NITRO

Trout in the Classroom Super Hero

Story by Chuck Dinkel
Illustrations by Erika Lawson

RoseDog Books
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Story by:

Chuck Dinkel

Chuck is a member of the Potomac-Patuxent Chapter of Trout Unlimited (PPCTU) in Maryland. He serves as one of the two coordinators for the state's Trout in the Classroom (TIC) program. Currently 100 schools and environmental organizations across the state participate in TIC. His professional career as an electronics engineer spanned 30 years and included employment with the Naval Oceanographic Office, National Oceanic and Atmospheric Administration (NOAA), and the National Institute of Standards and Technology (NIST). Publications for which he has written articles include: Small Boat Journal, Fine Gardening, Maryland Natural Resource, and United States Power Squadron magazine. His enjoyment of fly fishing and sailing have taken him to many beautiful locations in the U.S. including several western states and Alaska. **NITRO – TIC SUPER HERO** is Chuck's second Trout in the Classroom published book. **MAC – The Macroinvertebrate** came to print in 2018.

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Illustrations by:

Erika Lawson

Erika, a gifted artist, graduated with honors from high school in 2018. Currently she is a college sophomore in majoring in Criminal Justice while holding down a part time job. Despite this very busy schedule she has found time to collaborate with the author on his second Trout in the Classroom book. Once again, she has demonstrated her unique ability to turn the author's very crude stick figure drawings into the beautiful illustrations that decorate this book; a true testament to her creative and artistic abilities.

Dedication

"NITRO – Trout in the Classroom Super Hero" is dedicated to the memory of two wonderful men who each in his own way contributed significantly to the Maryland Trout in the Classroom (TIC) program. Both lost a battle to cancer in 2020.

Jim Marecki, a Christian brother, loved working with Trout in the Classroom students. He and I spent many a day assisting teachers set up their fish tanks in preparation for the arrival of trout eggs. At spring time trout releases you'd often find Jim instructing students in the fine art of casting a fly rod, but he'd readily support other activities as the need arose. Yes, Jim was my right-hand TIC volunteer, but he was also a cherished friend and fishing companion. He'll be greatly missed by all who had the opportunity to interact with him through his support of TIC.

Keith Lipscomb, a Frederick County teacher, spearheaded his school's TIC program for many years. Keith loved his tech ed. students and he loved the opportunity to share his passion for raising trout with them. In the fall of 2019,

knowing he was facing an uphill battle, Keith and his family made a special trip to the school one weekend to set up his TIC tank. I had an opportunity to visit with Keith while he was under hospice care. I told him that the eggs had hatched and that I'd met with several of his former students who had agreed to raise the fish in his absence. When I told him the students' names a smile lit up across his face. He knew the fish were in good hands. Keith also knew he was in God's hands.

Acknowledgments

My background is in electronics and computer science. What biology and chemistry I still remember are from high school days. Serving as a Maryland TIC program coordinator has had more than its share of challenges. On many occasions I've wished I had more of a bio-chem education. The internet has helped provide some answers, but I've been very fortunate to have two individuals who over my TIC career have provided many more.

Dr. Douglas Dent, Senior Vice President and Technical Director for Product Development, Ecological Laboratories, Inc., has been one of my mentors. Doug has spent many an hour on the phone and through emails patiently answering a myriad of my questions related to the nitrogen cycle of fish tanks. Despite an extremely busy schedule, Doug has also taken time to serve as a presenter at TIC annual training sessions.

Dr. Drew Ferrier, heads up the Center for Coastal and Watershed Studies at Hood College in Frederick, Maryland.

The Center's focus is on all aspects of coastal environments and the watersheds that culminate at our shorelines. Drew has been very generous with his time and expertise as well. In less than a half hour I can be in Drew's office having face to face discussions about TIC related topics. Drew and his staff have helped train hundreds of TIC teachers and volunteers in the fine points of trout culture. He comes to TIC with first-hand experience having raised trout at the college for several years.

Drew and Doug are the individuals I turned to for guidance and advice regarding the bio-chemistry in this story. I especially appreciate both having agreed to reviewing the manuscript when asked.

Trout in the Classroom in Maryland would not exist were it not for the strong base of dedicated and passionate volunteers who annually support the program. These individuals come from all walks and organizations and comprise a very special and unique partnership. Foremast among these are the school teachers and educators who have understood the goals of TIC and embraced them. Today far too many students lack a basic understanding of their watersheds and the unique role they play in their lives. Life depends on the existence of clean water – the kind of clean, cold, oxygenated water in which trout thrive. TIC teachers are the carriers of this message and the students are getting it.

Annually the Maryland Department of Natural Resources' Aquatic Resources Education Section has provided grant

money to enable schools to cover the initial cost of TIC participation as well as support in training and logistics. Albert Powell Hatchery donates over 15,000 fertilized eggs to the program and the food needed to feed the hatchlings. Biologists and support personal respond to phone calls and emails to answer questions and conduct tours of the hatchery.

Members of Trout Unlimited (TU) are the heart and soul of TIC. Both at the local and national level, their love of the outdoors and fly fishing for trout, take a back seat to the stewardship role they play when it comes to protecting and defending the environment and watersheds needed to ensure that future generations of fly fishers are afforded the same opportunities. Serving with these individuals has been a blessing. I tell TIC teachers that there is one important difference that distinguishes TIC volunteers from doctors - we still make house calls. The fact that these individuals take the time to meet with and interact with teachers and students is what plays such a key role in the success of the program.

From behind the scenes my wife, Mary Susan, has supported and encouraged my efforts coordinating the Maryland TIC program. In addition, she generously allows me time to indulge my fly-fishing hobby.

The universe and planet earth do declare the creative hand of God. Fly fishing affords one the opportunity to travel to and visit some of the world's most beautiful and pristine places. There are few if any trout streams that fail this test. I believe God designed them this way to provide us a respite

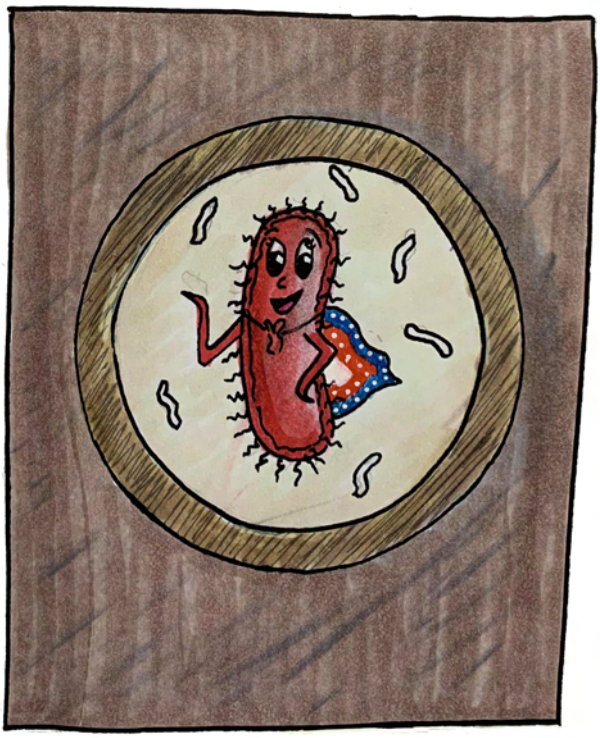
from the noise and clutter of daily life. As His stewards I hope this book encourages you to join me in protecting and defending these precious resources.

Note to Readers

When reading this story, you will come across words that when used for the first time are printed with **bolded** type. This indicates that you should refer to the Glossary at the end of the story for additional information about the word or topic. The exception to this is the names of characters in the story. Their names are always printed **bolded**.

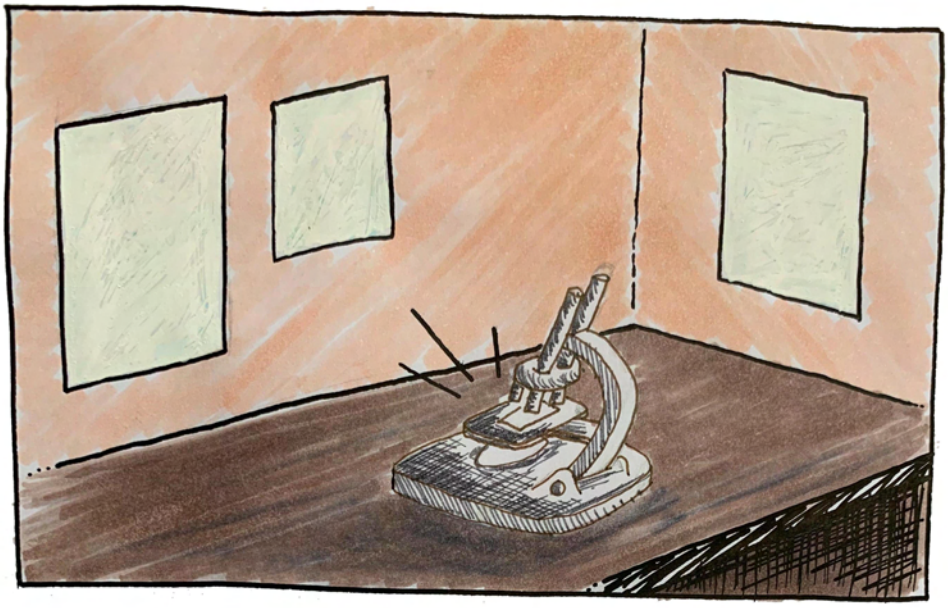
NITRO

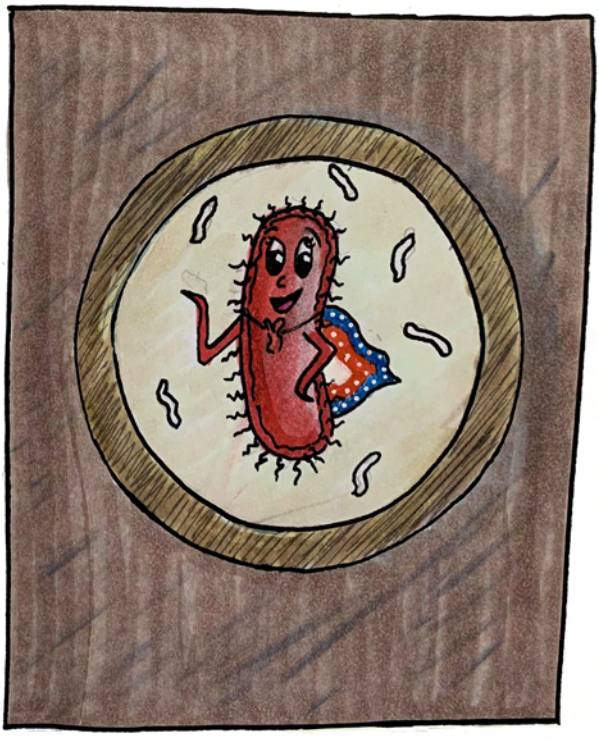
Trout in the Classroom Super Hero



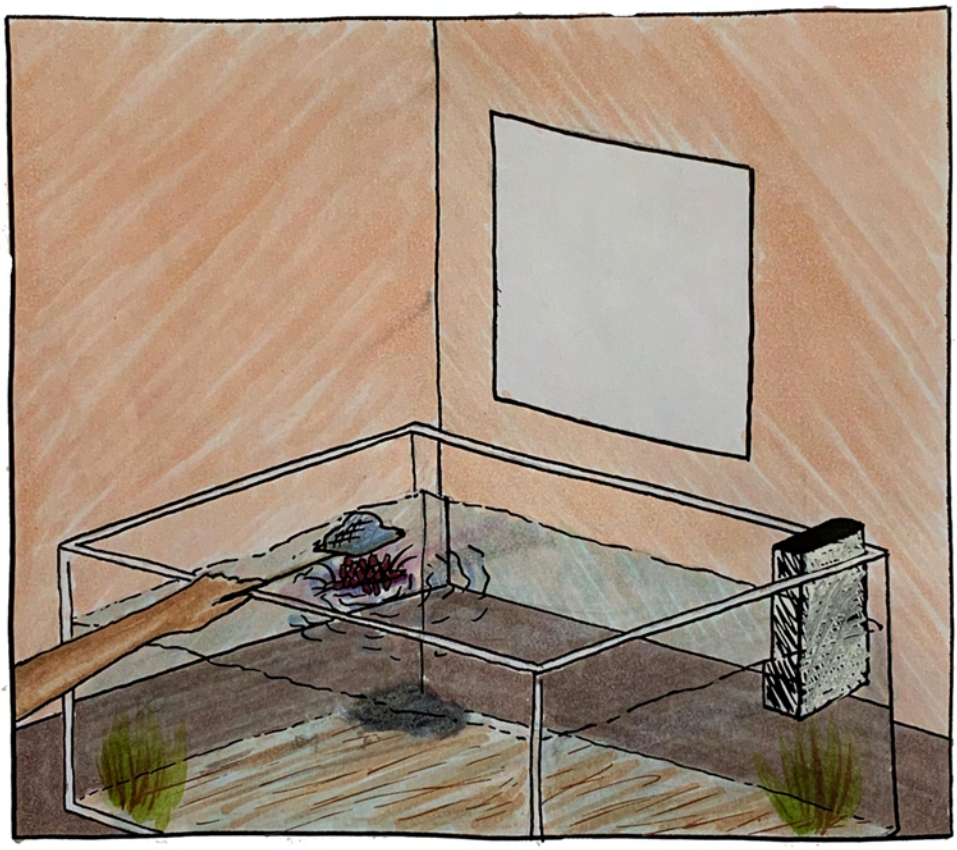
Hey, Trout in the Classroom students. Look, over here! No, over here, through your microscopes. Having a hard time seeing me? I'm **NITRO** – the TIC Super Hero! My Super Hero friends and I help you raise trout in your aquariums.

We're bacteria; very tiny bacteria





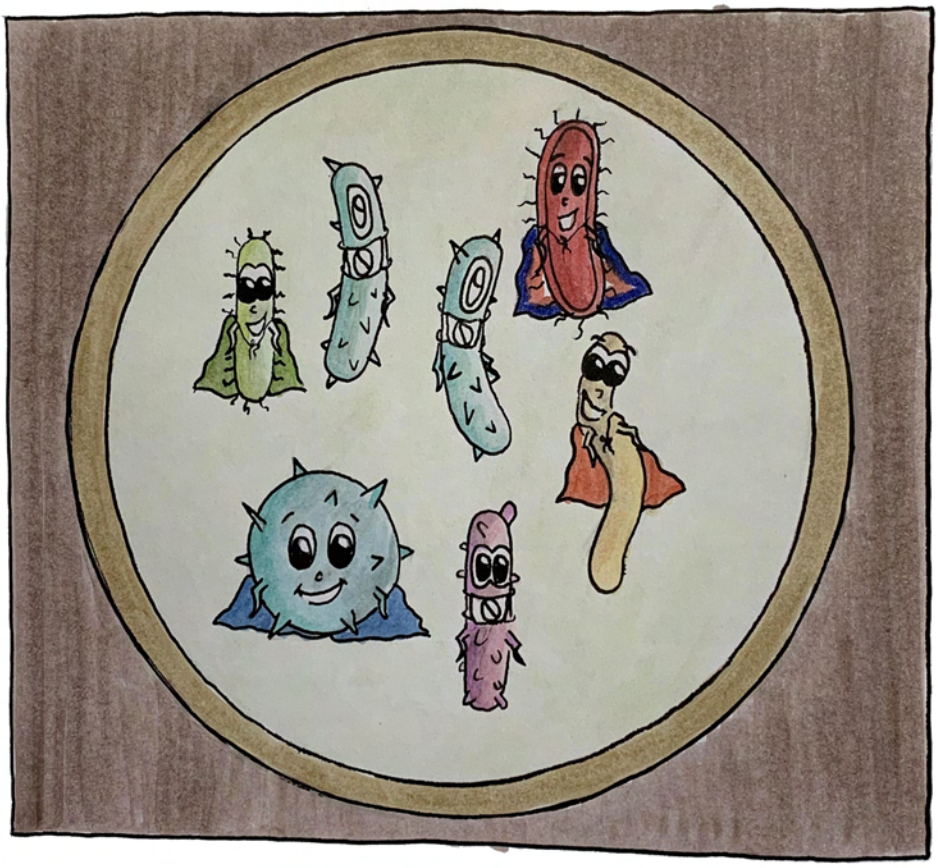
We may be small, but there's lots of us Super Heroes and adding us to your fish tanks is essential to the survival of your trout. Listen carefully. I have a short biology and chemistry lesson to share with you. It's all about how we Super Heroes maintain the water quality in your fish tanks.



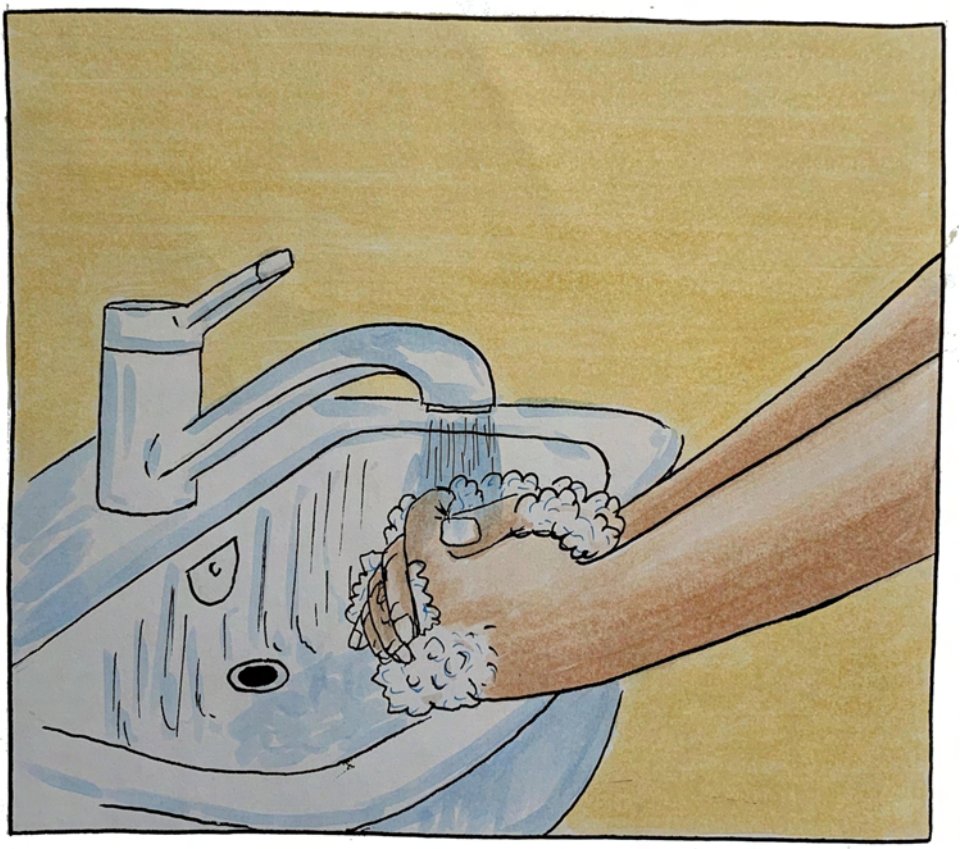
First of all, you need to know something about bacteria. We're tiny little organisms, but we're everywhere around you. You already know you can't see me without a microscope because we're so small. We live in the air, on your skin, in your bodies, in the ground, in your fish tank, and throughout nature.

We can survive in very harsh conditions including deep areas of the earth's crust and in radioactive waste.

There are around as many bacteria cells in a human body as there are human cells.

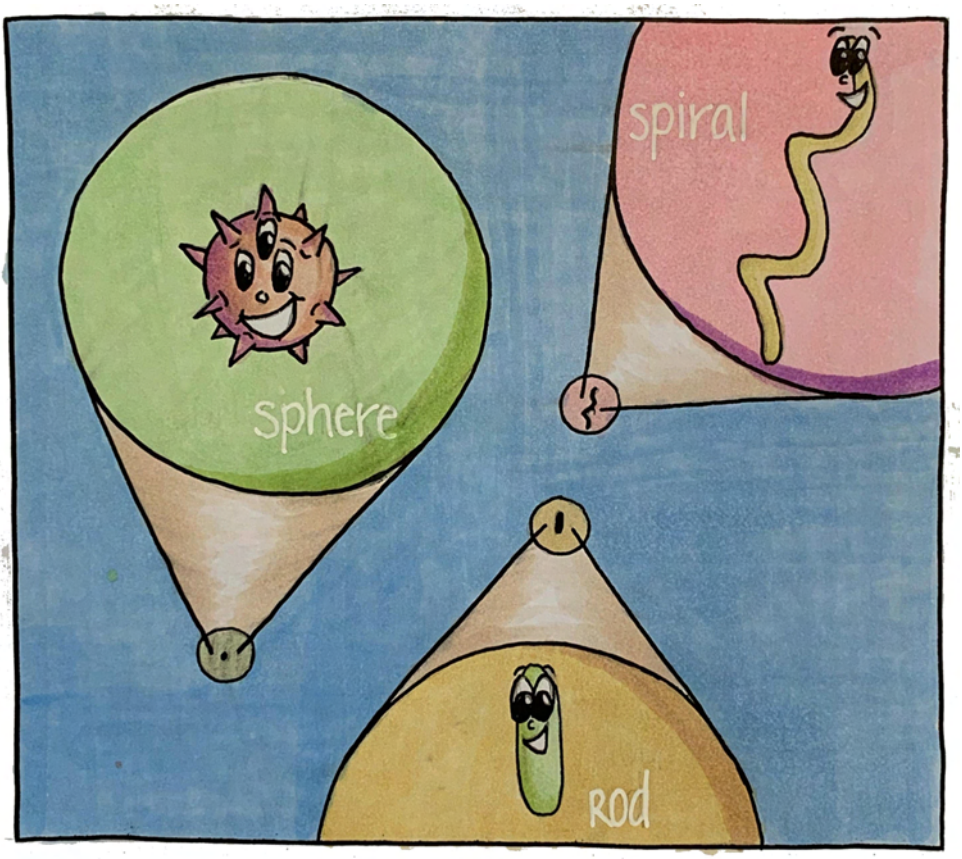


My guess is most of what you've heard about bacteria isn't good. It's true some bacteria can cause sickness and disease in plants and animals. Scientists and medical doctors have developed antibiotic medicines and products that help fight off bacterial infections. When you wash your hands with an antibiotic soap you kill harmful bacteria.

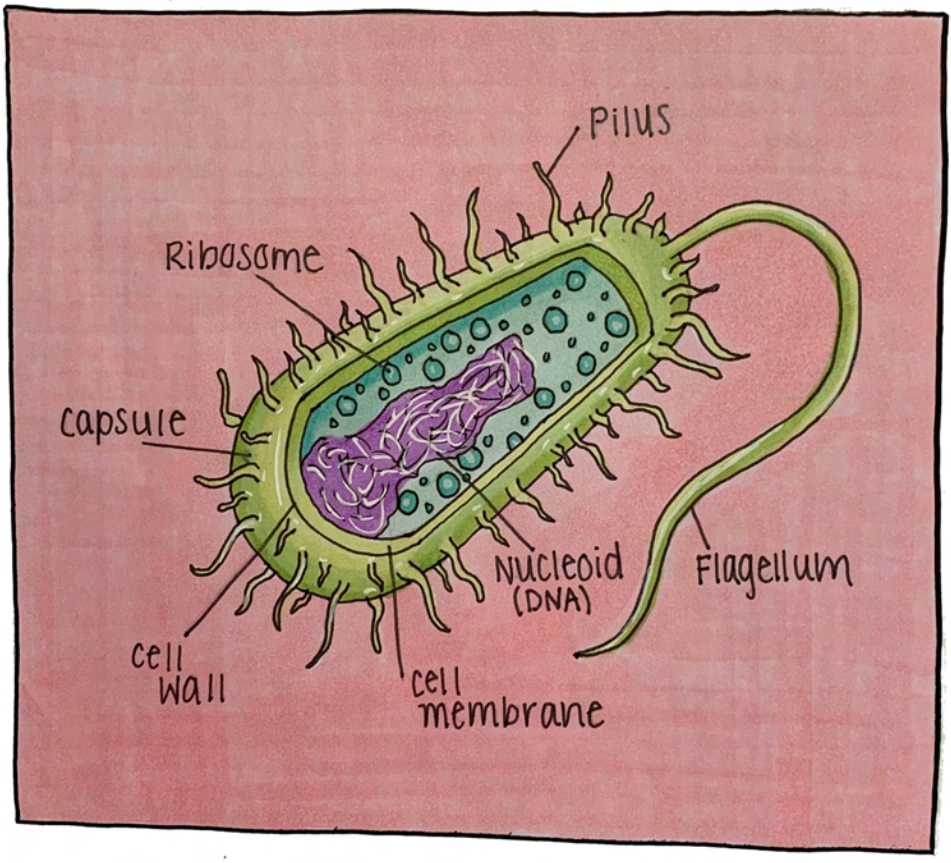


The good news is most bacteria aren't dangerous. As a matter of fact, if you've eaten yogurt, you've eaten bacteria. My Nitro friends and I play an important role in helping ensure the survival of planet earth, humans, and your trout.

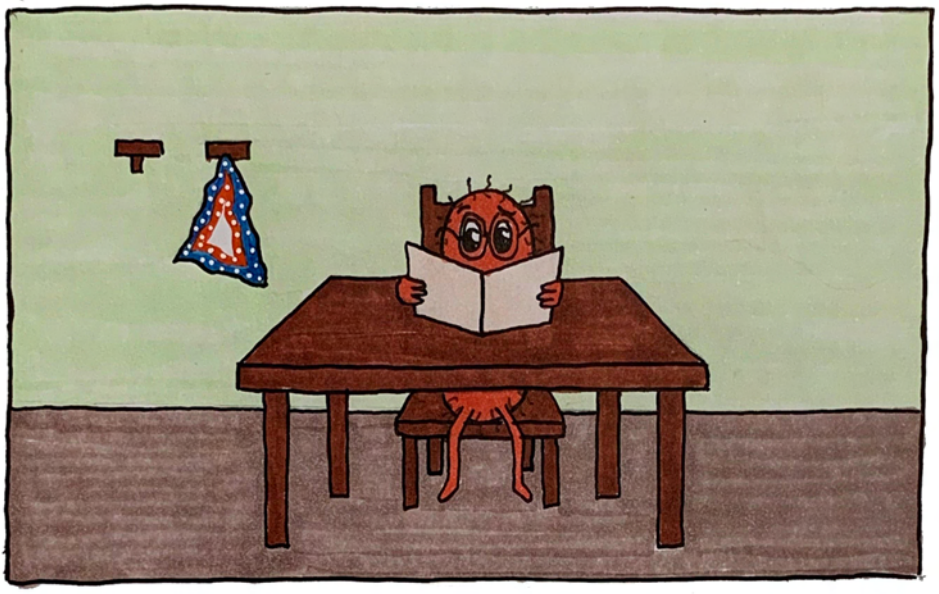
We come in all sorts of shapes including rods, spirals, and spheres. Some of us can even "swim" around using long tails called flagella.

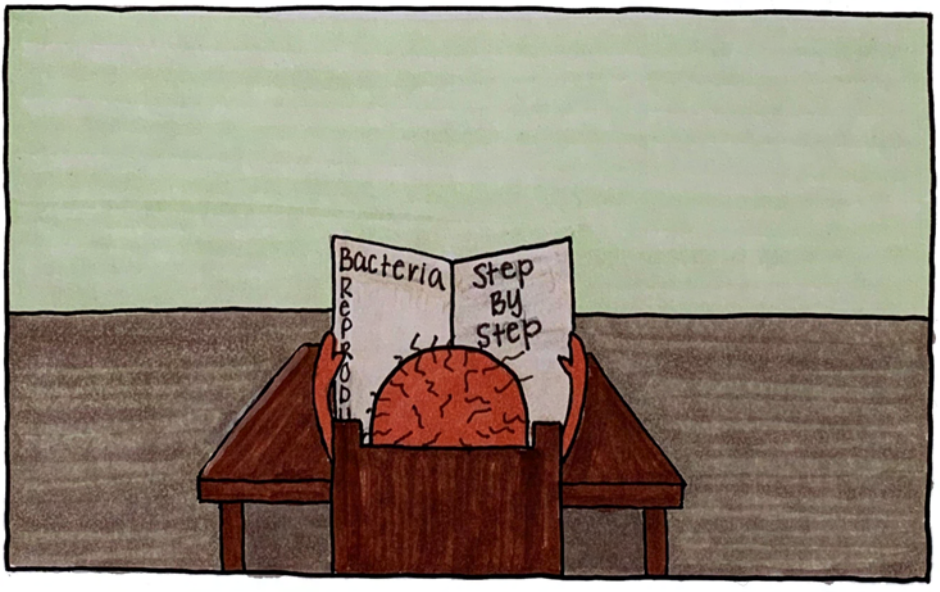


We're single-celled microorganisms. Our cell structure is unique in that we don't have a **nucleus** and most bacteria have cell walls similar to plant cells. The cell wall protects us. On the inside of bacteria, there is a gel that holds all the other parts we need to survive and reproduce.

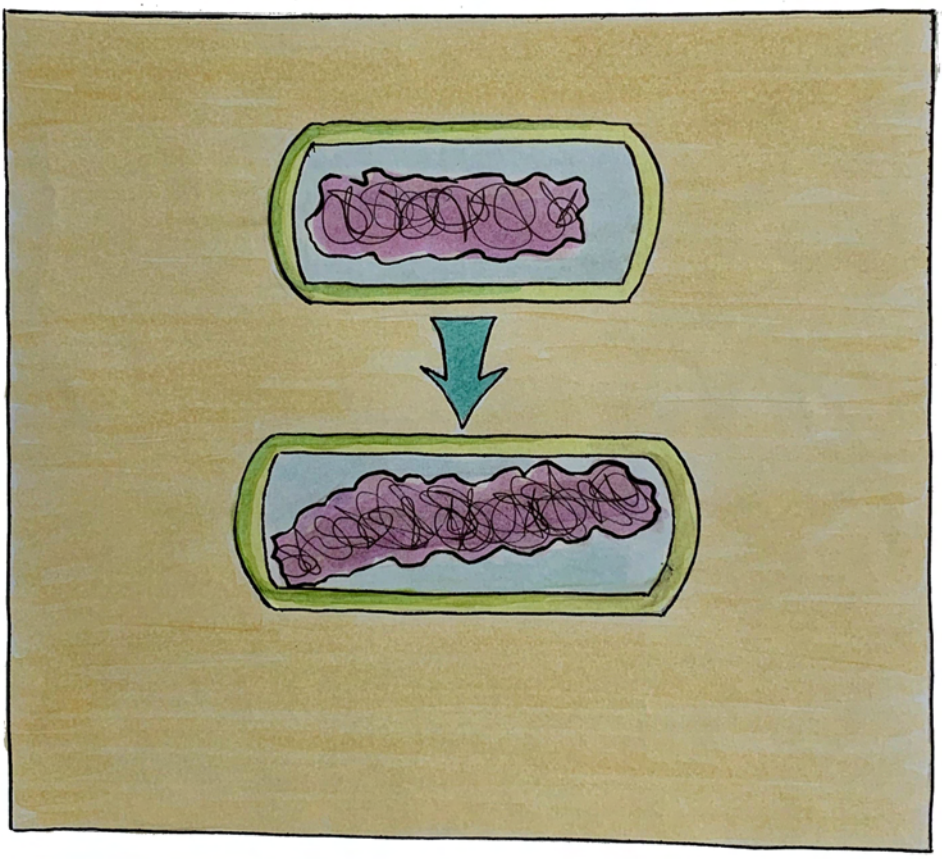


One of those important parts is **DNA**. DNA is something that every single living thing has. You can think of DNA as directions/instructions, or a blueprint. DNA tells cells what to do

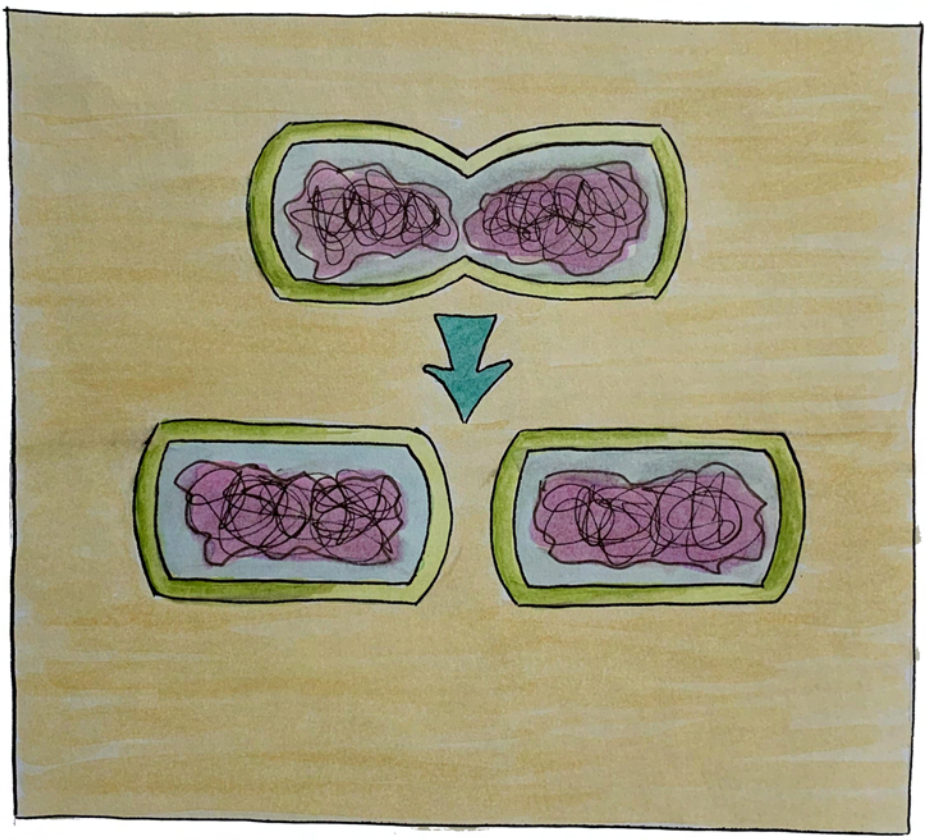




Bacteria reproduce by making an exact copy of ourselves. Most of us reproduce **asexually** by dividing in two, a process called **binary fission**. The first thing that happens in binary fission is that we make a copy of everything inside of us. Then the copies of all the stuff, including DNA, move to opposite sides of the cell.



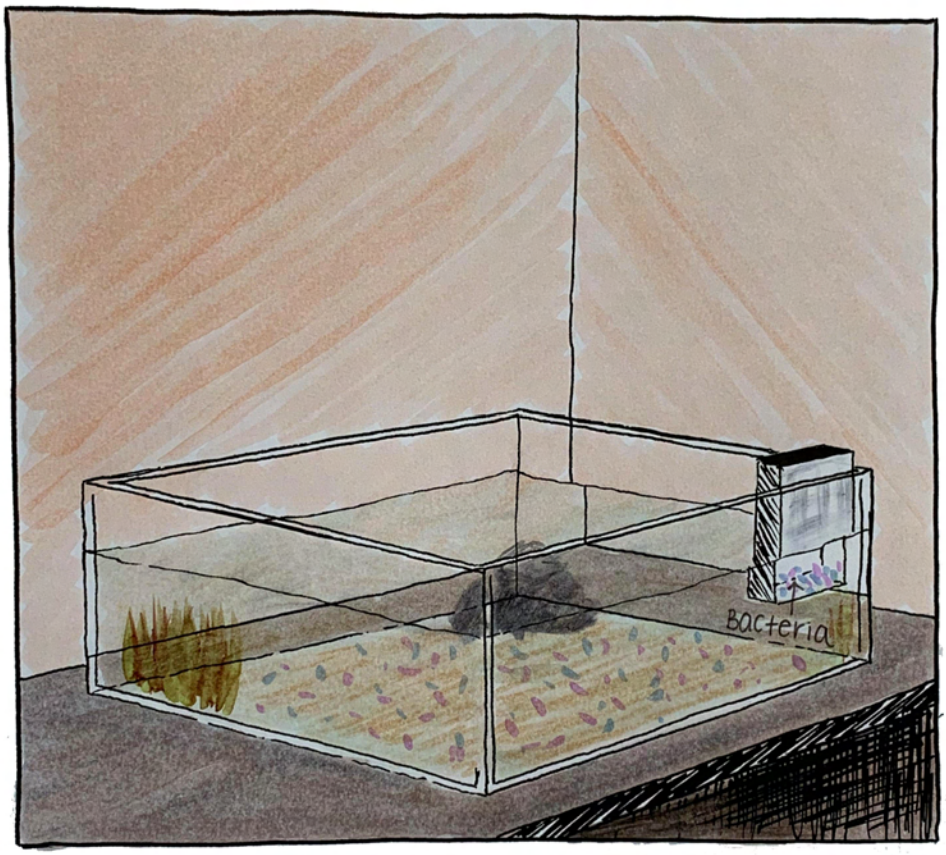
Once there are two sets of DNA and other important stuff, we create a new cell wall down the middle and the two halves start to pull apart. When the two halves have completely pulled apart, the new bacteria is called a **daughter**. The daughter bacteria is an exact copy of the original bacteria, or the **mother** cell. These two new cells grow, develop, and will likely each divide to form two new cells, resulting in a total of four cells with identical DNA from a single parent cell.



One thing of interest and importance to bacteria in your fish tank is that we are sensitive to temperature. Trout are **cold-blooded.** If you were to catch a trout and measure its body temperature you would discover it's the same as the water the fish was swimming in. As the water temperature gets colder, the rate at which trout **metabolize** (process) the food they've eaten also decreases. In cold water trout need less food to survive than in warmer water.

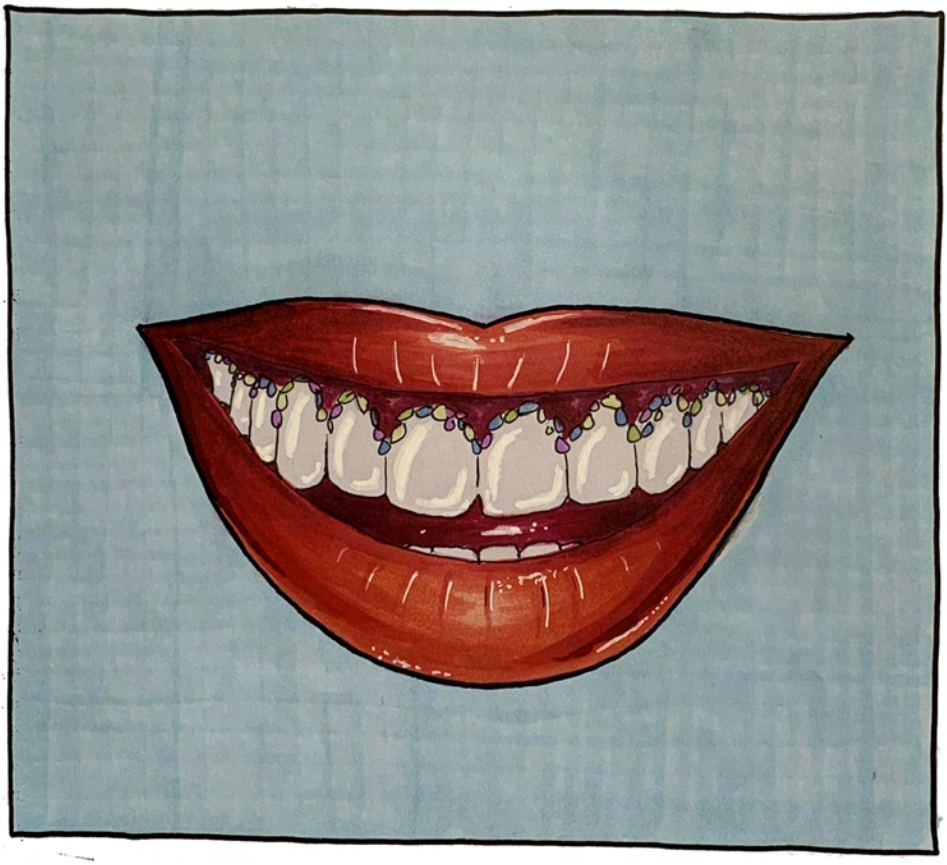
Bacteria are also sensitive to temperature changes. As the water temperature in your tank decreases, the rate at which we reproduce slows down. This is critical to how Nitro Super Heroes help your trout survive.

You're probably wondering where in your trout tank you need to look to find bacteria. The Super Hero bacteria in your tank make up what is called the **bio-filter**. Most of us live in media (charcoal, ceramic cylinders and foam) placed in the mechanical filter. If your tank has a gravel bottom some of us will reside there.



Colonies of reproducing bacteria form **biofilms**. These films are very complex and consist of multiple species of microorganisms. Living in a biofilm provides protection and allows sharing of nutrients and genetic material. An example of a biofilm most students are familiar with is dental plaque that can form on teeth.

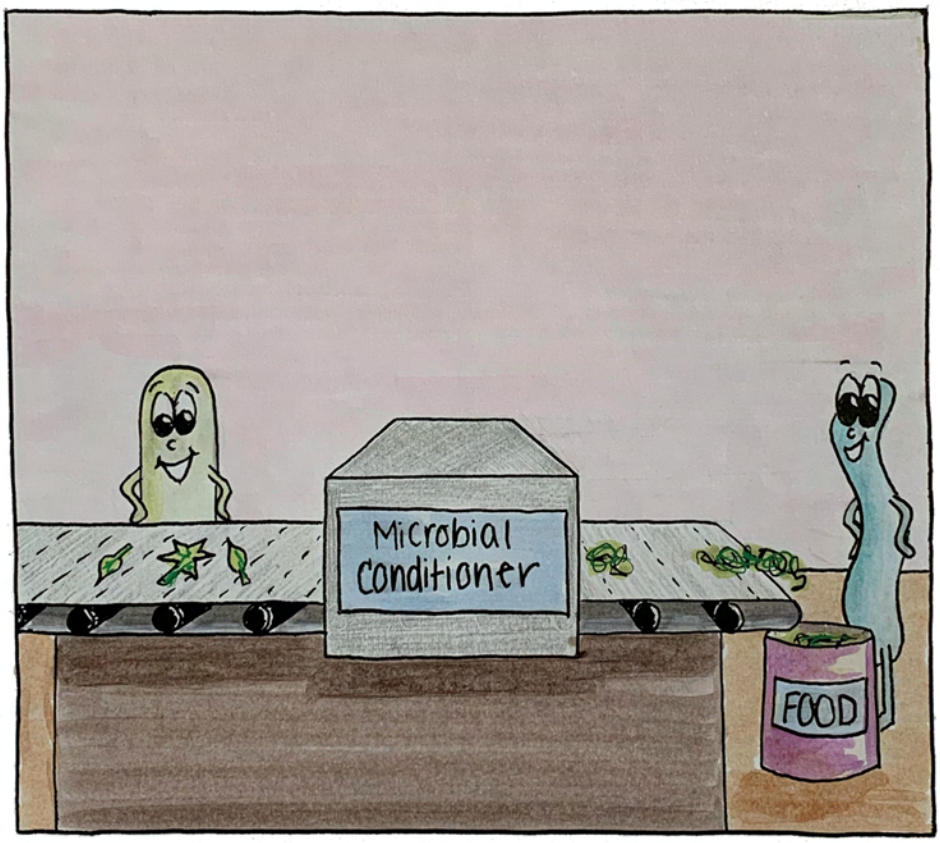
Biological processes, such as **decomposition** (organic breakdown**), nitrification** and **denitrification,** are also carried out in bio-films. I'll explain more about these as you continue reading.



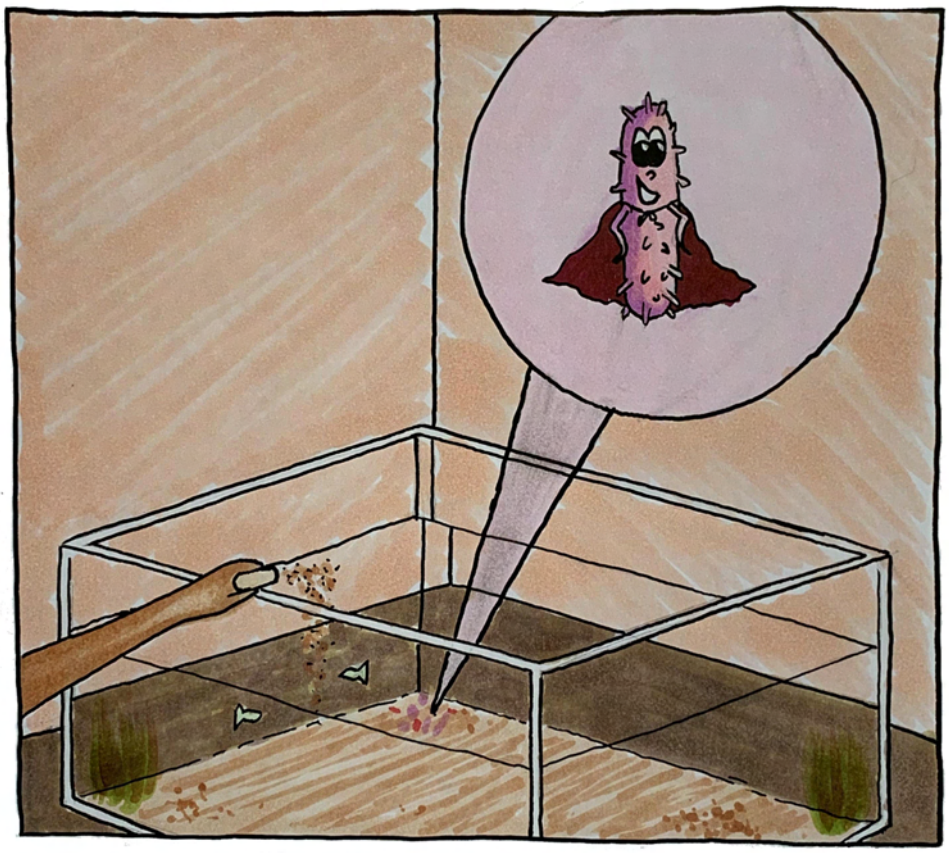
Those of you who have previous Trout in the Classroom experience know about **macroinvertebrates**. Aquatic macros live in lakes and streams. These insects do not have a backbone (invertebrate) and can be seen without the aid of a microscope (macro).

A major food source for many freshwater macroinvertebrates are leaves. However, before these aquatic insects can ingest leaves the tough plant matter has to be softened. A process called **microbial conditioning** softens leaves by breaking down complex chemical compounds, such as **lignin** and **cellulose**.

This takes place in bio-films colonized by bacteria, fungi and algae. Not only does the biofilm help to breakdown the leaf; the biofilm itself becomes an importance food source for the macros.

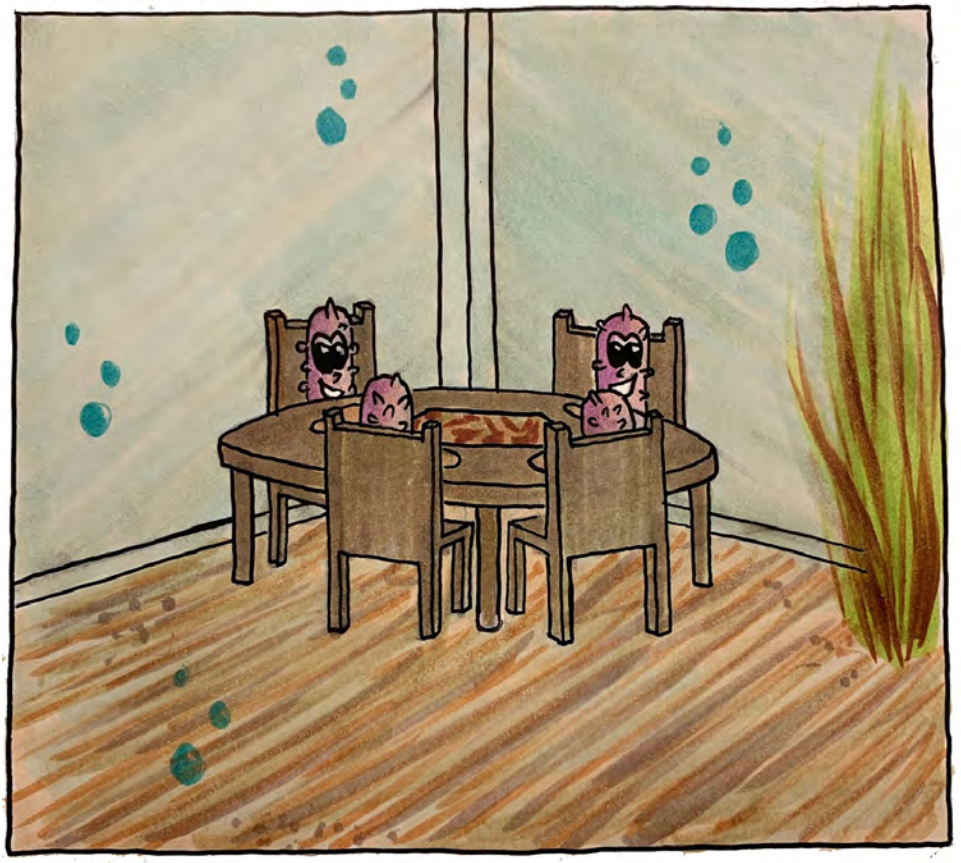


Like you, your trout need to be fed regularly to grow and thrive. This is when the Nitro Super Heroes take charge. Let me introduce you to **SAPRO**. **SAPRO's** family is made up of bacteria which scientists call "**waste degraders**". The scientific name for these bacteria is Saprophytic.



In the filter of your fish tank saprophytic bacteria form colonies in the bio-film and feed on fish waste and uneaten food. This helps break down waste into **inorganic** and **organic** byproducts including ammonia.

If this waste was left to accumulate in your tank the water quality would deteriorate and the trout would die. This process is called **decomposition.** This is also the first step of the **Nitrogen Cycle.**



Chemists use a type of mathematical formula or shorthand to express what happens during this first step:

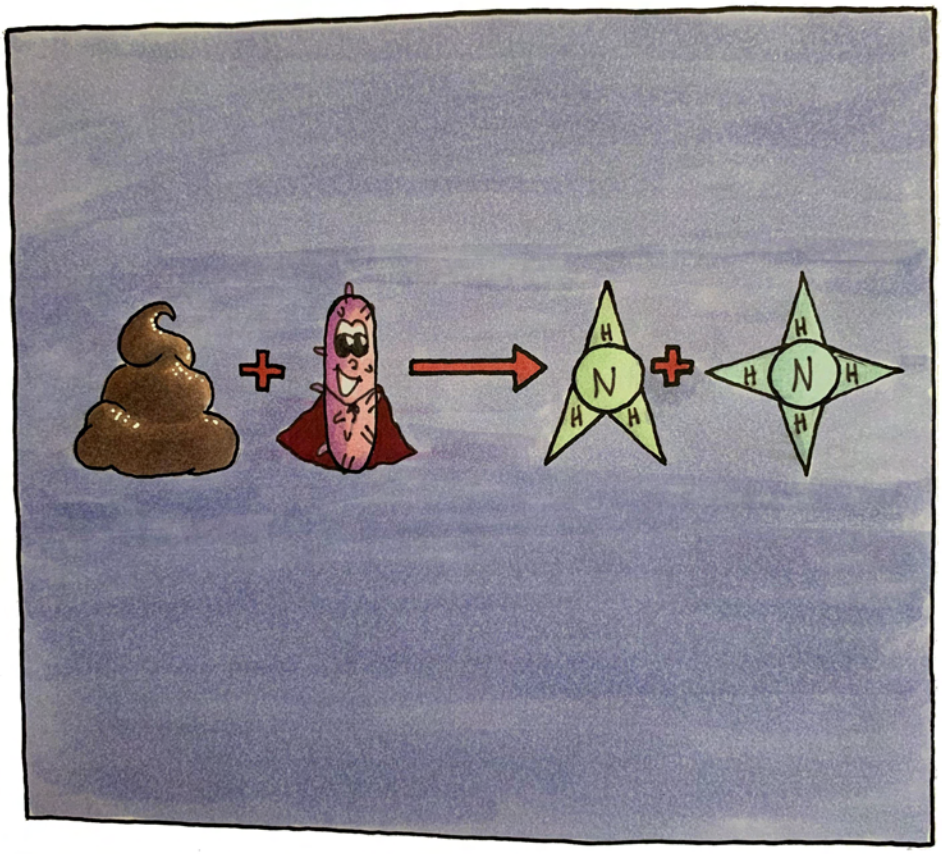
Organic waste(fish/food) + Saprophytic bacteria

Yield

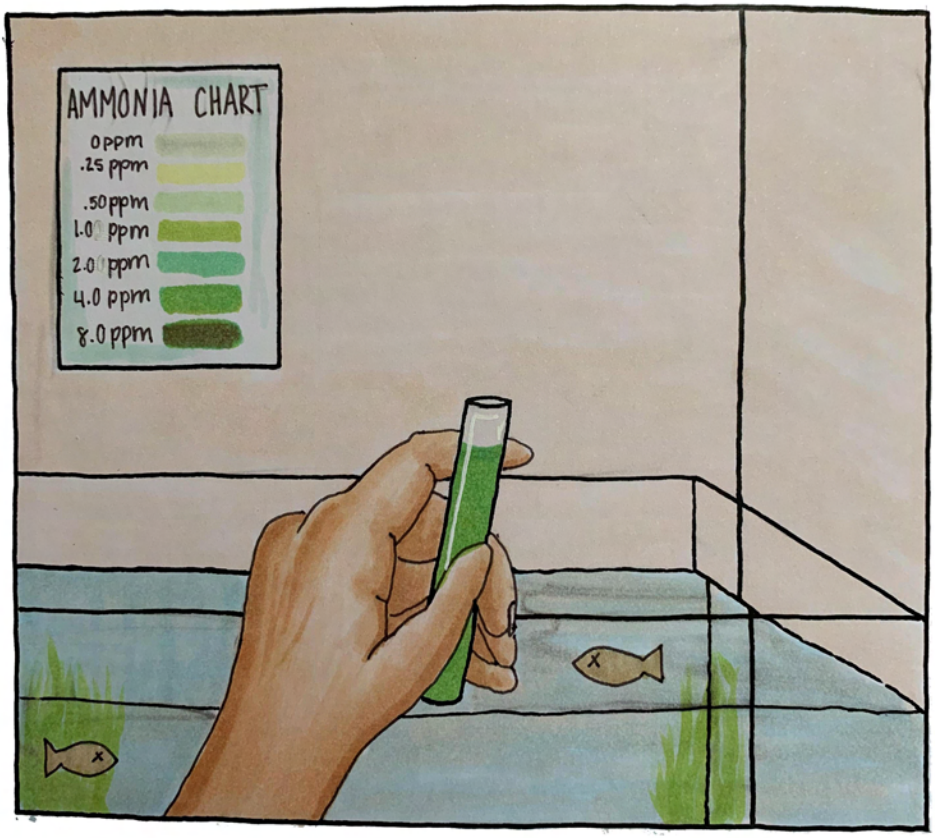
NH_3 NH_4

Ammonia + Ammonium

Here's a picture of this using atoms and molecules

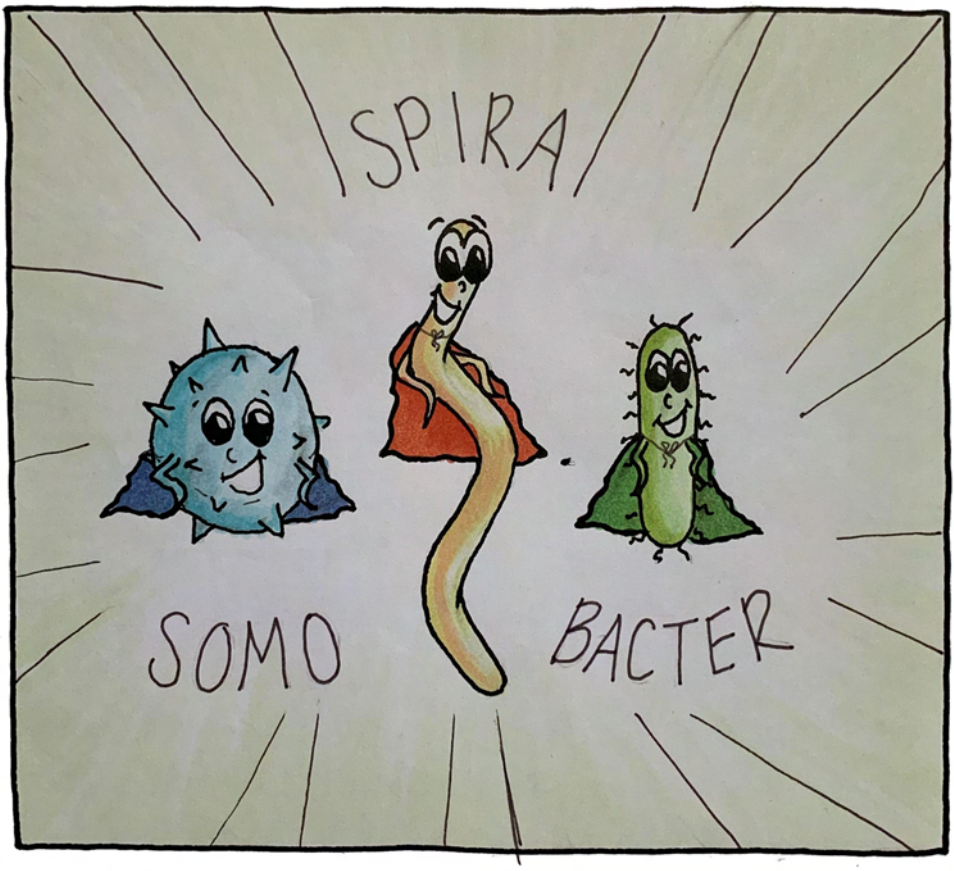


The buildup of ammonia formed by saprophytic bacteria during the decomposition of fish and food waste can kill your trout. Ammonia is also a waste product of a fish's protein metabolism and is excreted by their gills directly into the water. Ammonium, another by-product of decomposition, is not harmful to your fish.



As ammonia levels in your trout tank begin to increase three more of my Nitro Super Hero friends spring into action - **NITROSOMONAS NITROBACTER.** and **NITROSPIRA**

You can call them **SOMO**, **BACTER and SPIRA ;** I do.

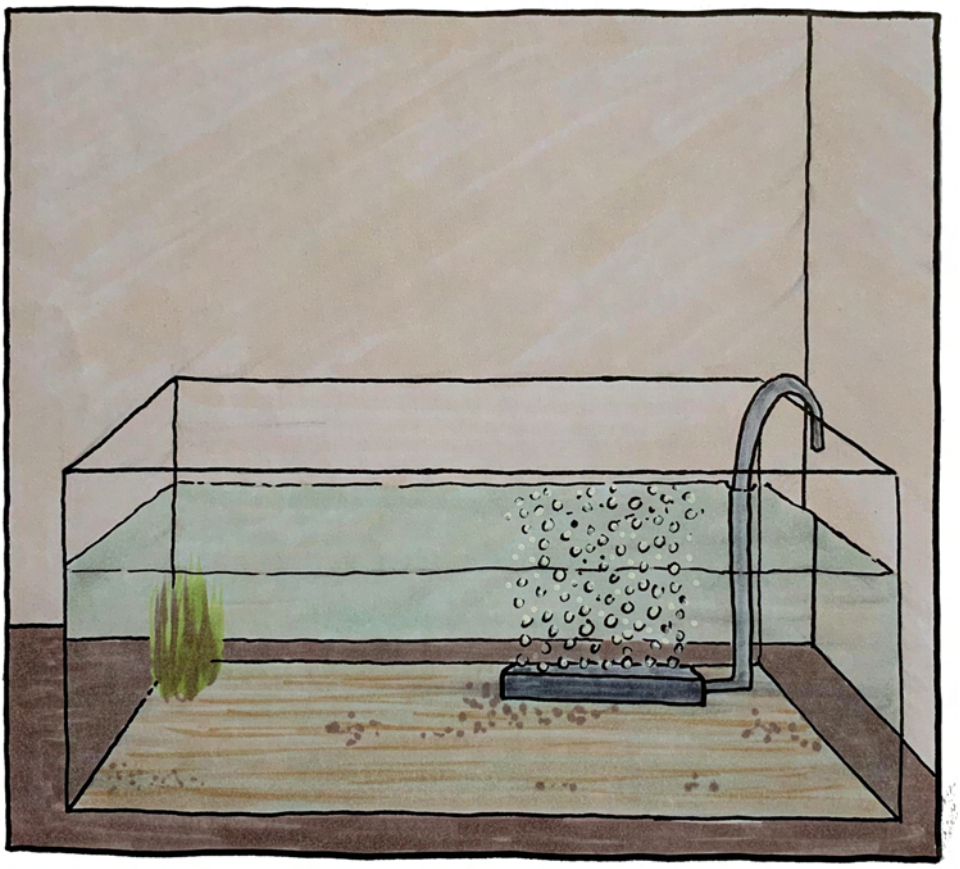


Nitrosomonas is type of spherical to rod-shaped bacteria. These bacteria use oxygen in the tank water to convert ammonia and ammonium into nitrite, a process known as **nitrification**. How does oxygen get into the water in your tank?

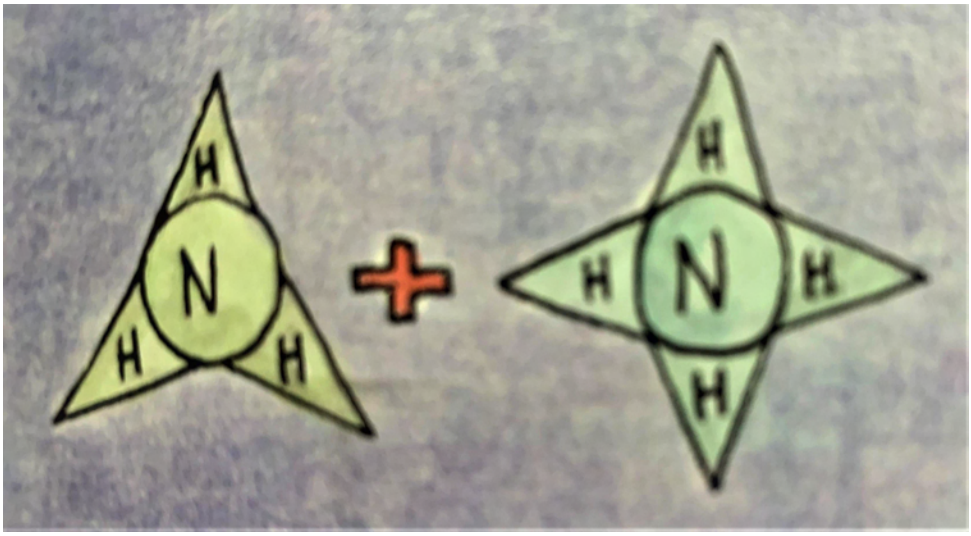
Dissolved oxygen nearly always comes from the mixing of oxygen in air with the water. In a stream this takes place when flowing water encounters obstacles such a rocks and tree branches. This is sometimes called "white water".

In your fish tank this takes place at the air/water interface. The bubbler (air stone) in your tank and the outflow from your filter are areas where mixing occurs. This helps provide an oxygen-rich environment for your fish and bacteria.

Another interesting fact is as the temperature of water is decreased, the amount of oxygen dissolving in the water increases. The cold-water trout like holds more dissolved oxygen than room temperature water.



A molecule of ammonia (NH_3) has three Hydrogen atoms connected to one Nitrogen atom; a molecule of ammonium (NH_4) has four Hydrogen atoms connected to one Nitrogen atom.



O_2 is the symbol for a molecule of oxygen and NO_2 is the symbol for a molecule of nitrites

Chemists might use something like this expression to describe what occurs when ammonia and ammonium are converted to nitrites by **SOMO** bacteria.

NH_3 NH_4 O_2

Ammonia/Ammonium + Oxygen + Somo Bacteria

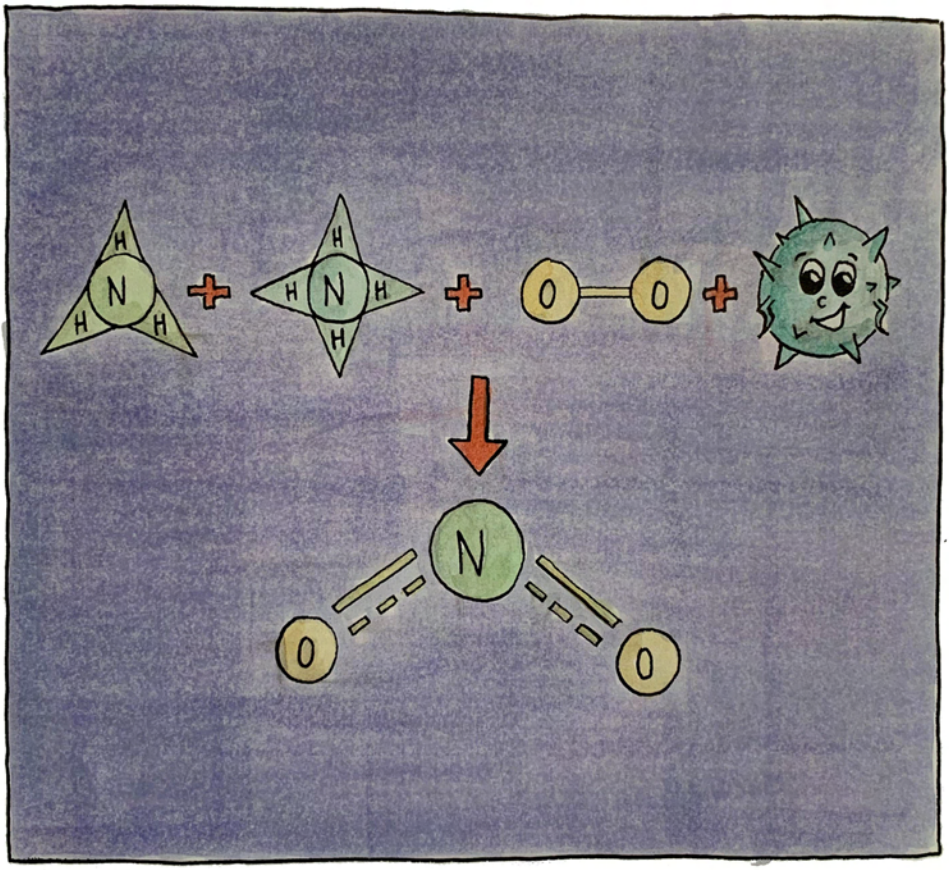
Yield

NO_2

Nitrites

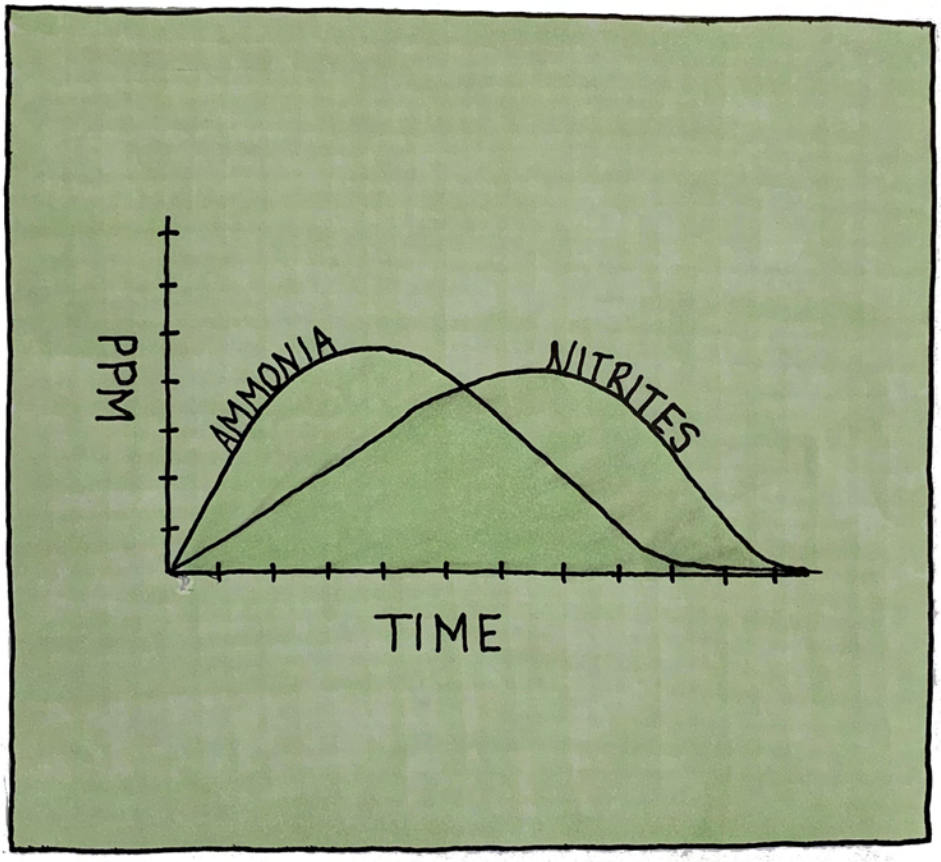
A picture of this process looks like this:

This is the second step of what takes place in your fish tank during the Nitrogen Cycle.

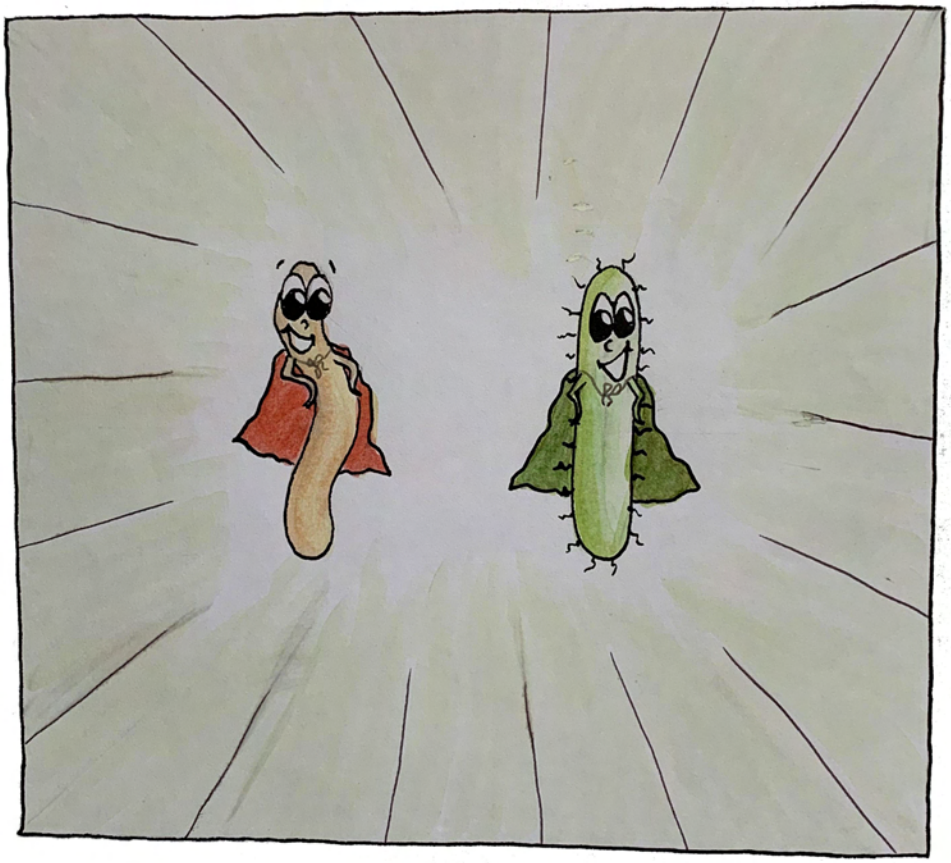


At this point in time, as you run daily water chemistry measurements for your tank, you should begin to see ammonia levels starting to decrease as colonies of **SOMO** Super Heroes convert ammonia into nitrites. However, nitrites are also very harmful for your trout and also need to be controlled in your aquarium.

Graphs are often used to illustrate how ammonia levels in parts per million (PPM) reach a peak in your tank while reproducing colonies of **SOMO** bacteria begin to convert them to nitrites. When this occurs, the ammonia level begins to decrease and the nitrites level starts to increase.

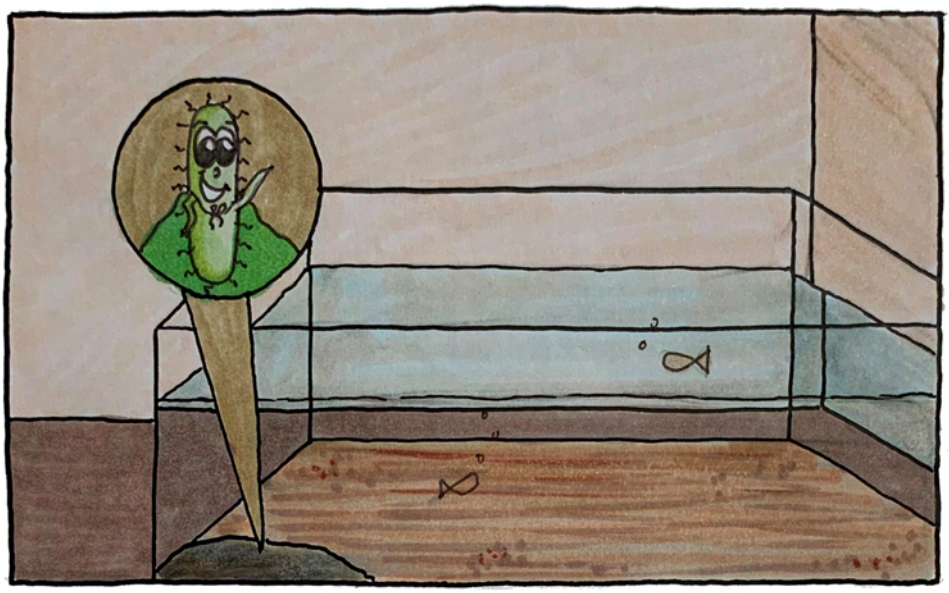


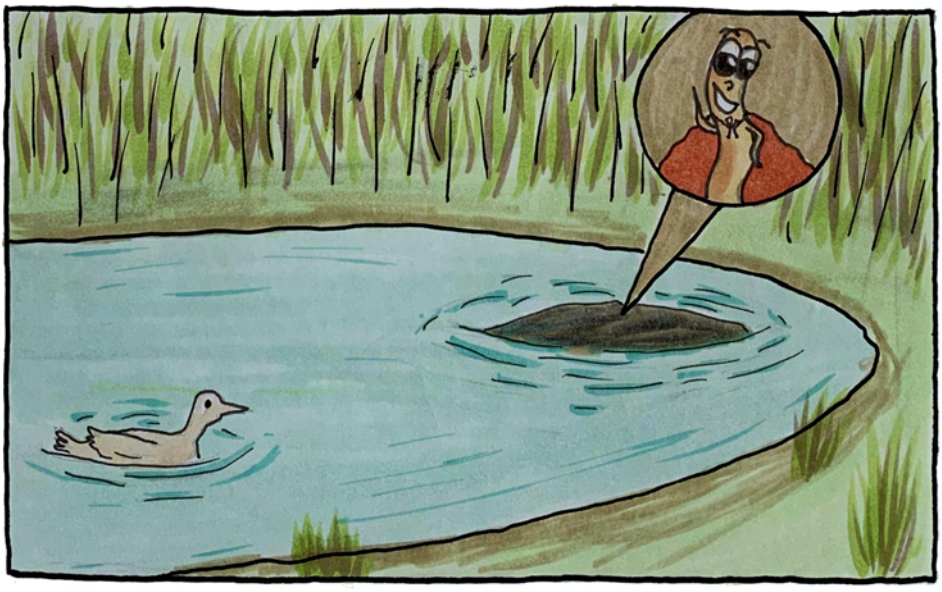
To the rescue come **BACTER** and **SPIRA** (Nitrospira and Nitrobacter bacteria). Biologists who study bacteria are discovering new and interesting facts about these two Super Hero friends of mine and have added much to the knowledge base in the past 10 years.



Current research indicates that both **BACTER** and **SPIRA** convert nitrites into nitrates, however, which one dominates seems to depend on the oxygen content of their surroundings. **BACTER** likes the higher oxygen conditions normally found in tanks in which you raise trout. **SPIRA**, on the other hand, does most of its work in lower oxygen situations such as lakes and ponds.

This information is important to keep in mind when you're selecting the bacteria products you add to your trout tanks.



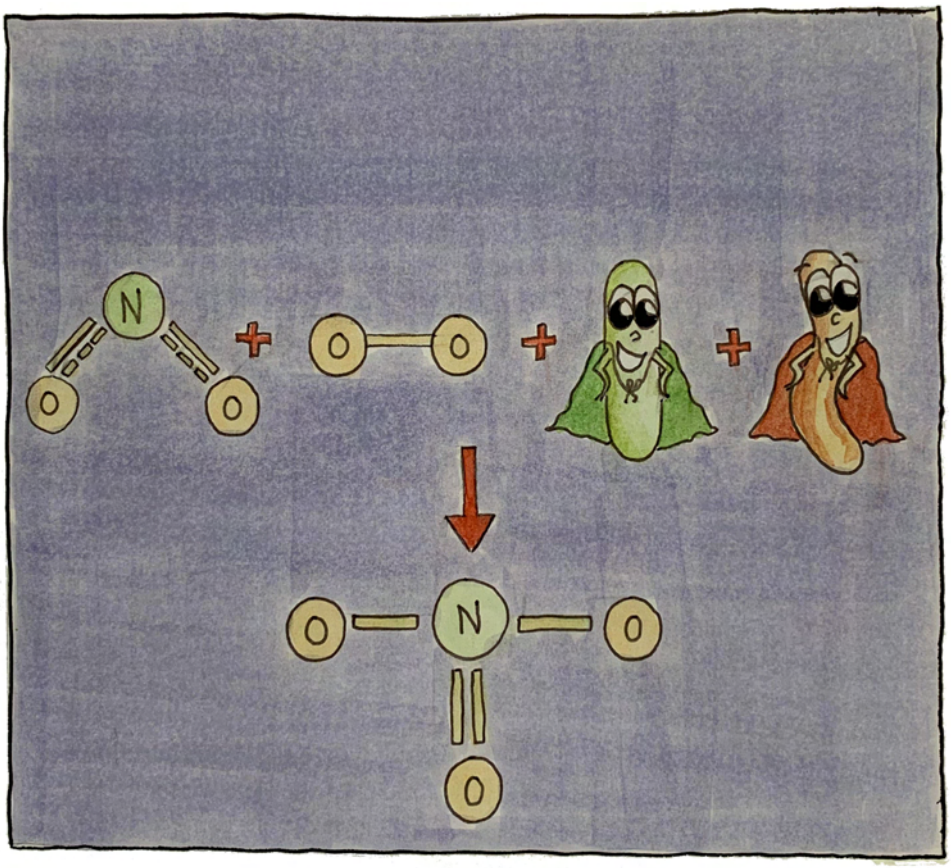


The conversion of nitrites to nitrates can be represented as follows: This is the third step of the Nitrogen Cycle

The Chemistry method:

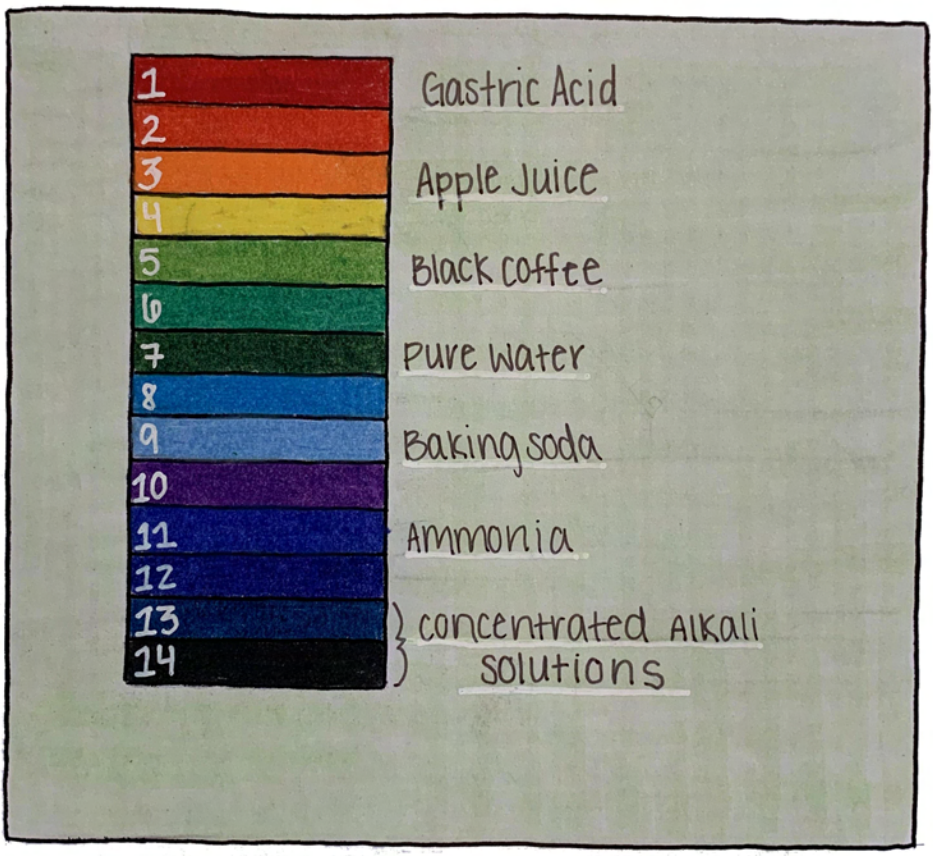
Nitrites(NO_2)+ Oxygen(O_2) + Nitrobacter/Nitrospira Bacteria ————> Nitrates(NO_3)

A simpler way using a picture:



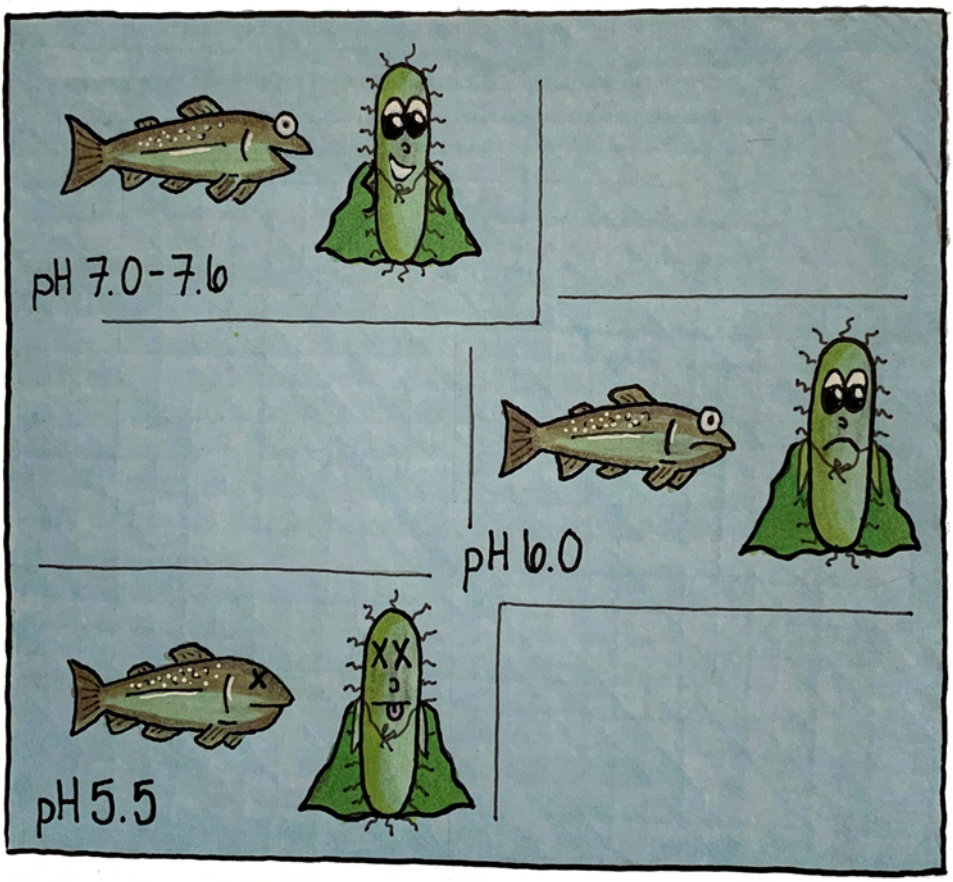
Just like **SOMO**, **BACTER** and **SPIRA** reproduce more slowly as the temperature decreases. All of my Nitro friends are also influenced by another water quality parameter you measure every day- **pH.**

If you haven't studied pH it's a measure of the acidity of your tank water. The pH scale has a range of 0-14. A value of 7 is considered neutral; neither acidic (below 7), nor basic (above 7). Nitro Super Heroes do our best work if the pH is around 7.0 – 7.6. This pH range is also ideal for trout growth.



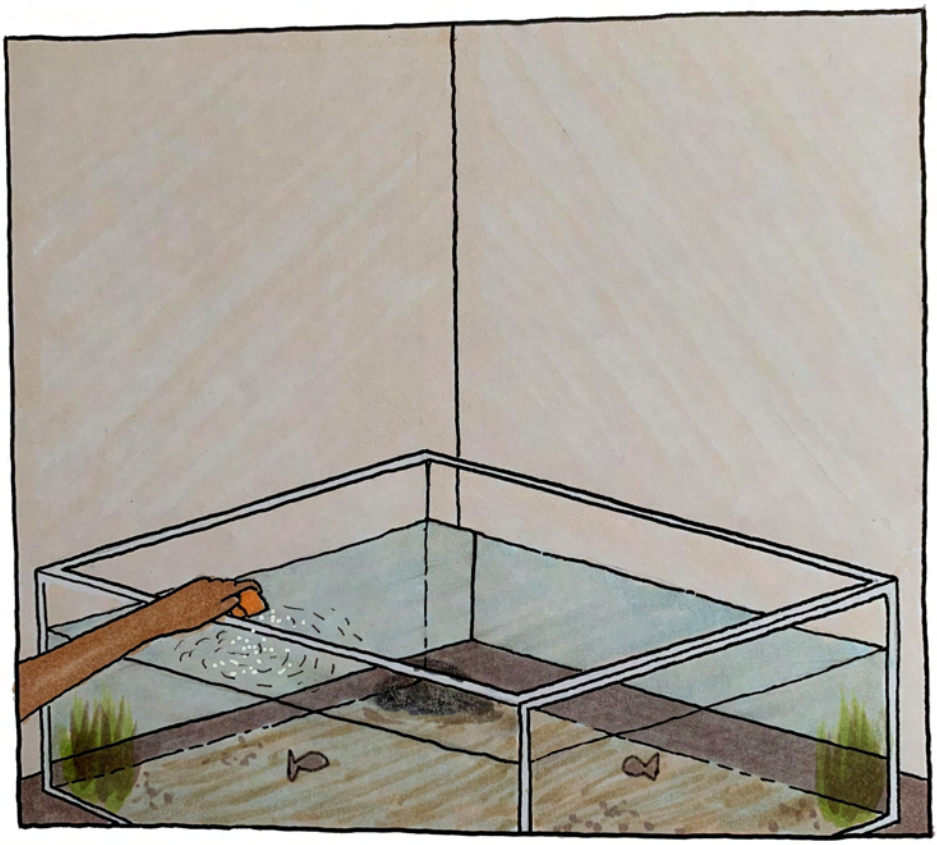
pH of Common Substances

If the pH gets too low, around 6.0, your Super Hero bacteria will stop reproducing. This means that ammonia, nitrite, and nitrate levels in your tank will start rising. If any of these substances get too high trout will start to die. If the pH gets as low as 5.5 we'll start dying. That's really bad for your fish!



If you begin to notice the pH in your tank starting to decrease this could be a signal that the **carbonate alkalinity** (**KH**) of your tank has fallen below 100 ppm (parts per million).

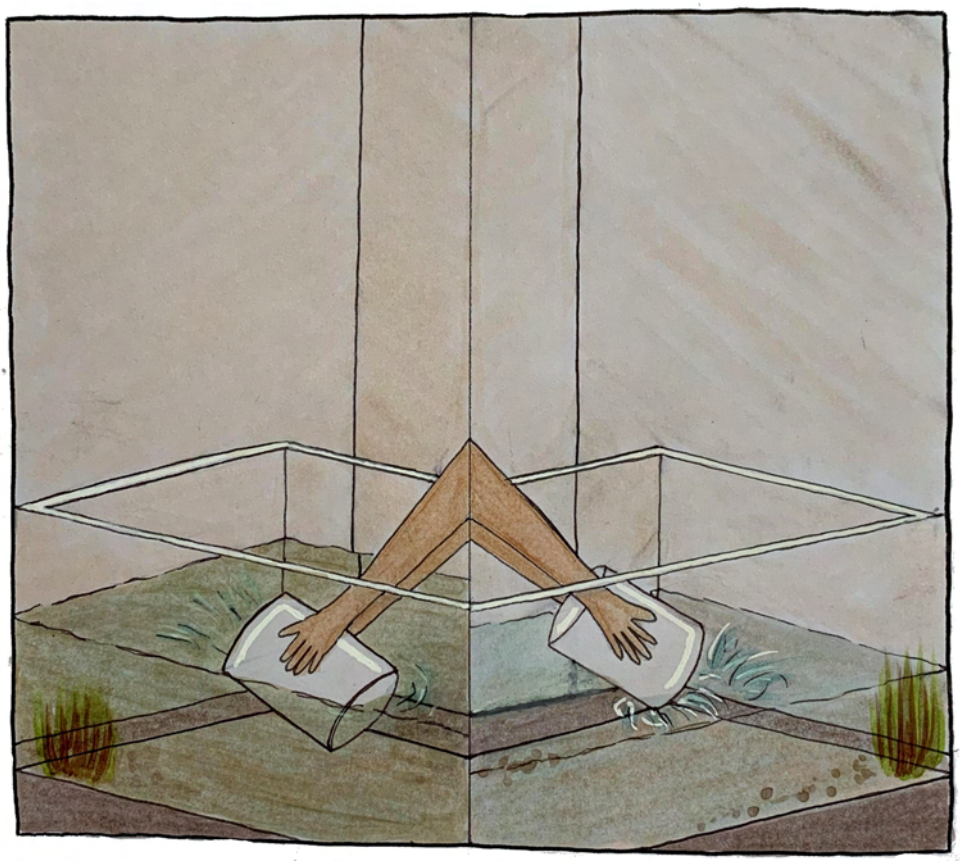
How do you fix this problem? Check your Trout Manual. Most suggest adding baking soda (**sodium bicarbonate)** to increase the KH. Baking soda has a pH of 8.4. It also provides carbon as an energy source for your bacteria.



What's next? At this point your ammonia and nitrite levels should be low (below 0.5 ppm). However, you'll most likely be experiencing an increase in nitrates.

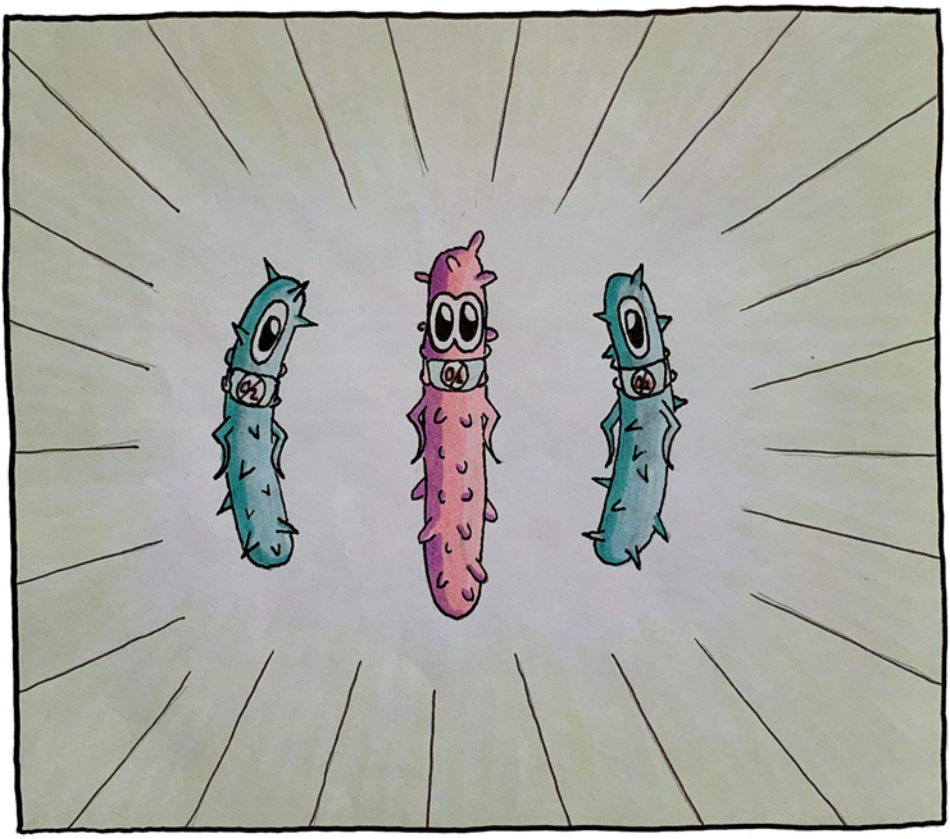
Fortunately, nitrates are not as harmful to your trout as are ammonia and nitrites. You still need to monitor them daily using your water quality test kit.

If nitrate levels exceed 40 ppm one of the best ways to reduce them is by performing a partial water change in your tank. Generally replacing 5-10 gallons of water weekly is all that's needed.



There is one other way my Nitro Super Heroes can reduce the nitrates in your tank, but it requires a very special type of bacteria. These bacteria need an environment in which oxygen is nearly absent. The name for this is **anaerobic.**

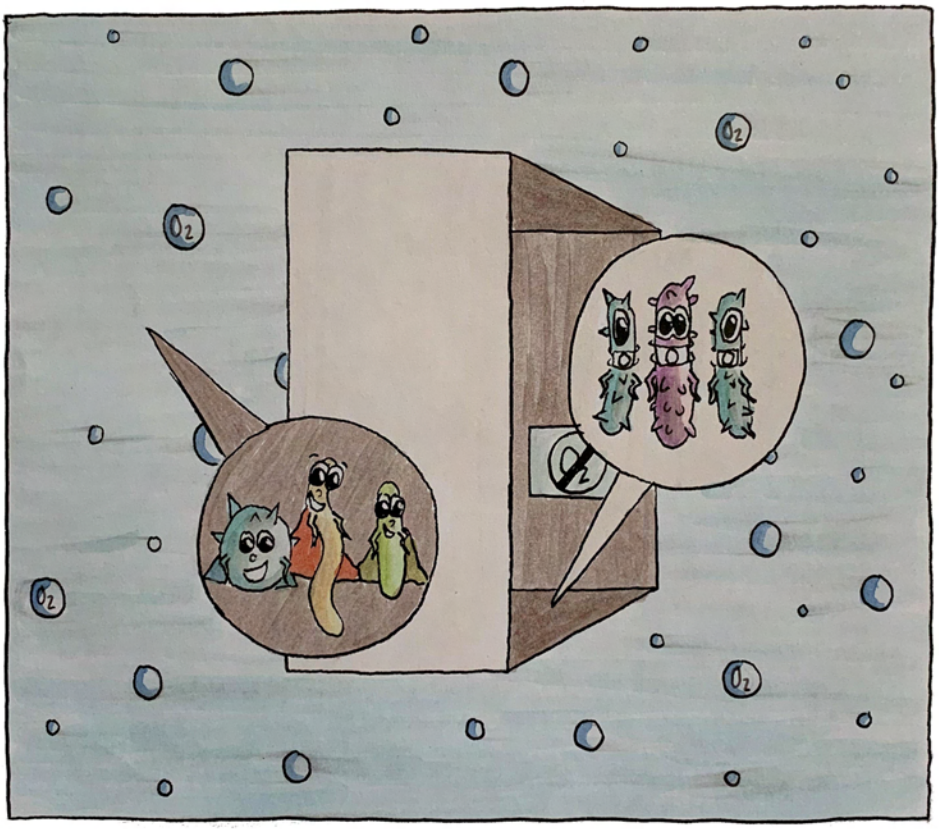
This is opposite the oxygen requirement of my other bacteria friends. Since I mentioned previously that your trout tanks generally have high levels of oxygen, you may be wondering if these bacteria could even survive in your tanks.



That is a tough question to answer. Remember the bio-films that develop in your tanks? These are the regions in the filter and gravel (if your tank has any) where bacteria live and reproduce.

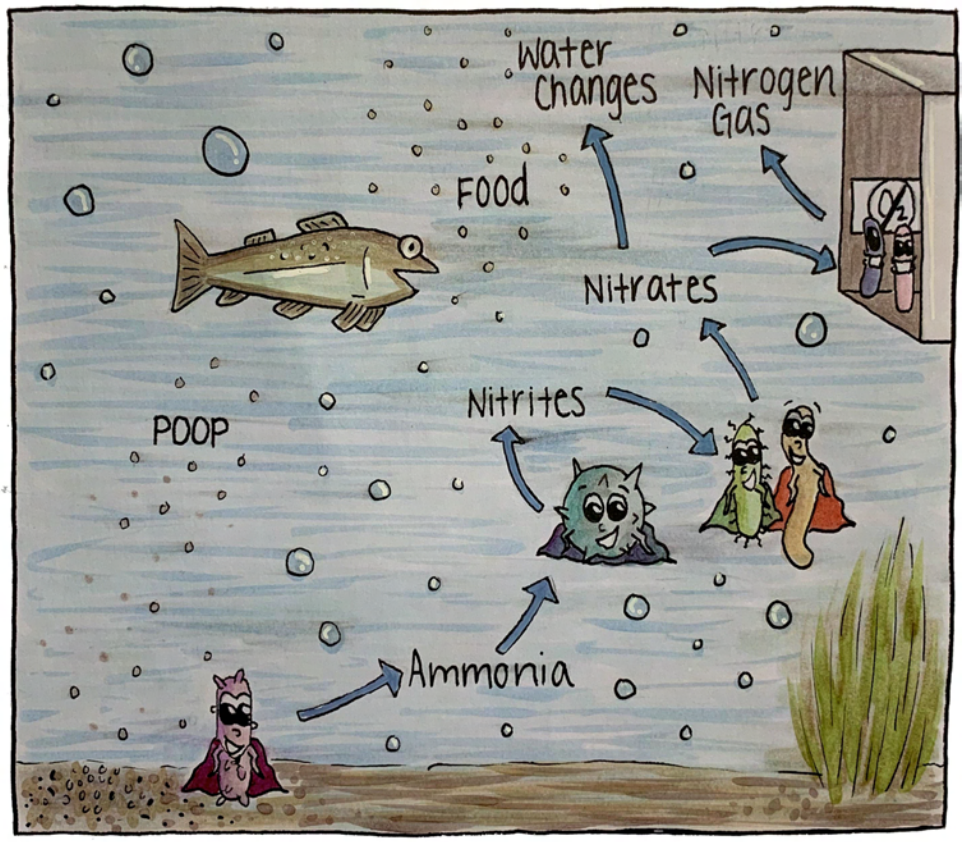
When my Nitro Super Hero friends check in with me rarely have they seen any anaerobic bacteria. On those occasion when they have, they tell me these bacteria reside in the inner portions of bio-films where there is very little oxygen.

If the concentration of O_2 in this area is less than 10% and there is a source of carbon to provide their energy, then these bacteria can remove aquarium nitrates by converting them to nitrogen gas that bubbles harmlessly into the atmosphere.



Generally, you cannot count on this happening in your trout tank. This process, when it occurs, is called **denitrification**.

This is the final step of the **Nitrogen Cycle** in your tank. I suggest you make a note of this as I hear this question sometimes shows up on a quiz your teacher may give you.



Nitrogen Cycle in a Fish Tank

I hope you've enjoyed meeting me and my Nitro Super Hero friends. Add us to your tanks, keep us fed (KH), check the pH levels, don't chill us too much, and provide proper O_2 levels.

That's a lot to ask of you, but I know you're up to the task. And when you do, all of us Nitro Super Heroes will do our best to help you raise healthy trout.

Bye for now. Your Super Hero friend, **NITRO**.

Glossary

Anaerobic bacteria – Bacteria which live, are active, or occur in the absence of air or free oxygen.

Asexually - Asexual reproduction is a type of reproduction by which offspring arise from a single organism, and inherit the genes of that parent only; it does not involve the fusion of gametes (a mature male or female germ cell) and almost never changes the number of chromosomes. Asexual reproduction is the primary form of reproduction for single-celled organisms such as bacteria.

Binary fission - A type of asexual reproduction wherein a cell divides giving rise to two cells, each having the potential to grow to the size of the original cell.

Biofilm - Biofilms are densely packed communities of microbial cells that grow on living or inert surfaces and surround themselves with secreted polymers. Many bacterial species form biofilms, and their study has revealed them to be complex and diverse.

Biofilter - A fully functioning biological filter, or biofilter, is one that is capable of removing a large majority of toxic nitrogenous wastes produced within a closed system. The biofilter is populated by two distinct groups of nitrifying bacteria: those that oxidize ammonia to nitrite and others that oxidize nitrite to nitrate).

Carbonate alkalinity (KH) - A measure of the carbonate and bicarbonate anions (see ion) in a solution. Carbonate and bicarbonate anions contribute to alkalinity due to their basic nature. This enables them to neutralize acid. KH is usually expressed as parts per million (ppm), milligrams per liter (mg/L) or in degree KH (dKH).

Cellulose - An important structural component of the primary cell wall of green plants and many forms of algae. Some species of bacteria secrete it to form biofilms. Cellulose is the most abundant organic polymer on Earth. The cellulose content of cotton fiber is 90%, that of wood is 40–50%.

Cold-blooded - Designating or pertaining to animals, such as fish and reptiles, whose blood temperature ranges from the freezing point upward, in accordance with the temperature of the surrounding medium.

Daughter cell - Either of the two cells formed when a cell undergoes cell division. Daughter cells are genetically identical to the parent cell because they contain the same number and type of chromosomes.

Decomposition - The process by which organic substances are broken down into a much simpler form of matter. Organisms, such as bacteria) that do this are known as decomposers.

Denitrification - A microbially facilitated process of nitrate reduction performed by a large group of anaerobic bacteria) that may ultimately produce molecular nitrogen (N2) through a series of intermediate gaseous nitrogen oxide products.

DNA – Deoxyribo Nucleic Acid, a self-replicating material which is present in nearly all living organisms as the main constituent of chromosomes. It is the carrier of genetic information.

Inorganic - Not consisting of or deriving from living matter.

Ion - An atom that has either gained or lost electrons and therefore has a charge.

An anion is an ion that has gained an electron from its neutral atomic state and is therefore negatively charged. A cation is an ion that has lost an electron from its neutral atomic state and is therefore positively charged.

Lignin - a complex organic polymer deposited in the cell walls of many plants, making them rigid and woody.

Macroinvertebrate - organisms without backbones, which are visible to the eye without the aid of a microscope. Aquatic macroinvertebrates live on, under, and around rocks and sediment on the bottoms of lakes, rivers, and streams.

Metabolism - The set of life-sustaining chemical reactions in organisms. The three main purposes of metabolism are: the conversion of food to energy to run cellular processes; the conversion of food/fuel to building blocks for proteins, lipids, nucleic acids, and some carbohydrates; and the elimination of nitrogenous wastes. These enzyme-catalyzed reactions allow organisms to grow and reproduce, maintain their structures, and respond to their environments.

Microbial conditioning - A method of multiplying microbial organisms by letting them reproduce in a predetermined culture medium under controlled laboratory conditions

Mother cell - A cell that divides to produce two or more daughter cells. Also called parent cell.

Nitrification - The biological oxidation of ammonia or ammonium to nitrite followed by the oxidation of the nitrite to nitrate. The transformation of ammonia to nitrite is usually the rate limiting step of nitrification. Nitrification is an important step in the nitrogen cycle in soil. Nitrification is an aerobic process performed by small groups of autotrophic bacteria.

Nitrogen Cycle - The biogeochemical cycle by which nitrogen is converted into multiple chemical forms as it circulates among atmosphere, terrestrial, and marine ecosystems. The conversion of nitrogen can be carried out through both biological and physical processes. Important processes in the nitrogen cycle include fixation, ammonification, nitrification,

and denitrification. The majority of Earth's atmosphere (78%) is atmosphere nitrogen, making it the largest source of nitrogen.

Nucleus - A dense organelle (specialized structures within a living cell) present in most cells, typically a single rounded structure bounded by a double membrane, containing the genetic material.

Organic – Relating to or derived from living matter.

pH - A number expressing the acidity or alkalinity of a solution on a logarithmic scale on which 7 is neutral, lower values are more acid and higher values more alkaline.

Sodium bicarbonate - A chemical compound commonly known as baking soda with the formula NaHCO3. It is a salt composed of a sodium cation and a bicarbonate anion.

Waste degraders - An enzyme and bacteria blend that is useful as a biological additive to aquariums. These bacteria feed on fish and food waste and convert them to ammonia.