

# Wastewater World

By N. J. Hayden

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— N.J. Hayden

It's seven a.m. and ten below zero. I pull my red neck warmer over my nose, and look out over the wastewater treatment plant. Everything is covered with glistening snow except the large settling and aeration basins. They're open and emit an ethereal mist that dissipates into the frosty air. There's no odor today. Nothing smells when it's this cold.

To most people the large concrete tanks sunk into the ground with their catwalks, grates and criss-crossing pipe networks look austere and formidable; an industrial badlands devoid of life, but I know better. I know that within the bubbling brown liquid of the gigantic aeration basins beats the heart and soul of the treatment plant. It's not chemicals that are responsible for the purification of the wastewater, but a complex mixture of microorganisms called activated sludge. This thriving ecosystem along with gravity provides most of the treatment for our human sewage.

Jim emerges from the control building. He's bundled up in his winter work clothes with his head down, a plant manager with a mission. I wave but he doesn't see me. Jim doesn't mind if I steal some wastewater, though. His standard reply for the past fifteen years has been "take as much as you like." An old line, but he says it in a way that still makes me smile.

It's time to get to work. I unscrew the sample bottles and put them carefully on the grate. I slip off my gloves, pocket them, and put on the stiff, cold vinyl ones. I need to move quickly now or my hands will freeze before I'm done. I throw the orange plastic bucket with a rope tied onto the handle into the brown frothy liquid. It tips sideways and takes in water. I pull it up, set it on the grate, and pour its contents into the two sample bottles, not caring if most of it spills,

although I'm careful that it doesn't spill on me. The surplus falls through the grate and back into the tank.

I screw on the caps, wipe the bottles with an old rag, and put everything into the empty bucket including the vinyl gloves. I slip my hands back into my pockets, and ball them into fists for a second to warm my fingers before fumbling for my winter gloves. Vinyl doesn't cut it in this kind of weather.

Back in my lab at the University, I set my bottles on the counter. I'm glad no one is around yet; there'll be no interruptions while I take a quick peak at my collection. The brown solids in the bottles quickly settle, and I'm careful not to disturb them while I lower the pipette and draw some particles into it. I place a drop onto the microscope slide, and put on a cover slip.

Even though I've looked at hundreds of these samples over the years, anticipation runs through me for I'm about to enter the magical world of activated sludge treatment. I place the slide on the microscope stage, set the magnification to two hundred times, and adjust the lights. Then I watch the dance unfold.

Immediately, small protozoa twirl and boogie through the microscope field only to quickly move off, then on, and then off stage. These critters belong to the group of organisms called ciliates because of the hair-like cilia they use to move through the water. They are single celled but contain internal organelles that are visible even at this magnification. The organelles look like bits of ice seen through dappled glass. A different ciliate now enters the field, moving slowly, almost crab-like as it gropes and crawls amid the floc. From the side it looks like a miniature combat helmet except that it's translucent and its cilia extend out from underneath 'the helmet.'

I move the stage and find a cluster of stalked ciliates nestled within the brown floc and increase the magnification. These are my favorite wastewater

microorganisms especially the ones that bunch together, sometimes dozens in a clump, like a bouquet of crystal flowers. Other types of stalked ciliates are solitary, lone sentinels on the lookout for a tasty meal. As their name suggests, these ciliates have a long thin stalk on which sits an elongated cell that culminates in tufts or ridges of cilia. The moving cilia create eddies and currents that draw in bacteria and other food toward their mouth opening. All of the ciliates in wastewater treatment are bacterivores — hunters, trappers, and grazers of bacteria. Some of the stalked ciliates even lie in wait with their stalk coiled up, and just at the right moment spring out at the unsuspecting bacteria, gobbling them whole. How they know their prey is there, is still a mystery.

The bacteria, also single celled, are generally much smaller than the ciliates, but their numbers are much greater. At four hundred times magnification, the bacteria look like little pinpricks of oscillating light. Some are rod shaped, some are spherical and some are spirals. The rods and spheres generally have flagella on one end of the cell that allows them to move through the water. Their flagella are long whip like structures too thin to be seen with my microscope, but their effects are witnessed in the constant twirling motion of the cells. This motility helps bacteria move away from toxins and toward optimal environmental conditions.

Because they are moving it's pretty hard to tell what's what or who's who except for the spiral bacteria. They are the largest, as long as some of the ciliates, but much thinner, like undulating worms. They are visible even at this magnification, but are not too prevalent in a fresh sludge sample. They'll be noticeable in higher numbers after I leave the bottle sitting on the lab counter all day. I'll still have to point them out to the students though; they're hard to notice at first because they look like tricks of the light or what my optometrist calls floaters.

I take a break and rub my eyes. It's tiring, but it's hard not to keep looking, too. I switch to a dark field. Dozens of rod shaped bacteria jiggle on stage, although they're not much to look at. That's because bacteria belong to the group of organisms without a nucleus or internal organelles. All of the specific cell activities, metabolism, energy storage, energy production, and reproduction take place within the soupy mixed up interior of the cell called the cytoplasm. They have a cell membrane that envelops their cytoplasm and separates them from the rest of the world, and most have a cell wall as well, a tough exterior coat that protects them from getting dried out and from toxins in their environment. At this magnification, however, they're just gray rods jostling for position.

The major challenge for bacteria, like most critters, is acquiring food. Just because their food is all around them dissolved in the wastewater, doesn't mean it's easy to get. They have no mouth opening to engulf their next meal. All of their food has to diffuse through the cell wall and cell membrane. Even though bacteria have developed mechanisms to speed up this type of transport, it's still tough. And it's not only food that has to diffuse through the cell wall and membrane, oxygen and other nutrients do, too. Likewise cellular waste products have to escape through that same membrane and wall. The movement of molecules through their cell membrane and wall means that very large molecules like some of the chemicals that humans have put down the drain including pharmaceuticals, anti-microbial soap ingredients and other personal care products can't get into the cell. And if they can't get in, bacteria can't break them down. That means some of these chemicals are getting into our lakes and streams still intact, and we are only beginning to recognize their impact on human health and the environment.

But don't blame the bacteria; they are the heroes in the wastewater treatment story. Bacteria are at the base of the food chain, eating the soluble carbon material in the wastewater. And contrary to popular belief, these bacteria

are not genetically-modified mutants, nor are they disease-causing organisms (pathogens). The wastewater bacteria are the creatures we find naturally in streams, sediments and soils recycling the world's organic matter. For domestic wastewater treatment, we select and concentrate these bacteria giving them plenty of food and oxygen so they can multiply and grow.

What about the disease-causing microorganisms and coliform bacteria from human intestines that get flushed down the toilet everyday? What happens to them? Many of them die a natural death because they can't live very long outside their human host. Others become a tasty meal for the various ciliates and other wastewater predators. Still others get trapped within the floc. The activated sludge, also called biological floc, contains bacteria (both good and bad), ciliates, small suspended solids such as toilet paper, feces and food debris, and anything else that congregates there. Floc is short for flocculated which means to come together and stick, thereby making bigger and heavier particles. Large floc settle quickly within the settling tank — the next tank after the biological activated sludge tank. The settling tank is also called a clarifier or sedimentation tank. After settling, the wastewater can undergo filtration or further treatment, or it might be clean enough for final disinfection and discharge. It all depends on the treatment plant.

The first steps for treating domestic wastewater begin with screening and settling the large solids — gross gross solids I call them. Toilet paper, condoms, tampons and lots of undistinguishable stuff gets removed. The screens look like large grates so the water can go through, but they catch the big stuff. They are either hand raked or cleaned mechanically. On plant tours, my students want to know about the strangest thing Jim's found on the screens. Any alligators? Not even a goldfish, or one he could identify anyway. The most interesting thing turned out to be a fifty-dollar bill that was mashed in with all the muck on the

screens. Did you keep it? They asked. Damn straight, although he washed and dried it first.

The screening and settling processes remove a large percentage of the grossest materials, but these aren't sufficient to keep our lakes and rivers clean. Before the 1970s, this was typically the entire wastewater treatment process; screening and settling solids with a shot of disinfectant to kill pathogens at the end. Some towns didn't even have that much. The wastewater pollutants left in the wastewater after this initial settling such as soluble organic matter, nitrogen and phosphorus, not to mention untreated chemical wastes, were being discharged into rivers, lakes and bays. The fouling of our waterways from this pollution was the main impetus for passing the Clean Water Act in the early 1970s. This federal Act required discharge permits for all public and private wastewater facilities and limits were established on how much could be discharged. The Act resulted in targeting priority pollutants for removal as well as requiring best available treatment technologies for treating wastewater. As a result of the Clean Water Act, wastewater treatment plants now need to remove most of the soluble pollutants remaining in the wastewater after initial settling, such as the urine, dissolved fecal matter, and a whole bunch of nastiness we flush down the toilet or pour down the drain everyday. Biological treatment processes are now installed in all domestic wastewater treatment plants in the U.S. to treat these dissolved components.

I change the magnification back to two hundred times and take another scan around the slide. I find a rotifer, one of the few multi-cellular organisms of the sludge, munching on the floc. These floc eaters look like elongated electric shavers, with rotating bristly heads that encircle and draw food into their "mouth." The mouth connects to a rudimentary beginning of a gut that I see as a dark central core of the rotifer. Near the critter's anterior end is little ballerina type "feet" that wriggle and cause the rotifers to pirouette and twirl as the mood

strikes them. The rotifers are higher up the food chain than bacteria and ciliates, and are generally evident, although not overly abundant, in healthy sludge. Healthy sludge contains lots of free-swimming ciliates, some stalked ciliates, and large numbers of rod and spherical shaped bacteria. These creatures are a good indicator of how well the treatment process is working.

One of the bottles goes into the refrigerator to preserve the healthy sludge for when the students come in for the afternoon lab session. Keeping the sludge cool slows down the respiration of the microorganisms, so they won't use up all the oxygen in the bottle. Most of the activated sludge organisms are aerobic. Like us they need oxygen to live.

The other bottle I leave on the counter, top off. In the warmth of the lab, the microorganisms in this bottle will quickly deplete the oxygen. This dramatic change in the environment will result in a whole new ecosystem developing. A small amount of oxygen will be available as it diffuses in from the surrounding air, but not enough to keep all the critters alive. I've done this before and been amazed at the changes that occur within the bottle. Different types of organisms, most that I didn't even see in the "fresh" sample will dominate. Spiral bacterial, different types of ciliates, amoebae, and even fungi with their long branched hyphae will show up. It's a great "compare and contrast" exercise for the students and it shows them how changes in treatment operating parameters can result in big changes in the microbial community, which in turn has dramatic effects on treatment efficiency.

Over the years, I've noticed that wastewater samples from different treatment plants often show differences in ecological makeup. I rarely see amoebae in Jim's activated sludge, but in activated sludge from another local plant they are common. Amoebae are single-celled critters that can move by reshaping themselves, slowly stretching themselves in a new direction. It's called cytoplasmic streaming. For us, it would be like if we were lying on the

floor and our arms and head grew at the expense of shrinking legs. Eventually we would make it across the floor, but we wouldn't look anything like the way we started.

The relationship between ecosystem makeup and the way the treatment plant operates isn't well known. There are too many aspects of the operation that affect the ecology of the activated sludge. But we do know certain things that cause upsets. Once when I looked at the floc from an ice cream wastewater treatment plant I was amazed to see thousands of long skinny strands. The floc looked like a tangled mass of hair, but was really a tangled mass of filamentous bacteria — the worst case I'd ever seen, including pictures from textbooks. The wastewater from this ice cream manufacturing facility had high concentrations of organic matter and soluble fats. The wastewater treatment plant had a difficult time supplying enough oxygen to the bacteria to keep the system aerobic and they frequently had really low oxygen levels in the wastewater. Certain types of filamentous bacteria do well in those kinds of conditions and out-compete other organisms that need higher oxygen levels. The problem with dense populations of filamentous bacteria is they don't flocculate (clump) properly. Then they won't settle. This is called bulking sludge and leads to a higher solids and higher organic matter content in the discharge. That's not good for the environment, nor is it good for the treatment plant.

Most of the time the floc does settle quickly within the sedimentation basin. Some of the brown sludge at the bottom of this tank is then recycled back into the activated sludge tank to keep a high population of microorganisms there. That's where the term "activated sludge" comes in; this recycling keeps the biological system active. But not all the sludge can be recycled; some of it needs to be separated, treated and then properly disposed. At Jim's plant where I typically get my samples, they pump the sludge to an anaerobic (meaning without oxygen) digester. All the solids settled in the beginning of the treatment

plant get pumped there, too. In the digester, very different consortia of bacteria convert the sludge into carbon dioxide and methane. Jim's treatment plant uses some of the methane as a fuel source to heat the digester, as well as to fuel microturbines that generate electricity for the plant. Even after the digestion, there is still some sludge that needs to be disposed of. Some plants send it to a landfill, while others make a soil amendment from it that can be sold. Digested sludge is great for soil as long as it doesn't have toxic metals or toxic chemicals in it. If we could only get people not to dump household and industrial wastes down sinks and toilets!

There are dozens of different types of biological wastewater treatment systems being used today. Some people have their own right in their backyard and don't even realize it. Septic tanks and leach fields function in the same way as a municipal wastewater treatment plant. The septic tank acts as the initial settling basin as well as an anaerobic digester, albeit not a very efficient one since there's no mixing and no heat. From the septic tank the wastewater flows through trenches and infiltrates into the soil. What most people don't realize is there are all sorts of bacteria, ciliates, amoebae and other microorganisms happily living within the soil and trench, similar to those critters mentioned earlier except these organisms attach themselves to the soil particles and rock surfaces. We call this a biofilm.

Biofilms exist throughout nature. That film on your teeth in the morning when you forget to brush is a biofilm. Those rocks in a clear mountain stream have biofilms on them, and that's what makes them so slippery. The bacteria in the biofilms are eating any soluble organic material that come their way, and in turn are being eaten by bacterivores.

Constructed wetlands are another example of biological wastewater treatment processes that add additional ecological complexity (green plants) to the system. As is typical, initial settling occurs prior to entering the constructed

wetland. Once in the wetland bacteria, ciliates and other microorganisms consume the organic matter just as they do in other treatment systems. But the addition of the plants and the additional habitat they create through their roots and upper vegetation provides greater biodiversity within the system. Wetland plants also shunt oxygen to their roots as a mechanism to keep the root zone aerobic. Bacteria and other microorganisms use the excess oxygen supplied by the plants to consume some of the organic matter in the wastewater. Unfortunately, it's difficult to supply enough oxygen to the microorganisms if the microorganisms are too concentrated, so constructed wetlands tend to take up more aerial space than other types of wastewater treatment plants. They can be less expensive to operate though since oxygen (usually as compressed air) might not be needed. Aeration and sludge treatment and handling are two big operating expenses at wastewater treatment plants that wetlands can avoid.

My graduate student comes in and asks me what I'm doing. I show her the rotifer and some of the other cool critters on the slide. Like me, this microscopic world mesmerizes her. She's also interested in wastewater treatment because we're trying to understand what happens to a compound called nonylphenol within the treatment process.

Nonylphenol is an anaerobic breakdown product of a commonly used chemical (nonylphenolethoxylate) found in detergents and cleaners – commonly used, but not easily said. Nonylphenol is currently of concern because of its toxicity, estrogenic properties (hormone mimicking) and its widespread contamination. As an estrogen mimic, it falls into the category of emerging environmental contaminants called endocrine disrupting compounds. These endocrine-disrupting chemicals can mess up the hormonal system in humans and other animals and have shown some alarming effects especially in aquatic animals. Alligators and fish have been found with characteristics of both sexes, resulting in sterility.

Although nonylphenol is typically found in low concentrations in water and wastewater, low concentrations of hormones can still have an effect. Nonylphenol is also difficult to degrade by the microorganisms in the wastewater treatment plant, although it is attracted to the solids. One of the ways it gets removed from wastewater is through the removal of the sludge. What happens to it in the sludge is less clear, but some evidence suggests that it is not degraded there either. Some of the nonylphenol entering the plant can also volatilize into the atmosphere during treatment, which is not necessarily a good thing, but it keeps it out of the wastewater discharge.

Our results suggest that nonylphenol is present in the wastewater coming into Jim's plant and that it is being reduced during treatment, but it was still measured in the discharge. This isn't uncommon, nor is it uncommon to find a whole bunch of other chemicals such as active ingredients of antimicrobial soaps, perfumes and additives in shampoos and soaps, and even caffeine. In fact, most of the medicines from our medicine cabinet show up in wastewater discharges. Some studies have shown that some of these are treated, but most aren't. The wastewater organisms can't take up these strange chemicals and eat them, so many just keep going right through the wastewater plant and into our lakes and rivers.

I turn off the microscope and wash up, wondering what substances in this soap will and won't be treated when this water finally makes its way to the treatment plant. I don't buy antimicrobial soaps and other products that have some of the chemicals I know aren't good for the environment, but what about all the unknowns? Maybe our wastewater heroes, the bacteria, will come to our rescue, but maybe we can't count on them. They weren't much help with DDT and some of our other diabolical creations. I believe we need to practice more precaution in the types and numbers of new chemicals we introduce into the

market every year. We also need to remember the lessons from our past, the biggest being to stop spewing synthetic chemicals into the environment when we don't know the long-term effects. We're wrong if we think the biological floc can handle them. Those creatures evolved to eat and recycle natural carbon substances, not synthetic fragrances, painkillers and chemotherapy drugs.