

Microorganisms in waste water treatment process



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When people think of microorganisms, they tend to think of unsafe pathogens. While this may be of concern there are beneficial microorganisms living ubiquitously around us as well. Microorganisms that live in air, soil, and groundwater live in a symbiotic cycle, consuming harmful chemicals and masses of organic materials. Therefore, it is only natural that scientists would harness the natural biodegradation of these in the field of Wastewater Management. Biotechnologists prefer to call this process bioremediation. They have been using bioremediation on wastewater for many years and have discovered a plethora of usable microorganisms. Due to the vast amounts of microorganisms capable of bioremediation, this paper is focusing on bacterium capable of breaking down organic material useful in treating wastewater.

Wastewater treatment is performed on a variety of waste sources such as agricultural, residential, and industrial waste. Many bacteria such as Nitrifactor, nitrobacter and paracoccus are important players in the treatment of industrial and sewage waste. The use of microbes in wastewater treatment plants is an integral piece of the wastewater treatment process due to the fact that microbial population in a facility can become depleted resulting in system back-ups, organic material build-up and overall reduction in system efficiency. It is at this point when supplementation of a microbial product becomes necessary. There are three stages of wastewater treatment: primary, secondary, and tertiary where microorganisms can be added or encouraged to grow in wastewater. The first two stages are concerned with large debris and organic matter removal by the use of a variety of filtration and sedimentation processes. Microbial

organisms are stimulated in the second and third stages and the goal is to degrade excessive amounts of contaminants such as nitrogen, phosphates, oils, chemicals and heavy metals by the third stage. One common practice used in residential wastewater in the second stage, is using activated sludge techniques, which aerates the waste to stimulate denitrifying and nitrifying microorganisms to biodegrade the waste. Most large sewage treatment plants use a two-phase digestion system in which organics are metabolized by bacteria, anaerobically. In the first stage, the sludge is heated and mixed in a closed tank for about 15 days, while digestion takes place. The sludge then flows into a second tank, which serves primarily for storage and settling. Sludge digestion is a biological process in which organic solids are decomposed into stable substances. Nitrifying and denitrifying organisms, both aerobic and anaerobic are added to convert about half of the organic sludge solids to liquids and gases (Siezen & Galardini, 2008).

Nitrosomonas europae was isolated in 1892 by Russian microbiologist Sergio Winogradsky. It has been a useful bacterium in wastewater treatment, usually added in the secondary treatment process due to its ability to breakdown organic material. If given an aerobic environment, ammonia is oxidized first to nitrite by ammonia-oxidizing bacteria, then nitrite is oxidized to nitrate by nitrite-oxidizing bacteria which makes *N. europaea* primarily important in the nitrification cycle (Arp and Bottomley, 2006). *Nitrosomonas europaea* is a bacillus shaped, gram-negative obligate chemolithoautotroph; which is an autotroph that gets its energy from oxidation of inorganic substances in the absence of light. It is a mobile bacteria with flagella located in its polar region. It commonly inhabits places rich in ammonia and

inorganic salt, such as in soils, freshwaters, stone monuments, and sewage. It obtains most of its energy from its ammonia-oxidizing capabilities, an unusual process for most bacteria. Cell division may take several days due to its need for large amounts of ammonia consuming about 25 moles of ammonia per mole of carbon dioxide assimilated into cellular biomass (Arp and Bottomley, 2006). Due to its long delay in cell division, scientists tend to avoid studying *Nitrosomonas europaea*. *N. europaea* gains carbon from the atmosphere by converting carbon in a gaseous form into carbon bound up in organic molecules. Its genome consists of a single circular chromosome with 2,812,094 bases. Its gene structure denotes that it must take in Fe and suggests it can take in other metals such as Cu, Cd, Zn, and Co as well (Chain, Lamerdin, Larimer, Ragala, Lao, 2003). *N. europaea* functions best at a basic pH but can tolerate a pH between 6.0-9.0 and it prefers temperatures between 20-30 degrees Celsius.

Nitrobacter hamburgensis got its name because it was isolated in soil of the Old Botanic Garden in Hamburg. It is a gram-negative bacteria that lives mainly in soil, building sandstone, and sewage sludge. It is pear-shaped and has one sub-polar flagellum. There is one circular DNA chromosome and three circular DNA plasmids with 4,406,967 base pairs on the chromosome. (Kaipa, et al, 2010). *N. hamburgensis* gains energy from oxidation of nitrite to nitrate and has the ability of metabolizing nitrogen in nitrite from its environment. It is found mainly in soil and freshwater. (Arp & Bottomley, 2006). The bacteria has provided a solution to removing high levels of nitrogen from municipal effluents of wastewater treatment plants. Biofilms with different nitrifying bacteria including *N. hamburgensis* have been

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constructed. Before the invention of these biofilms very large and expensive reactors were used for this purpose.

Paracoccus denitrificans, an organism that removes high levels of nitrogen in wastewater when paired with *Nitrosomonas europaea*, a nitrifying organism which reduces ammonia to nitrate. *P. denitrificans* is a spherical coccus shaped gram-negative bacteria having a double membrane cell wall. It inhabits soils in either aerobic or anaerobic environments. First isolated in 1910 by Martinus Beijerinck, a Dutch microbiologist gave the organism the name *Micrococcus denitrificans*, only later to be changed by Diana H. Davis in 1969 to the current name *Paracoccus denitrificans* after the discovery that the bacteria contained many features known to be in mitochondria, possibly an ancestor to the eukaryotic mitochondria. (Davis, et al, 1969). The genome of *P. denitrificans* consists of two circular chromosomes and one plasmid. The first chromosome has 2, 852, 282 base pairs and the second chromosome has 1, 730, 097 base pairs. The plasmid has 653, 815 base pairs (Swiss Inst., 2007). Many of the proteins transcribed and translated from the plasmid is what gives *P. denitrificans* its unique features of the ability to metabolize ammonium to nitrogen gas. Due to *P. denitrificans* ability to produce more than 5000 proteins it is useful in biotechnological applications (Uemoto & Saiki, 2007).

Another process commonly used in wastewater treatment is the use of biofilms, various trickling rock filters that encourage biofilms. (Sillankorva, Neubauer, Azeredo, 2008) These biofilms build microorganism communities enclosed in a matrix of extracellular polymeric substances separated by water channels. Within these colonies are a variety of bacteria, fungi and

algae which biodegrade waste. *Pseudomonas fluorescens*, *P. syringae* and *P. putida* are a few of the bacteria found in biofilms. As well as fungus like *Mycelium* and algae. The biofilm community is an optimal environment for cell-cell interactions, including the cellular exchange of genetic material, and nutrient exchange within the community. The matrix protects the microorganisms from UV exposure, metal toxicity, acid exposure, dehydration and salinity, phagocytosis, antibiotics, and antimicrobial agents (Hall-Stoodley, et al 2004).

Pseudomonas fluorescens makes a great contribution to the turnover of organic matter and while present in soil, is abundant on the surfaces of plant roots and leaves. *P. fluorescens* grows at an optimum temperature of 25° Celsius but can also survive in temperatures as low as 0° degrees Celsius make it a rare pathogenic in humans. The bacteria's degrading ability has been applied to pollutants such as styrene, TNT and, polycyclic aromatic hydrocarbons (Sillankorva, Neubauer, Azeredo, 2008).

Notable is *P. putida* possessing a high biodegrading metabolism. It can breakdown styrene which is a highly polluting synthetic chemical, used to make plastics (Park, et al, 2005)). *Pseudomonas putida* is a gram-negative shaped bacteria, similar to *Pseudomonas aeruginosa*, a known pathogen to humans, however it is missing key gene segments that *P. aeruginosa* possess making it nonpathogenic. The biochemistry of *P. putida* makes it an aerobic, gram negative, fluorescent colored, rod-shaped bacteria. It a motile organism with one or more polar flagella. They are usually found in moist soil and water environments and grow optimally at room temperature. Certain strains have the ability to grow on and break down many dangerous

pollutants and aromatic hydrocarbons such as toluene, benzene, and ethylbenzene. *P. putida* can also be used in petroleum plants to purify fuel. *P. putida* is also closely related to *Pseudomonas syringae*, an abundant plant pathogen, but again it lacks the gene that causes such disease (DOE, 1998).

The first isolation of *Pseudomonas syringae* occurred in 1902 by van Hall from a diseased lilac. *Pseudomonas syringae* are aerobic rod-shaped gram negative bacteria that are motile with the use of several polar flagella. *Pseudomonas syringae* secretes a plant toxin making it a known plant pathogen. Therefore, it is easy to see its use in the biodegradation of organic waste. Each strain of this bacteria has a specific plant it targets and is often found on plant leaves. An interesting quality is its ability to form ice crystals, *P. syringae* is responsible for causing frost injury to frost-sensitive plants. (Feil, et al, 2005) This discovery led to its production of artificial snow.