

# What is MiCroBial Aqua Product?

The core MiCroBial Aqua is a natural biocatalyst made via a novel fermentative process that has been continually refined by microbiologist in India. MiCroBial Aqua consists of a select consortium of bacteria, enzymes, nutrients and co-factors that degrade organic matter as Carbohydrate, Proteins & Fat (FOG). These microbes produce different types of enzymes as amylase, Protease, Lipase, Cellulase etc. MiCroBial Aqua works in aerobic and anaerobic conditions as it contents aerobic and facultative anaerobic microbes.

# Composition

Denitrifying bacteria, Nitrifying bacteria, Fermenting bacteria, Hydrolytic enzymes etc.

MiCroBial product contains specifically selected microbes for their specific function that works in wastewater treatment It contains about 9 different strains of microbes and hydrolytic enzymes cocktail. Each microbe performs their own function. About 2 billions/Gms of each microbe are present in MiCroBial products. Key Benefits

• Reduction in sludge production, Sludge handling & disposal cost

- Reduce BOD, COD, TSS, Biological nutrients
- Improve MLSS, Biomass in the system
- Reduces odors
- Reduces aeration; Energy savings
- Reduces need for chemical additives
- Improves plant stability
- Reduces hydrogen sulfide, ammonia and nitrates
- Enhance nitrogen and phosphorus removal
- Inhibit the growth of pathogens by competing with them for nutrients

## **Mode of Action**

Nitrification & Denitrification Bacteria remove nitrogen from wastewater by a two step biological processes: nitrification followed by denitrification. Technically, it is a three step process: Ammonification precedes nitrification and denitrification.

## Ammonification

While traveling through sewer pipes, the majority of the nitrogen contained in raw sewage (urea and fecal material) is converted from organic-nitrogen to ammonia through a process called hydrolysis. Technically, in the majority of situations, more ammonium than ammonia is created during ammonification. The actual ratio is influenced by pH and temperature.

## **Nitrification by Nitrifying Bacteria**

Nitrification process performed by nitrifying bacteria such as Nitromonas in aerobic treatment.

The biological conversion of ammonia to nitrate nitrogen is called Nitrification. Nitrification is a two-step process. Bacteria known as Nitrosomonas convert ammonia and ammonium to nitrite. Next, bacteria called Nitrobacter finish the conversion of nitrite to nitrate. The reactions are generally coupled and proceed rapidly to the nitrate form; therefore, nitrite levels at any given time are usually low. These bacteria known as "nitrifiers" are strict "aerobes," meaning they must have free dissolved oxygen to perform their work. Nitrification occurs only under aerobic conditions at dissolved oxygen levels of 1.0 mg/L or more. At dissolved oxygen (DO) concentrations less than 0.5 mg/L, the growth rate is minimal. Nitrification requires a long retention time, a low food to microorganism ratio (F:M), a high mean cell residence time (measured as MCRT or Sludge Age), and adequate buffering (alkalinity). The nitrification process produces acid. The optimum pH for Nitrosomonas and Nitrobacter is between 7.5 and 8.5; most treatment plants are able to effectively nitrify with a pH of 6.5 to 7.0. Nitrification stops at a pH below 6.0. The nitrification reaction consumes 7.1 mg/L of alkalinity as CaCO3 for each mg/L of ammonia nitrogen oxidized. An alkalinity of no less than 50-100 mg/L is required to insure adequate buffering. Water temperature also affects the rate of nitrification.

Nitrification reaches a maximum rate at temperatures between 30 and 35 degrees C. At temperatures of 40o C and higher, nitrification rates fall to near zero.

At temperatures below 20 degrees C, nitrification proceeds at a slower rate, but will continue at temperatures of 10 degrees C and less. However, if nitrification is lost, it will not resume until the temperature increases to well over 10 o C. Some of the most toxic compounds to nitrifiers include cyanide, thiourea, phenol and heavy metals such as silver, mercury, nickel, chromium, copper and zinc. Nitrifying bacteria can also be inhibited by nitrous acid and free ammonia.

# $\rm NH3+O2 \rightarrow \rm NO2+3H_{+}+2e_{-}$

 $NH_3 + O_2 + 2H_+ + 2e_- \rightarrow NH_2OH + H2O$  $NH2OH + H2O \rightarrow NO2 + 5H_+ + 4e_-$ 

# **Denitrification by Denitrifying Bacteria**

The biological reduction of nitrate (NO3) to nitrogen gas (N2) by facultative heterotrophic bacteria is called Denitrification. "Heterotrophic" bacteria need a carbon source as food to live. "Facultative" bacteria can get their oxygen by taking dissolved oxygen out of the water or by taking it off of nitrate molecules.

Denitrification occurs when oxygen levels are depleted and nitrate becomes the primary oxygen source for microorganisms. The process is performed under anoxic conditions, when the dissolved oxygen concentration is less than 0.5 mg/L, ideally less than 0.2. When bacteria break apart nitrate (NO3 - ) to gain the oxygen (O2), the nitrate is reduced to nitrous oxide (N2 O), and, in turn, nitrogen gas (N2). Since nitrogen gas has low water solubility, it escapes into the atmosphere as gas bubbles. Free nitrogen is the major component of air, thus its release does not cause any environmental concern.

The formula describing the nitrification reaction follows:

# $NO2 + H2O \rightarrow NO3 + 2H_{+} + 2e_{-}$

Optimum pH values for denitrification are between 7.0 and 8.5.

Since denitrifying bacteria are facultative organisms, they can use either dissolved oxygen or nitrate as an oxygen source for metabolism and oxidation of organic matter. If dissolved oxygen and nitrate are present, bacteria will use the dissolved oxygen first. That is, the bacteria will not lower the nitrate concentration. Denitrification occurs only under anaerobic or anoxic conditions.

Another important aspect of denitrification is the requirement for carbon; that is, the presence of sufficient organic matter to drive the denitrification reaction. Organic matter may be in the form of raw wastewater, or supplemental carbon. Conditions that affect the efficiency of denitrification include nitrate concentration, anoxic conditions, presence of organic matter, pH, temperature, alkalinity and the effects of trace metals. Denitrifying organisms are generally less sensitive to toxic chemicals than nitrifiers, and recover from toxic shock loads quicker than nitrifiers.

Temperature affects the growth rate of denitrifying organisms, with greater growth rate at higher temperatures. Denitrification can occur between 5 and 300 C, and these rates increase with temperature and type of organic source present. The highest growth rate can be found when using methanol or acetic acid. A slightly lower rate using raw wastewater will occur, and the lowest growth rates are found when relying on endogenous carbon sources at low water temperatures.

Wastewater cannot be denitrified unless it is first nitrified. Biological Solution to Save Environment

## **Role of Fermenting Bacteria in Waste Water Treatment**

The mixture of domestic sewage, industrial effluent and ground water which has infiltrated into the sewers is present in Wastewater which is 99.99% water, with a small amount of dissolved or suspended solid matter.

One of the applications of fermentation process is in the treatment of wastewater. In the process of sewage disposal, sewage is digested by enzymes secreted by bacteria. Solid organic matters are broken down into harmless, soluble substances and carbon dioxide. The digested solids, also known as sludge, accumulated in a wastewater treatment process must be treated and disposed of in a safe and effective manner. The purpose of digestion is to reduce the amount of organic matter and the number of disease-causing microorganisms present in the solids.

Biological treatment of waste can be carried out aerobically, process known as composting or anaerobically through anaerobic digestion. The degradable organic compounds e.g. carbohydrates, proteins, fats, etc. in the waste water are broken down by aerobic micro-organisms mainly bacteria and rarely fungi. The result is an effluent with drastically reduced organic matter content. The materials difficult to digest form a sludge is treated separately either by aerobic breakdown of raw waste-water or anaerobic breakdown of sludge.

## Aerobic Digestion of Raw Waste Water

The most common method involve in aerobic breakdown of raw waste water is the activated sludge system. Activated sludge system involves the production of an activated mass of micro-organisms capable of stabilizing the organic content of a waste aerobically.

Waste water is introduced into an aerated tank of micro-organisms which are collectively referred to as activated sludge or mixed liquor. Aeration is achieved by the use of submerged diffused or surface mechanical aeration which maintain the activated sludge in suspension.

A wide range of bacteria are involved, including Pseudomonas, Lactobacillus and Yeast.

Microorganisms have to cope with an uncontrollably diverse range of organic and inorganic compounds some of which may be toxic to the organisms. The microorganisms occur in discreet aggregates known as flocs. Some of the settled biomass is recycled as 'returned activated sludge' inoculate the incoming raw sewage because it contains a community of organisms adapted to the incoming sewage. The solid undigested sludge may be further treated into economically valuable products.

#### Use of Fermented Sludge in Nutrient Removal

Fermentation can be used to produce carbon source for both nitrogen and phosphorus removal. It is a tool to reduce operational costs associated with biological nutrient removal. Removal of nitrogen and phosphorus from wastewater effluents is important to ensure environmental protection of surface waters. High concentrations of nutrients in rivers can cause eutrophication, oxygen depletion in the river waters and can stimulate algae growth. Thus it has been proposed as an efficient process to remove nutrients from wastewater.

#### **Biological Nitrogen Removal**

The processes for biological nitrogen removal can be incorporated into both activated sludge and percolating filter plants. The overall mechanism follows the route of nitrification (oxidation of ammonia to nitrite and nitrate) and denitrification (reduction of nitrate sequentially to nitrite, nitric oxide, nitrous oxide and nitrogen)]

Biological denitrification processes generally recirculate nitrified effluent as a source of nitrate. This means that 100% removal can never be achieved. Only the nitrate contained in the recirculated stream will be removed.

## **Biological Phosphorus Removal**

Biological phosphorus removal is dependent mainly on the ability of the bacterium Acinerobacter spp. to release phosphate under anaerobic conditions and to absorb it under aerobic conditions.

The mechanism of Phosphorus removal by Acinetobacter spp. includes the following steps:

Under anaerobic conditions, readily biodegradable organic matter becomes fermented to short chain fatty acids. These are stored in the cell as poly-hydroxyl butyrates (PHB).

Under aerobic conditions the stored PHB is oxidized and energy is released allowing the assimilation of soluble orthophosphate. The ortho-phosphate is metabolized by the cell and excess quantities are stored in the cell as polyphosphate. The storage of excess phosphate is known as 'luxury uptake' of Phosphorus and it is this particular ability of the cell that is exploited in the nutrient removal process.

## Anaerobic Digestion of Sludge

The commonest method of treating sludge however is by anaerobic digestion. This can be done by allowing the sludge to decompose in digesters under controlled conditions for several weeks. Digesters themselves are closed tanks with provision for mild agitation, and the introduction of sludge and release of gases. About 50% of the organic matter is broken down to gas, mostly methane.

Anaerobic Microbial Digestion usually involves four steps:

- Extracellular Hydrolysis (e.g., Cellulose)
- Fermentation leading to organic acids (VFAs), acetate, CO2 and H2
- Fermentation leading to acetic acid (CH3COOH), H<sub>2</sub> and O<sub>2</sub>.
- Methanogenesis leading to CH<sub>4</sub>, CO<sub>2</sub> and H<sub>2</sub>O.

The fermentation process occurs in two main steps: hydrolysis and acidogenesis. During hydrolysis the long chain molecules are broken down into smaller dissolved molecules by extracellular enzymes that are then converted to VFAs during the acidogenesis step.

First, during hydrolysis, water-insoluble biopolymers such as carbohydrates, proteins, and fats are decomposed by extracellular enzymes to water-soluble monomers (e.g., monosaccharide's, amino acids, glycerin, fatty acids) and thus made accessible to further degradation.

In the second step (acidification) the intermediates of hydrolysis are converted into acetic acid (CH3COOH), hydrogen (H2), carbon dioxide (CO2), organic acids, amino acids, and alcohols by different groups of bacteria. Some of these intermediate products (acetic acid, hydrogen, and carbon dioxide) can be directly used by methanogenic bacteria, but most of the organic acids and alcohol are decomposed into acetic acid, hydrogen, and carbon dioxide during acidogenesis. Only these products, as well as methanol, methylamine, and formate, can be transformed into carbon dioxide and methane (CH4) by methanogenic bacteria during the third and last step, methane formation.

A great advantage of anaerobic fermentation is the production of biogas that can be used as a source of energy. A major advantage of anaerobic digestion in comparison with aerobic composting is the ability of engineers to have total control over gaseous and liquid emissions, as well as having the potential to recover and use methane gas generated as the wastes degrade. Biological Solution to Save Environment

## Is MiCroBial Products Safe to Use?

MiCroBial products are safe for environment, animal, aquatic life and soil additives. It's completely biodegradable, 100% organic and non pathogenic.

# **Optimum Conditions for Application of MiCroBial Products**

COD: BOD ratio – Min 2:1 PH – 6-8 HRT – Min 24 hrs MLSS – 2000 – 4000 mg/l DO – Min 0.5 mg/l

TKN, TP, Ammonia, Nitrite, Nitrate, TSS, F: M ration, RAS/WAS, Temperature will also play vital role in biological treatment of waste water and monitored during application of MiCroBial products.

## Key Principles of Use:

There are four variables that most impact the effectiveness of MiCroBial product and are as follows;

## **Product Dosage:**

There is a wide range of effective concentrations or "doses" of MiCroBial product for certain applications, however the optimum range is generally between 2 and 10 milligrams per liter of volume—that is milligrams of MiCroBial product per liter of waste water or substrate. This is roughly equivalent to parts per million.

#### **Contact Time**

To maximize the MiCroBial product treatment program, MiCroBial product should be kept in contact with the target substrate to the greatest extent possible. This means that when applying MiCroBial product to a large volume of wastewater, such as a large lagoon, tank or vessel, it is important to maximize the **distribution** of MiCroBial product throughout that medium. Additionally, the **frequency** of MiCroBial product dosing should match or exceed residence time within the wastewater system. For example, if a septic tank has a residence time of 14 days, then MiCroBial product should be applied at least every 14 days. If a grease trap fills and empties completely every day, then daily dosing or continuous dosing with MiCroBial product would be advised.

#### Temperature

MiCroBial product can operate effectively in a wide range of temperatures. However, the catabolic break-down of organic waste molecules into inert compounds Slows significantly as the temperature of the substrate approaches the freezing point 0°C. There is a similar loss of effectiveness at temperatures above approximately 60°C. The ideal operating temperature is between 10°C to 49°C.

#### PH

MiCroBial product operates within a broad pH range, from 5.5 and 9.5, but the optimum pH is between 6.5 and 8.5 with neutral preferred.

# **Oxygen Level**

When using MiCroBial product in an aerobic process the performance of MiCroBial product can be improved by higher levels of dissolved oxygen. Optimum levels in Aerobic processes are DO>1.5mg/L, however MiCroBial product is equally effective in Anaerobic processes with lower DO such as 0.5 mg/L. Mixing of MiCroBial Products:

The MiCroBial product are packaged in freeze-dried powder form and delivered in vacuum-sealed bags. The powder must be pre-hydrated by mixing into fresh water before being applied to the wastewater or target medium. This step allows MiCroBial product to properly activate and form a high concentration of catalysts. Several factors govern the effectiveness of this MiCroBial product dose pre-activation phase:

#### **Mixing Time:**

1 ok of product can be mixed with fresh water and allow it to stand for 30 mins. It's recommended that for better results, MiCroBial product can be pre hydrated in fresh water for 30 mins.

#### **Concentration of Product:**

For pre-hydrating, MiCroBial product should be mixed with water to make solution at a concentration 10,000 milligrams of MiCroBial product per liter of water (parts per million). The ideal concentration is approximately 10,000 mg/L. Because MiCroBial product is packaged with its own nutrient source, it is important to use a relatively nutrient-free source of water for pre-hydrated mixing. It's recommended that 1 kg MiCroBial product can be mixed into 50-100 liter of fresh water.

#### **Temperature of Fresh Water:**

The range of optimum temperature for pre-hydrated MiCroBial product is not as broad as its effective temperature range once mixed with water. It is ideal to keep the pre-hydrated culture at a temperature above 15°C and below 55°C.

#### **Quality of Fresh Water:**

The product activation is a biological process. It is important that the water used does not contain any harsh chemicals, such as MiCroBial inhibitors, that would prevent the growth of the MiCroBial product Culture.

For dosing concentration and application, please contact to technical team.

Please refer to the MiCroBial Technologies website for the appropriate MSDS, www.microbialtech.com



Address : A Plot no 566, Karale Nagar, Degaon, North Solapur, Maharashtra, India Contact : Ph.: 8888885375 E : support@microbialtech.com