment and are capable of exhibiting greater sensitivity to toxicity than bacteria. An indication of possible toxicity in a treatment system is the absence of or a lack of mobility of protozoa. The presence of large numbers of highly evolved protozoa in the biological mass in a wastewater treatment system is indicated as a hallmark of a well-operated and stable system<sup>53,42</sup>.

They can be placed into one of five groups, according to their means of locomotion. These groups are the free-swimming ciliates, crawling ciliates and stalked/sessile ciliates, flagellates and amoebae<sup>52,51</sup>. The three types of ciliates are free-swimming ciliates, crawling ciliates and stalked ciliates. All of these three have short hair-like structures or cilia that beat in unison to produce water current for locomotion and capturing bacteria. The water current moves suspended bacteria into a mouth opening<sup>54,48</sup>.

In the aeration tank of biological processes, a true trophic web is established. The biological system of these plants consists of populations in continuous competition with each other for food. The growth of decomposers, prevalently heterotrophic bacteria, depends on the quality and quantity of dissolved organic matter in the mixed liquor<sup>55</sup>. For predators, on the other hand, growth depends on the available prey. Dispersed bacteria are thus food for heterotrophic flagellates and bacterivorous ciliates, which, in turn, become the prey of carnivorous organisms. The relationships of competition and predation create oscillations and successions of populations until dynamic stability is reached. This is strictly dependent on plant management choices based on design characteristics aimed at guaranteeing optimum efficiency<sup>55,42</sup>.

Some ciliates, however, are predators of other ciliates or are omnivorous, feeding on a variety of organisms including small ciliates, flagellates and dispersed bacteria. All bacterivorous ciliates rely on ciliary currents to force suspended bacteria to the oral region<sup>52</sup>. Ciliated protozoa are numerically the most common species of protozoa in activated sludge, but flagellated protozoa and amoebae may also be present. The species of ciliated protozoa most commonly observed in wastewater treatment processes include *Aspidiscacostata*, *Carchesiumpolypinum*, *Chilodonellauncinata*, *Operculariacoarcta*, *Operculariamicrodiscum*, *Trachelophyllumpusillum*, *Vorticella convallaria* and *Vorticella microstoma*<sup>51,49</sup>. Figure 2.2 indicates the trophic web in the activated sludge.

There is indication that the free-swimming ciliates such as *Litonotus* sp. and *Paramecium* sp., which have cilia on all their body surfaces are typically found suspended or swimming freely in the bulk solution. On the other hand, the crawling ciliates, such as *Aspidisca* sp. and *Euplotes* sp. possess cilia only on their ventral or belly surface where the mouth opening is located. The crawling ciliates are usually found on floc particles while the stalked ciliates, such as *Carchesium* sp. and *Vorticella* sp., possess their cilia around the mouth opening only and are attached to floc particles. They have enlarged anterior portion and a slender posterior portion. The beating of the cilia and the springing action of the stalk produce a water vortex that draws dispersed bacteria into the mouth opening<sup>54,56</sup>.

In wastewater systems, two types of amoebae are predominant, naked, such as *Actinophrys* sp., *Mayorella* sp. and *Thecamoeba* sp. and the shelled amoebae or testate amoebae, e.g. *Cyclopyxis* sp. The naked amoeba lack any protective covering while shelled amoebae possess a protective covering that consists of calcified material<sup>57,56</sup>. flagellated protozoa are oval in shape and possess one or more whip-like flagella. In wastewater treatment systems, the flagellated protozoa are propelled through the system by the help of the flagella in a cork-screw pattern of locomotion<sup>58,56</sup>.

### **Viruses**

Viruses are also found in wastewaters, particularly human viruses that are excreted in large quantities in faeces. Viruses that are native to animals and plants exist in smaller quantities in wastewater, although bacterial viruses may also be present<sup>4,5,86</sup>. They are the causative agents of some water-related infections in humans, such as gastrointestinal and respiratory infections, conjunctivitis and meningitis. It is reported that a majority of waterborne diseases due to unidentified sources were caused by enteric viruses<sup>46</sup>. They are very notorious and persistent when present in wastewater and can remain a viable source of infection for months after their entry into the wastewater<sup>60</sup>.

### **Fungi**

Fungi are also part of the microorganisms found in wastewater treatment systems. Fungi are multicellular organisms that are also constituents of the activated sludge. Under certain environmental conditions in a mixed culture, they metabolize organic compounds and can successfully compete with bacteria. Also, a small number of fungi are capable of oxidizing ammonia to nitrite, and fewer still to nitrate. The most common sewage fungus organisms are *Sphaerotilus natans* and *Zoogloea* sp.<sup>61,62</sup>.

A number of filamentous fungi are found naturally in wastewater treatment systems as spores or vegetative cells, although they can also metabolize organic substances. A number of fungi species, such as *Aspergillus*, *Penicillium*, *Fusarium*, *Absidia* and a host of others have been implicated in the removal of carbon and nutrient sources in wastewater<sup>38</sup>. Some fungi are also reported to have the ability to breakdown organic matter present in the sludge system. In a system with low pH, where bacterial growth is inhibited, the main role of the fungi is the breakdown of organic matter. Additionally, some fungi use their fungal hyphae for trapping and adsorbing suspended solids to accomplish their energy and nutrient requirements. Some filamentous fungi have been reported to secrete some enzymes which help in the degradation of substrates during wastewater treatment<sup>87</sup>.

### **Algae**

Algae can be found in wastewater because they are able to use solar energy for photosynthesis as well as nitrogen and phosphorus for their growth leading to eutrophication<sup>64</sup>. Some types of algae that can be found in wastewater include *Euglena* sp., *Chlamydomonas* sp., and *Oscillatoria* sp. Algae are significant organisms for biological purification of wastewater because they can be able to accumulate plant nutrients, heavy metals, and pesticides, organic and inorganic toxic substances. The use of microalgae in biological wastewater treatment has gained a lot of importance over the years<sup>65</sup>.

High rate algal pond is shallow and equipped with mechanical aeration and mixing by means of paddle wheels, 90% of BOD and 80% of nitrogen and phosphorus are treated in high rate algal ponds. Construction and energy costs are highly lower and the land requirement is not up to that of facultative pond in constructed wetlands<sup>66</sup>.

### **Helminth**

Nematodes are aquatic animals present in fresh, brackish and salt waters and wet or humid soil worldwide. Freshwater nematodes can be present in sand filters and aerobic treatment plants. They are present in large numbers in secondary wastewater effluents, biofilters and biological contractors. Freshwater nematodes inhabit freshwater below the water table with species utilizing oxygen dissolved in the fresh water. Nematodes are part of the ecosystem, serving as food for small invertebrates<sup>61,42,89</sup>. They crawl onto floc particles and move in whip-like fashion when in the free-living mode. They secrete a sticky substance to be able to anchor to a substrate (media), so that anchored nematodes can feed without interference from currents or turbulence. A lack of nematode activity can be one of the bio-indicators of a toxic condition that may be developing in the treatment process<sup>44,46,89</sup>.

There are many species of parasitic worms that have human hosts. Some of them have the ability to cause serious illnesses. They can be grouped into three, the roundworms, the flatworms and the annelids. The flatworms are divided into two groups, the tapeworms, which possess segments on their body and the flukes, which have a single, flat and unsegmented body. A common characteristic of most helminthes is that they reproduce through eggs, although the eggs may differ in size and shape. In wastewater, most



helminthes eggs are not always infectious. For the eggs to be infectious they need to be viable and larval development must occur. The larval development occurs at required levels of temperature and moisture. Helminth eggs can remain viable for 1-2 months in crops and it takes longer months in soil, fresh water and sewage and can take years in faeces and sludge<sup>67,68,69,70</sup>. Helminthes eggs are inactivated at elevated temperature (above 40°C) and reduced moisture (below 5%), although these conditions are not readily achieved during wastewater treatment; In typical water treatment, helminthes eggs are removed by physical means, such as sedimentation, filtration or coagulation-flocculation<sup>71,88</sup>.

### CONCLUSION

The main reason for treating wastewater is to prevent the spread of diseases by safeguarding water sources against pollution. Treatment of wastewater is one of the strategies for the management of water quality. Due to some drawbacks over the years concerning chemical treatment, biological treatment is now employed to avoid the unpleasant conditions in natural water resources. The incidence of biological nitrogen and phosphorus in wastewater treatment systems has been extensively investigated. Most nutrient removal studies are based on the presence of bacteria and their roles in the removal of phosphorus and nitrogen have been well documented. The role of protozoa in the removal of nutrient in wastewater treatment has also evolved over the years.

In recent years the main role of protozoa was in the effectiveness of the purifying process by feeding on bacteria thereby reducing their number and also in the process of nutrient mineralization. Large amount of microorganisms that actively contribute to the removal of nutrient is responsible for eutrophication of the water sources, therefore there is need for more nutrient related research monitoring unit in order to achieve unpolluted wastewater discharge into water bodies. This will help in ensuring effluent standards and limitations as set by regulatory bodies which will help in clearer understanding and explanation of observed microbial life in wastewater treatment systems especially in constructed wetlands due to migratory birds.

Fungi have also been reported to increase the degradability, settleability and dewaterability of wastewater sludge and contribute to the sludge management strategy. Our understanding of the microbial community structure in wastewater treatment systems continues to advance rapidly owing to the ongoing development and application of molecular methods. Today, for most of the important processes in wastewater treatment systems; the use of uncultured prokaryotic organisms have been identified which has generated a lot of enormous opportunity for the investigation of important microorganisms. Fundamental studies have shown that the diversity of functional important prokaryotic groups in wastewater treatment systems can be influenced by the plant design and also by the changes in the process stability. Contaminants like hydrocarbon, heavy metals, nitrogen and phosphorus in distributed water and discharged wastewater is a constant area of concern.

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