Biological Denitrification Evaluation for Pirana at Massachusetts Alternative Septic Systems Test Center

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Removal of nitrogen from wastewater is a national concern. Because of its impact on the environment, the USEPA has established limits for safe drinking water of 10 mg/l of water. Salt-water estuaries are also threatened by this productivity-limiting nutrient. Given an inventory in the United States of over 26 million septic systems, that discharge of nitrogen biological denitrification is considered essential to the lifecycle management of wastewater treatment systems. Septic systems inject an estimated six billion gallons of treated effluent a day into the groundwater and, assuming no losses, likely contribute1.76 million pounds of nitrogen to the environment. States along the eastern seaboard have for the most part ignored onsite treatment as a regional option for nitrogen removal and have instead focused on extending sewers and adding nitrogen removal systems to municipal treatment plants.

The major barrier faced by state and municipal authorities seeking to extend sewers and upgrade municipal treatment as the solution to meeting denitrification requirements is cost. Attempts to use more advanced onsite systems have similar cost barriers, along with the new requirement for managing these far more complex onsite systems. The advent of a simple pretreatment septic tank insert that maintains a custom blend of nitrogen consuming bacteria, and easily upgrades existing septic tanks into biological denitrification systems, dramatically reduces these barriers. The validation of this promising new technology by Massachusetts's Alternative Septic System Test Center (MASSTC), documents the potential of this technology to reduce nitrogen in residential systems. Areas needing this advancement, given the very high cost of proposed sewers and advanced treatment systems, include communities adjacent to Long Island Sound, Cape Cod, and the Chesapeake. Towns struggling to reduce nitrogen to protect drinking water supplies or developers seeking more efficient land use will also find this technology very useful.

This paper reviews the fundamental chemistry and processes of biological denitrification relative to the conventional onsite technologies approved in Massachusetts. It presents and reviews pilot scale testing of the Pirana system and the theory behind this new biological process. Through this exercise, we can evaluate the validity of this process and thereby accelerate its regulatory review and approval. This is essential to bring it

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into the market place and to make this valuable wastewater treatment option readily available to small communities and citizens across the country.

What is a Pirana System?

The Pirana technology consists of two Patent Pending components: an aerobic reactor that is simply inserted into an existing or new septic tank and a microbial generation process that continuously inoculates the wastewater with Pirana Blend bacteria, see Figure 1.. A fine bubble diffuser located at the base of the reactor circulates aerated effluent throughout the tank. A 40-watt linear pump generates the air required to drive the treatment process. Existing septic tanks are easily upgraded by inserting the reactor. Optimum nitrogen removal is accelerated by removing the sludge, which contains vast quantities of organic nitrogen, prior to startup.

Biological denitrification – a fundamental process for returning nitrogen to the atmosphere

Nitrification

Urea is a fundamental waste product from man. This compound, when discharged into a septic system, is classified as ammonia. Oxidation of ammonia into nitrate occurs in the soil under aerobic conditions. The process normally depends on two types of bacteria, Nitrosomonas that convert ammonia to nitrite and Nitrobacter that covert nitrite to nitrate. These organisms derive energy and food for cell synthesis from this nitrogen compound and produce a very valuable and essential nutrient that supports photosynthetic plant productivity. This oxidation process depends on a number of variables including oxygen and alkalinity. Appropriate levels of carbon, temperature, and pH also affect the efficiency of these bacteria. Given competition from other carbon loving bacteria, BOD is reduced prior to nitrification beginning. Because the nitrification process demands about 4 mg of oxygen and 7 mg of alkalinity (expressed as carbonate) for every milligram of ammonia processed, we normally observe a drop in pH as the carbonate is utilized to build cell mass. Equations 1 and 2 define the stoichiometry of biological nitrification with equation 3 describing the assimilation of the ammonium ion into cell tissue.

$NH_4 + 1.5\ O_2 \rightarrow NO_2 + 2H + H_2O$ (typically by Nitrosomonas) (1)

$NO_2 + 0.5\ O_2 \rightarrow NO_3$ (typically by Nitrobacter) (2)

$NH_4 + 1.731 O_2 + 1.96 HCO_3 \rightarrow 0.038 C_5H_7H_2O + .962\ NO_3 + 1.07 H_2O + 1.769 H_2C0_3$

Denitrification

Denitrification converts nitrate to nitrogen gas through a series of reactions that utilize the oxygen molecules as electron acceptors. Denitrifying bacteria derives energy for cell synthesis from the conversion of nitrate to nitrogen gas. This process is driven by synthesis of carbon and the resulting need to oxidize this food. The denitrification process normally occurs under anoxic conditions, i.e. an environment rich in both nitrate

and soluble carbon but void of oxygen. In most wastewater treatment processes such as septic systems, the carbon is too low in the soil and the oxygen levels are too high, therefore nitrate moves into the groundwater. Given a choice, facultative bacteria will use oxygen first, then nitrate, and finally sulfate as an electron acceptor. Because wastewater treatment systems remove carbon prior to nitrification, soluble carbon must be added to the wastewater. Another option includes nitrification followed by recycling of the nitrate rich effluent into a carbon rich low-oxygen septic tank. The later approach results in a mixing of fresh and processed effluent and frequently leads to a blending of effluent, with both ammonia and nitrate present in the end product. Given this blending and short-circuiting of ammonia, total nitrogen below 10 mg/l is difficult to achieve.

The denitrification process utilizes the oxygen in the nitrate molecule as an electron acceptor. Enzymes within individual cells or between cells strip the oxygen, one molecule at a time. Therefore, nitrite is formed as part of both nitrification and denitrification. Microbiologists who have worked with forest ecosystems recognize that a number of facultative bacteria can participate in the nitrification/denitrification process. Dr. Dan Wickham has run experiments in forest ecosystems and found little nitrate in waters following treatment using an enhanced bacillus culture. Experts in the field of microbiology recognize the fact that these soil-based bacteria can compete for the nitrite and therefore participate in the denitrification process. The research and performance data suggest that the Pirana system can create a biological community where this competition inhibits Nitrobacter and thereby blocks the conversion of nitrite into nitrate.

As a facultative organism, bacillus will process carbon, with or without oxygen. They evolved this ability, given the need to survive and process leaf litter and because this environment can be either aerobic or anaerobic depending on soil moisture. What is very significant is the ability of these bacteria to utilize nitrite. Even in an aerobic environment, they will select nitrite over nitrate and potentially compete with Nitrobacter for this nutrient. Under normal wastewater processes Nitrobacter wins this contest, however in the soil with an exceptionally high numbers of bacillus little nitrate is formed.

The ability of bacillus to utilize nitrite requires manipulation of the wastewater process. While much simpler than an intermittent aerobic process or a conventional recirculating sand filter (RSF), the patent pending process reactor is needed to mix and aerate the effluent. This devise provides continuous inoculation by blending aerated effluent rich in carbon with the bacillus culture, and thereby generates billions of these organisms. Because these single cell organisms can double in as little as 30 minutes, they can very rapidly dominant the treatment process and exclude other organisms that frequently invade ATU's. This continuous inoculation process thereby reduces the diversity and development of higher order aerobic organisms such as protozoa that commonly invade ATU's and municipal wastewater treatment systems.

Because the nitrification/denitrification reaction occurs in the soil, effluent collected in a lysimeter after traveling through the soil is essential for documenting the effectiveness of the Pirana treatment process. Based on an understanding of the stoichiometry of the nitrification and denitrification, we would therefore expect the Pirana to condition the

wastewater for treatment in the soil. Because of the extremely high number of bacillus bacteria, we also suspect competition with Nitrosomonas in the pretreatment resulting in the need for less dissolved oxygen and less alkalinity. The fact that the bacteria are facultative also increases their efficiency in the tank demand increased energy for complete mixing.

Test Method and Procedures

MASSTC Laboratory Method

Figure 2 (MASSTC Lab Report) shows actual lysimeter sample test result from a lysimeter receiving Pirana effluent and lists EPA's approved laboratory method used by the MASSTC's laboratory. In addition to these laboratory procedures, note that the sample was collected by G. Heufelder, Director for the Center, who immediately transported them to the lab. Additional routine calibration of all pumps that feed the tanks is conducted by full time field technicians.

To insure an unbiased side-by-side comparison, the same ETV/EPA loading strategy used for Title 5 septic tanks was adopted for the septic tanks with the Pirana insert. Figure 3 below shows the percent of daily flow throughout the day for all test systems. The computer time control system applied a similar dosing strategy with pump run times adjusted (based on weekly pump calibration) to insure equivalent flows into all tanks.

Three replicated Title 5 test systems used individual 1,500-gallon septic tanks. Each tank discharged into a separate trench that was 25 percent of a residential system. Proportional loading was accomplished by a d-box fitted with a Dipper that dumps into four 4-inch pipes, one going to the 14-foot by 3-foot gravel trench and the other three to a bypass sump. This design preserved proportional loading for this pilot scale side-by-side test facility.

In June 02, one of the three Title 5 tanks was converted to the Pirana process. As this treatment system relies on the soil for denitrification, the stress tests called for increasing the loading rate to the soil while maintaining the 330-gpd loading of the tank. In order to increase hydraulic loading, the bypass outlets in the d-box were successively closed off. A second Pirana reactor was added to another septic tank that drains into three replicate tire chip aggregate test trenches. These trenches were 2 feet wide and 11 feet long with only 12 inches of sand above a lysimeter liner. Flow from this tank was distributed via a d-box fitted with six Equalizer outlets, three discharging to three replicate trenches and three to a by-pass return sump.

Table 1 shows a successive increase in wastewater applied to the Title 5 trenches (F1&F3), Pirana trench F2, and three replicate tire chip (TC) trenches. In the later case, all three bypass outlets were closed in one step, thus doubling the flow into these test trenches after only two weeks of operation. With three replicate trenches receiving

effluent from one tank, only 110 gallons per day could be applied to each TC trench. The F2 trench was successively increased from normal Title 5 loading to four times normal.

System Operation and Sampling Frequency

System startup consisted of simply adding the Pirana to the existing tanks and turning on the aeration. Solids monitoring data provided by the Worldstone SepticWatch on the F3 control tank indicated 25 percent solids with over 11 inches of sludge in the tank. The initial samples thus reflect processing of these solids.

Daily readings of DO and pH provide an indication of the state of the system, while weekly samples defined raw, d-box, and pan lysimeter effluent quality. On August 7, this phase of testing ended when the test tanks were pumped and refilled with fresh water. After three weeks of feeding 330 gpd into these tanks, the sampling resumed on a biweekly schedule. Sampling continued into the winter, until early February when the feed lines to the TC tank froze.

Results and Analysis

A parallel operation of control tanks supports a side-by-side comparison of conventional septic system denitrification with a recirculating sand filter and the Pirana system. As the sand soil media plays an integral part in each system, we will compare effluent quality following treatment in the soil. We will begin by reviewing the transformation of nitrogen in the standard septic system control and RSF control system, as these two conventional systems represent the standard treatment processes approved in Massachusetts.

Septic Control

Figure 4a, 4b, and 4c shows the total nitrogen in the raw wastewater feed to all system, total nitrogen leaving the septic tank, and finally pan lysimeter concentrations after 24 inches of sand. Total nitrogen is defined as the sum of Total Kjeldahl Nitrogen (TKN), nitrite, and nitrate. The variability in the effluent quality and time lag for samples collected on a given day at each location requires a comparison of average concentrations over a longer time frame. The relationship or scatter of these data over time helps us identify possible trends in these data. Not surprisingly, the data were evenly distributed about the mean in the raw influent and pan lysimeter effluent. What may be surprising to many is that these data showed that nitrogen was conserved. While the septic tank generates more nitrogen than it receives in the summer, the total nitrogen leaving the soil is about the same concentration as the raw wastewater. The soil appears to be a great equalizer, with average release of nitrogen of 39 mg/l equivalent to raw wastewater entering the test facility. As nitrogen is not generated in a septic tank, we can surmise that winter operation data would show a slight reduction in total nitrogen discharged from the tank due to reduced biological digestion, better settling, and solids storage.

Recirculating Sand Filter Control - TN Performance

A summary of the monthly performance for a recirculating sand filter at the MASSTC is presented in Figure 5. Note that the monthly average total nitrogen shows that this

technology is sensitive to temperature. A second performance related issue concerns the possible influence of stored carbon in this mature filter. A decline in late summer performance is likely due to reduced concentrations of organic solids in the filter. Once this carbon reserve was exhausted, the systems nitrogen removal performance declined. With an average annual TN of 22.5 mg/l, the technology reduces nitrogen by about 29 percent.

Pirana TN Removal Performance

Both Pirana units started operation by processing the existing residuals anaerobic sludge in the septic tank, while also receiving 330 gallons per day. The process started up very quickly with a pH in the range of 7.5. Dissolved oxygen varied and was lower as the tank temperature rose. After monitoring the systems for about two months, both tanks were pumped and all residual solids were removed in early August. While total nitrogen was dropping after 60 days of operation, the removal of the residual sludge accelerated the denitrification process in the soil. One indication of the change concerns nitrite levels, which are normally in the range of 0.025 mg/l. Figure 6 shows the relative increase in nitrite levels, which approach 10 to 80 times the concentration normally found in wastewater treatment systems. This indicated significant inhibition of nitrification, though some nitrate was still present in the effluent in the TC tank. TN leaving the TC tank was about 1/3 less.

Once the effluent moved into the soil, we begin to see the effects of elevated concentrations of soil-based bacteria. Figure 7 shows the historical total nitrogen in the sump draining the TC trenches. This reflects a fairly steady reduction in total nitrogen with levels below drinking water standards by January 03.

Comparing this performance with that of the RSF, we see that the process improved over time and was not retarded by lower temperatures. The increased solubility of oxygen in water may have supported more favorable conditions for bacterial generation. In this case, dissolved oxygen rose to between 5 and 8 mg/l in December and January, up from 1 to 3 mg/l, with many readings less than 1 mg/l during the summer. The TC tank also showed a lower pH, still above 7, indicating that nitrification and denitrification was occurring in this tank. In general, a mix of nitrate and ammonia was discharged into the TC sand trenches with a concentration of TN in the mid 20s.

Complete nitrification was occurring in the soil until January, when 3 mg/l of TKN remained in the TC sump effluent. However, a dramatic shift occurred in December with nitrate dropping dramatically from levels in the mid teens to less than 5.2 mg/l. The sensitivity of Nitrobacter to colder temperatures in the TC tank may have contributed to the change, which was observed in December. The F2 Pirana system was less consistent in its transformation of the soil microbiology. Figure 8 shows it's performance history after two feet of soil, with effluent captured in a pan lysimeter.

Please keep in mind that the F2 trench was loaded at three times the TC trench rate with 330 gallons per day and a flux rate (Q/wetted surface) of almost eight gallons per day. In the winter, when temperatures dropped, some ponding appeared in all trenches and flux

rates dropped as shown in Figure 9. Prior to December, no ponding was observed in F2 and the uneven gravity distribution and resulting microbiology was certainly more dynamic. It's very likely that a pocket of Nitrobacter could continue to thrive in soil pores in this trench and generate nitrate, thereby the pan samples may only provide part of the performance picture.

Both the TC and F2 Pirana tanks showed initial TN concentrations similar to that for the standard septic system. Certainly residual organic material was also present in these sludge laden septic trenches. The 2-foot pan lysimeters for the F2 trench captured effluent from the fringe of the trench. This effluent quality likely reflected the transformation of nitrogen in the zone adjacent to the trench sidewall, and may be quite different from a composite sample collected from three replicated TC sumps. To better understand the average total nitrogen, samples from the F sump that drained all three F trenches were collected (Table 2) and analyzed. These samples reflected a mix of effluent from the two septic trenches generating 33 percent of the flow and from the highly loaded Pirana trench representing 67 percent of the flow. They also reflect greater travel through the unsaturated media and much longer detention times as effluent moved through five feet of sand.

Only by performing a mass balance on the F trenches can we estimate the average total nitrogen draining from the soil below the Pirana trench. This exercise also requires that we adjust for possible dilution due to rainfall. Average values for nitrogen in the F3 Title 5 lysimeter pan at two feet (Figure 4c) were used to define nitrogen contributed by the conventional system. The end result of this calculation is an estimated TN of 5 mg/l or less coming from the Pirana. CODs of less than 2 mg/l in the two-foot effluent pan indicated that this process is likely working with extremely little carbon. Extremely low fecal Coliform of less than 10 cfu/100 ml (Table 2) also indicates that these pathogens are likely consumed in this soil-based process.

Application of this research

The implications of this new treatment process vary with the interests of each party. Given that we are increasing the hydraulic load by 400 percent for the Pirana fed trenches, we are collecting samples that represent an equivalent of four homes on Pirana and two homes on conventional septic. The end result is 15 to 20 mg/l of Nitrogen, slightly less than the effluent concentration documented for a Massachusetts's Department of Environmental Protection (DEP) approved RSF. What's very interesting is that in order to achieve these levels, the Pirana must reduce the TN to 1 to 5 mg/l. To community officials seeking to reduce nitrogen while keeping onsite treatment, this means that 1/3 of the homes may not need to upgrade to an RSF or other DEP approved denitrification system to meet MA performance expectations. For more environmentally sensitive areas, when applied to all homes the technology can meet drinking water standards or significantly reduce the nitrogen draining into sensitive estuaries. For developers in central Cape Cod, these performance levels will eventually mean more flexible development at higher densities.

Conclusion

The MASSTC proved invaluable with existing pilot scale septic systems easily converted to the Pirana process and with comprehensive evaluation by a professional staff and lab. The existing protocol, reviewed and approved by EPA, includes a full-scale lysimeter. This provided a challenging evaluation and generated essential information for regulatory officials seeking to define the potential of this process. Given the challenge of comparing soil treatment, MASSTC is likely the only facility in the country that could evaluate this soil-based process.

Biological denitrification with continuous inoculation of "designer" bugs looks promising given its simplicity, rapid startup, and overall superior performance.

While the results are very impressive, more research is needed by microbiologists and environmental engineers to understand the chemistry and engineering science required to establish the wide scale design of this unit process. When this is done, we should expect the application of the small flow technology on a much larger scale, meeting the needs of commercial and small communities with centralized treatment. Given this advancement in wastewater treatment, the need for sewers to control the release of nitrogen is greatly diminished making onsite technology the superior life cycle solution.

References

Crites, Ron and George Tchobanoglous 1998, Small and Decentralized Wastewater Management Systems, WCB/McGraw-Hill

Wickham, Daniel 2001, Biological Innovation in Wastewater Nutrient Elimination, Present at California Water Environment Association, Redding California by Piranico, Occidental, CA

Woods, Craig, H Bouwer, R. Svetick, S. Smith, and R. Pettyman, 2002 Study Finds Biological Nitrogen Removal in Soil Aquifer Treatment System Offer Substantial Advantages, in Small Flows Quarterly, Vol 3, Num 3 pp14-21.

Figure 1 – Pirana Component Cross Section

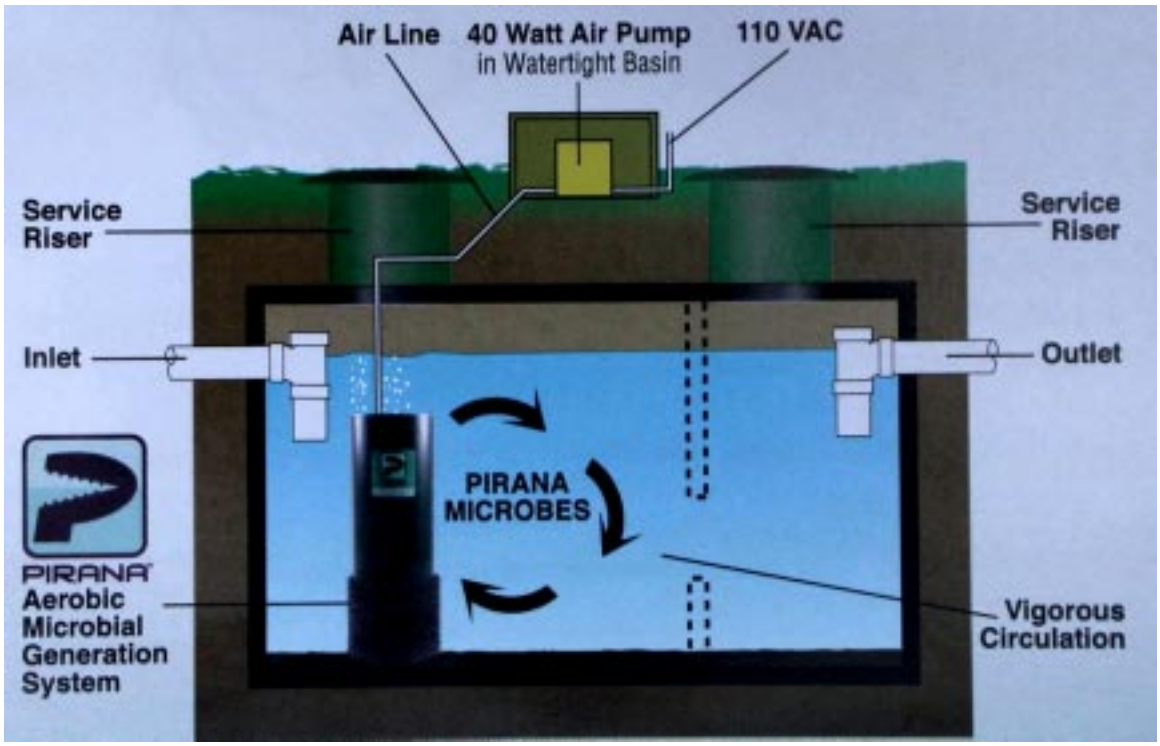
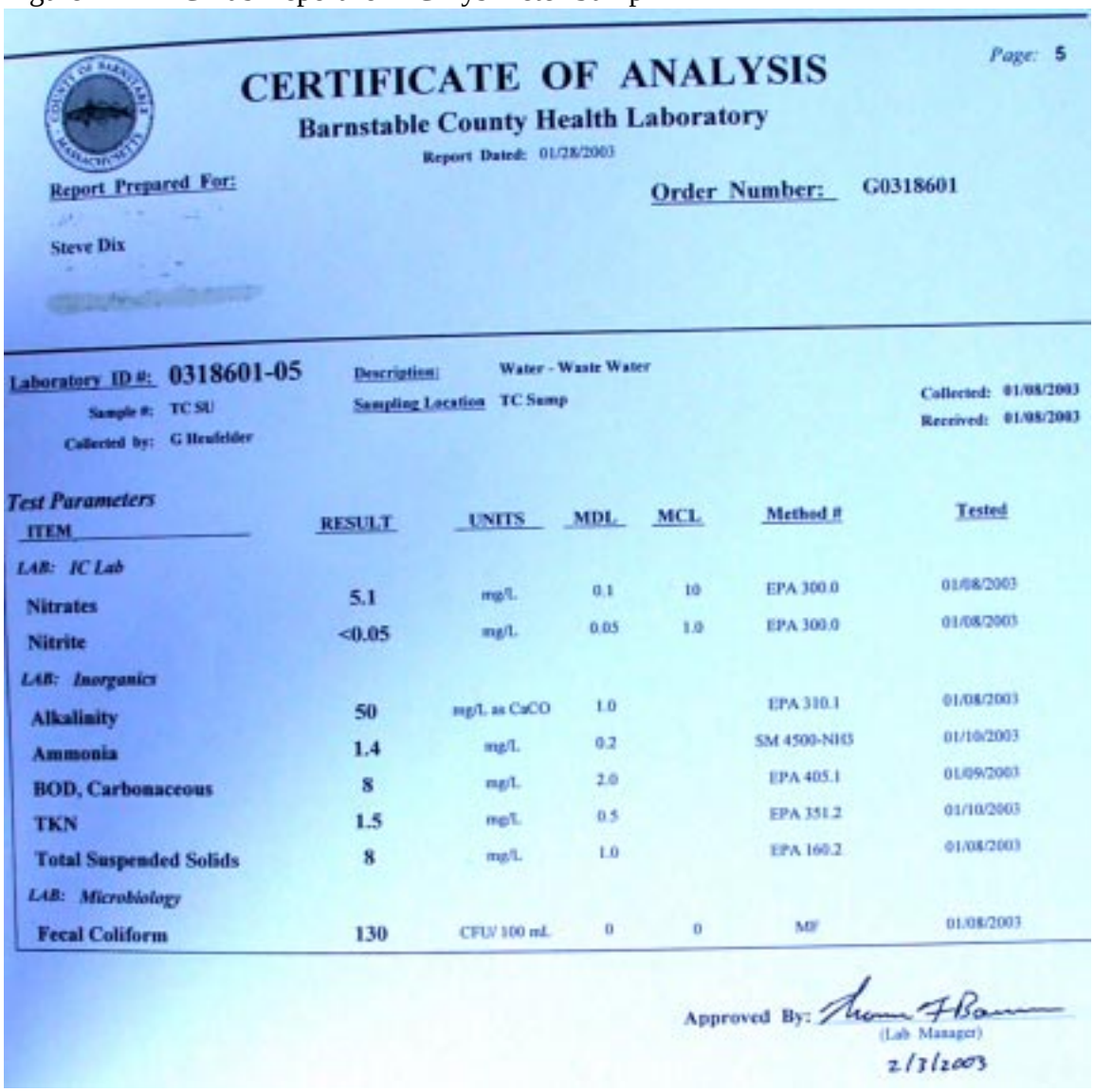


Figure 2 MATC Lab Report for TC Lysimeter Sump



CERTIFICATE OF ANALYSIS

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Barnstable County Health Laboratory

Report Dated: 01/28/2003

Report Prepared For:

Steve Dix

Order Number: G0318601

Laboratory ID #: 0318601-05
Description: Water - Waste Water
Sample #: TC SU
Sampling Location: TC Sump
Collected by: G Heufelder
Collected: 01/08/2003
Received: 01/08/2003

Test Parameters

ITEM	RESULT	UNITS	MDL	MCL	Method #	Tested
LAB: IC Lab						
Nitrates	5.1	mg/L	0.1	10	EPA 300.0	01/08/2003
Nitrite	<0.05	mg/L	0.05	1.0	EPA 300.0	01/08/2003
LAB: Inorganics						
Alkalinity	50	mg/L as CaCO	1.0		EPA 310.1	01/08/2003
Ammonia	1.4	mg/L	0.2		SM 4500-NH3	01/10/2003
BOD, Carbonaceous	8	mg/L	2.0		EPA 405.1	01/09/2003
TKN	1.5	mg/L	0.5		EPA 351.2	01/10/2003
Total Suspended Solids	8	mg/L	1.0		EPA 160.2	01/08/2003
LAB: Microbiology						
Fecal Coliform	130	CFU/100 mL	0	0	MF	01/08/2003

Approved By: Thomas F Bamm
(Lab Manager)
2/3/2003

Figure 3 Percent Loading of Pretreatment Tanks at MASSTC

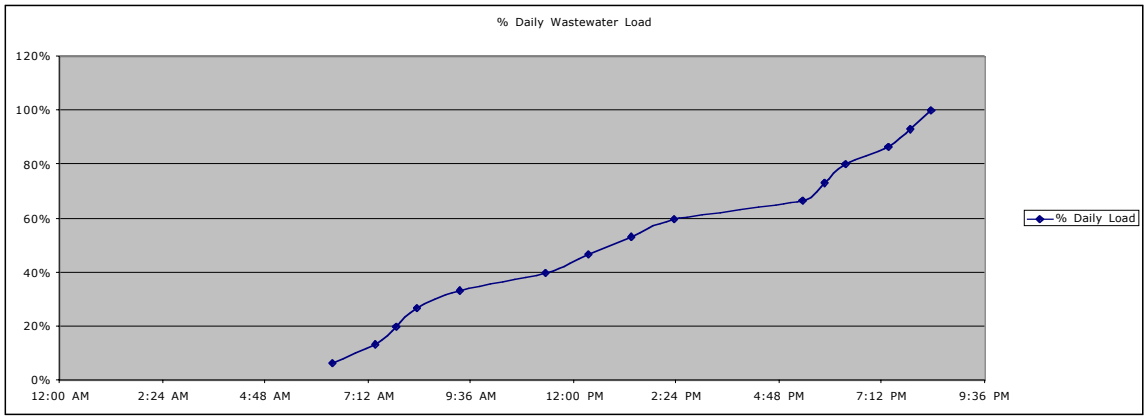

Table 1 Trench Loading Rates in 2002

Date	Loading Rates (gpd)		
	F1&F3	F2	TC
7/2	82.5	82.5	55
7/3	82.5	82.5	55
7/12	82.5	110	55
7/17	82.5	110	110
7/24	82.5	110	110
7/31	82.5	110	110
8/2	82.5	110	110
8/7	82.5	110	110
8/14	82.5	110	110
8/21	82.5	165	110
8/28	82.5	165	110
9/4	82.5	165	110
9/11	82.5	165	110
9/18	82.5	330	110

Figure 4a Total Nitrogen in the influent wastewater (R=Correction Coefficient)

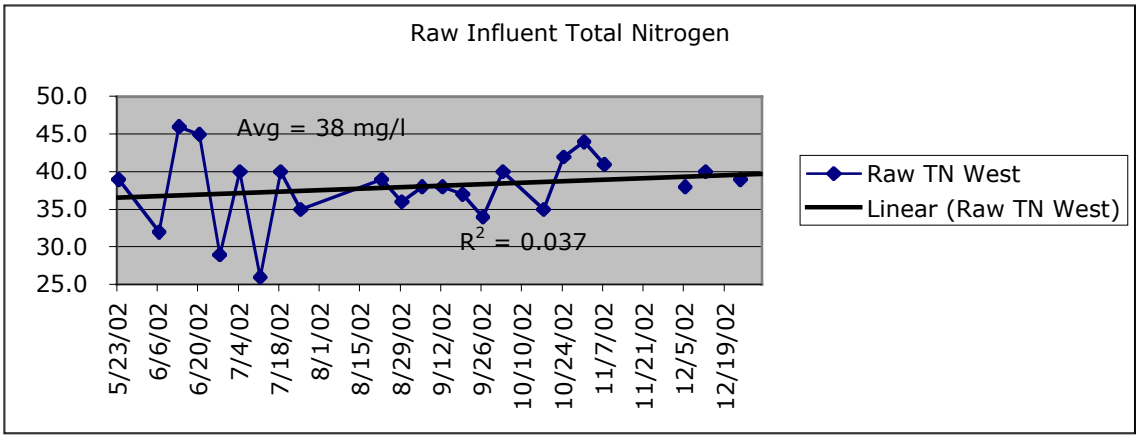

Figure 4b Total nitrogen in septic tank effluent

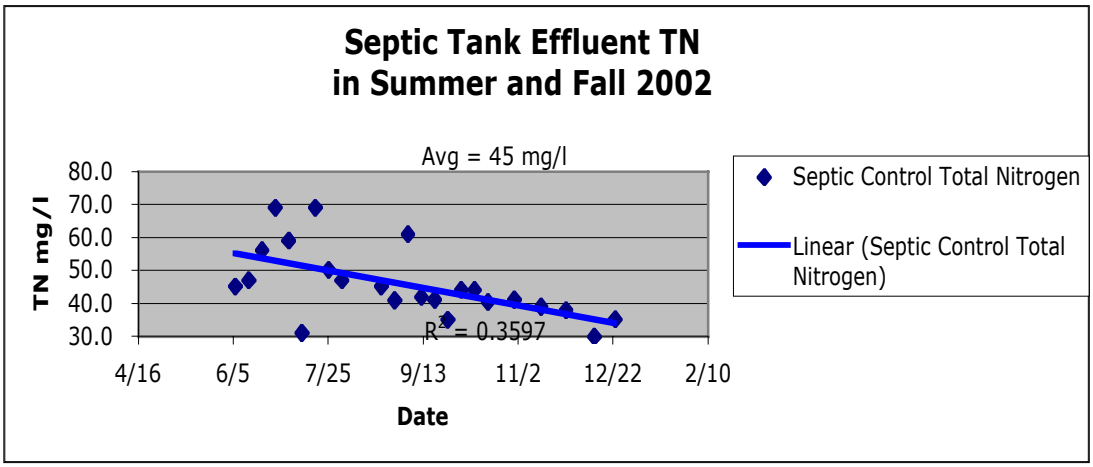

Figure 4c Total nitrogen in septic tank effluent after 24 inches of sand

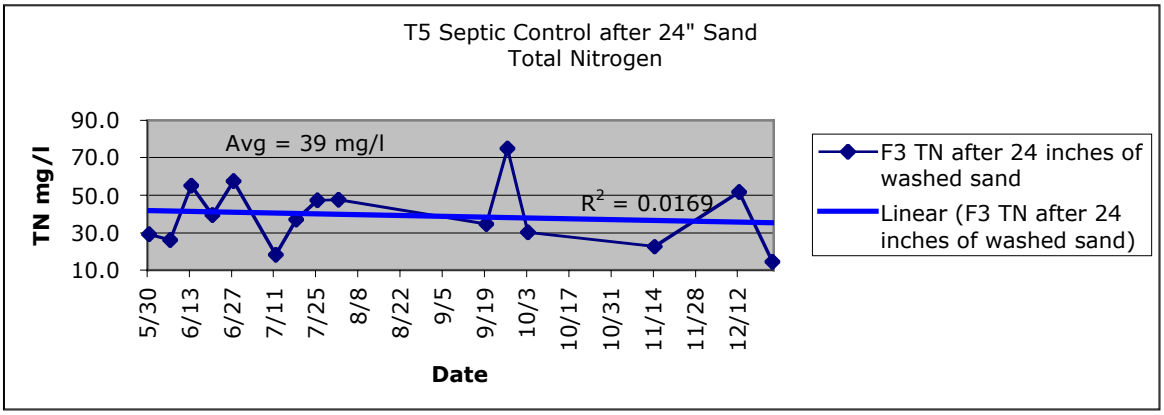

Figure 5 MASSTV Recirculating Sand Filter (RSF) Total Nitrogen by Month

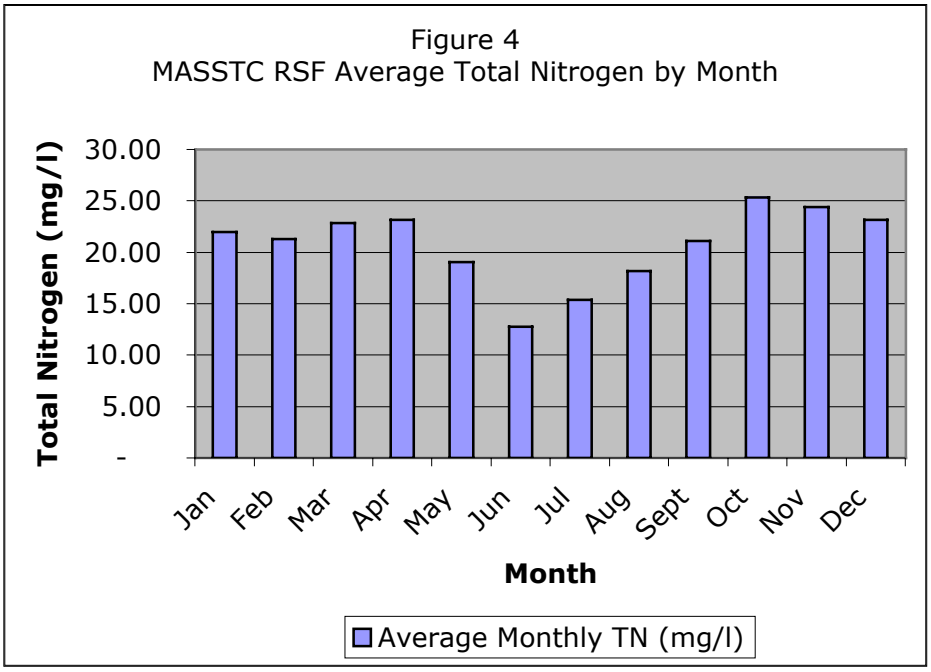

Figure 6

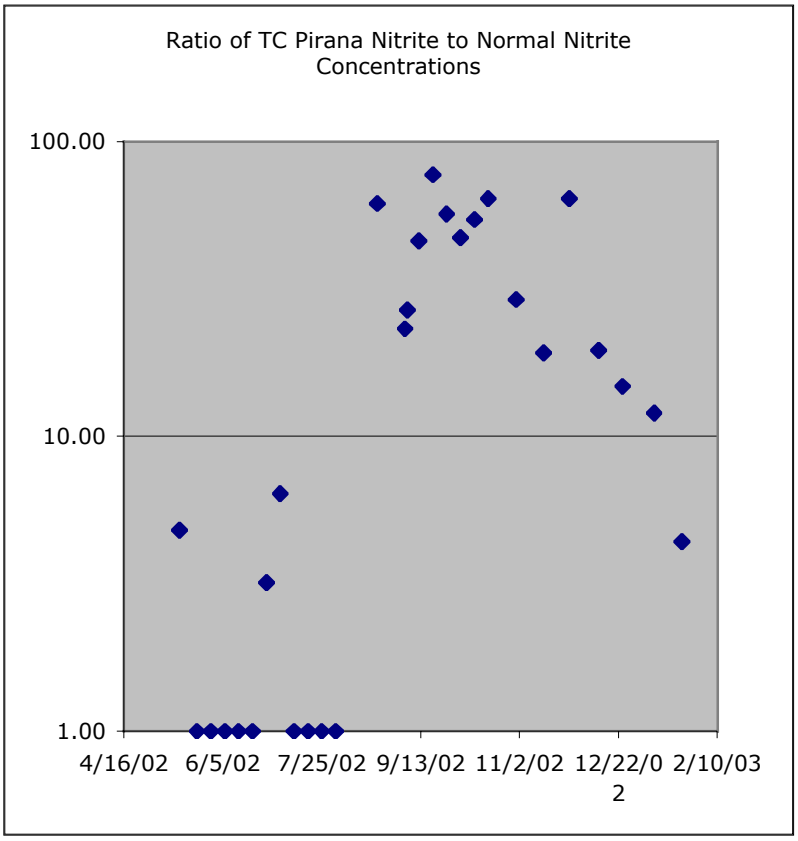

Figure 7 Tire Chip Pirana TN in TC Sump after 12 inches of sand

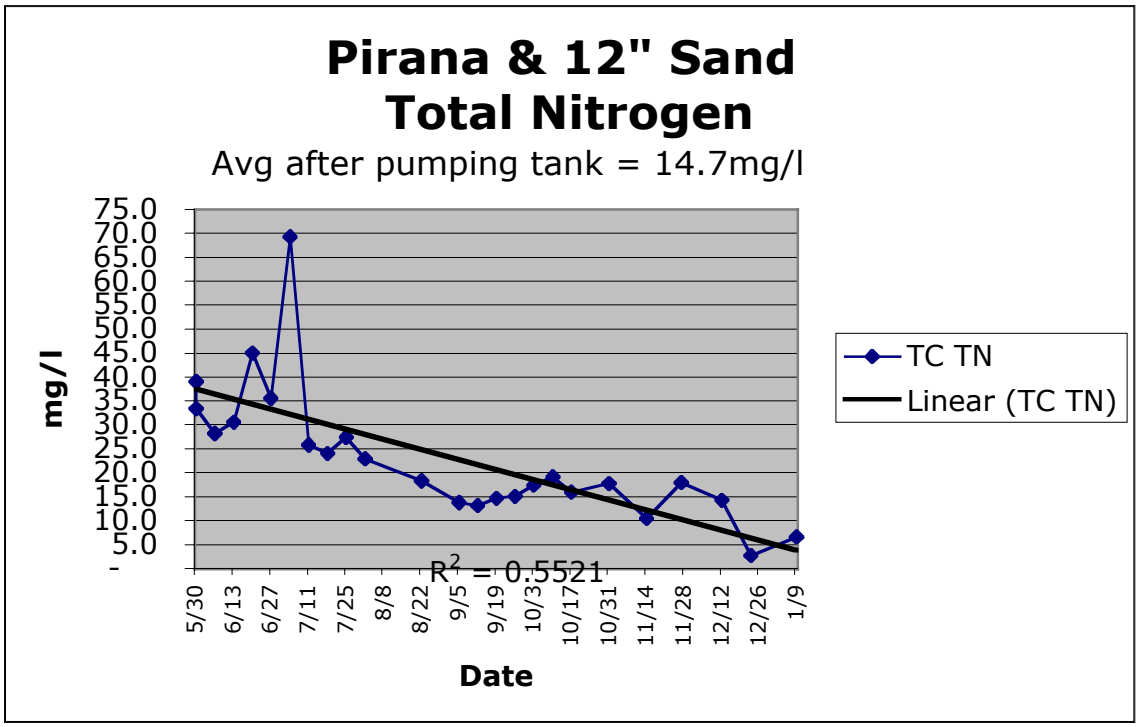

Figure 8 F2 Pirana TN After 2 feet of sand

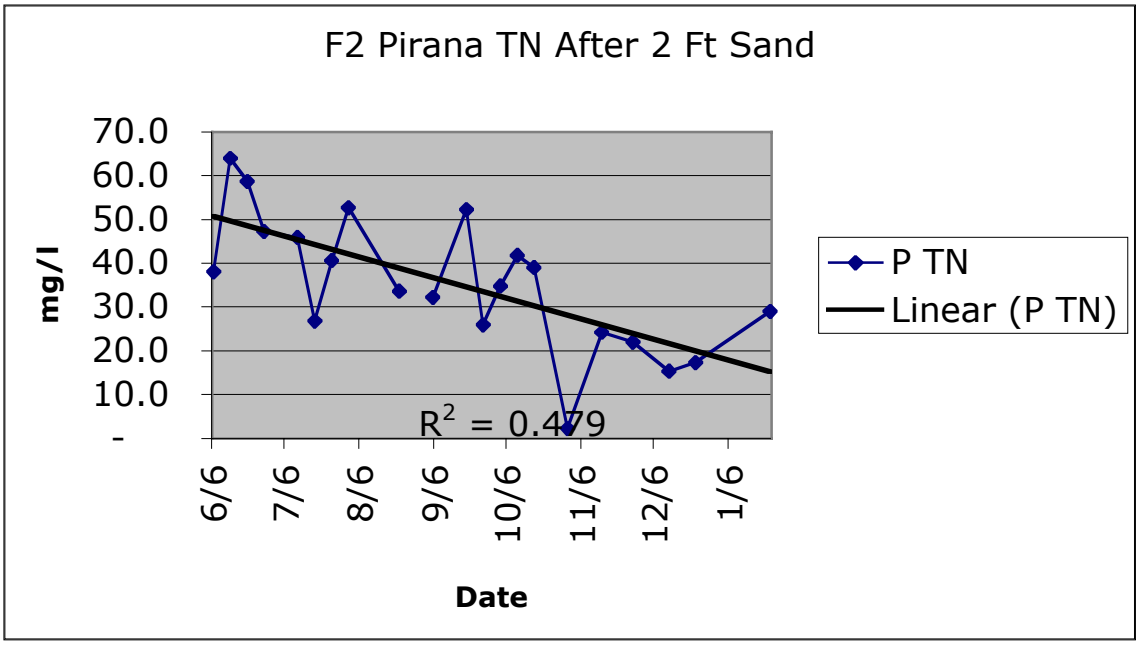

Figure 9 Absorption rates for Title 5 Control and for Pirana Effluent

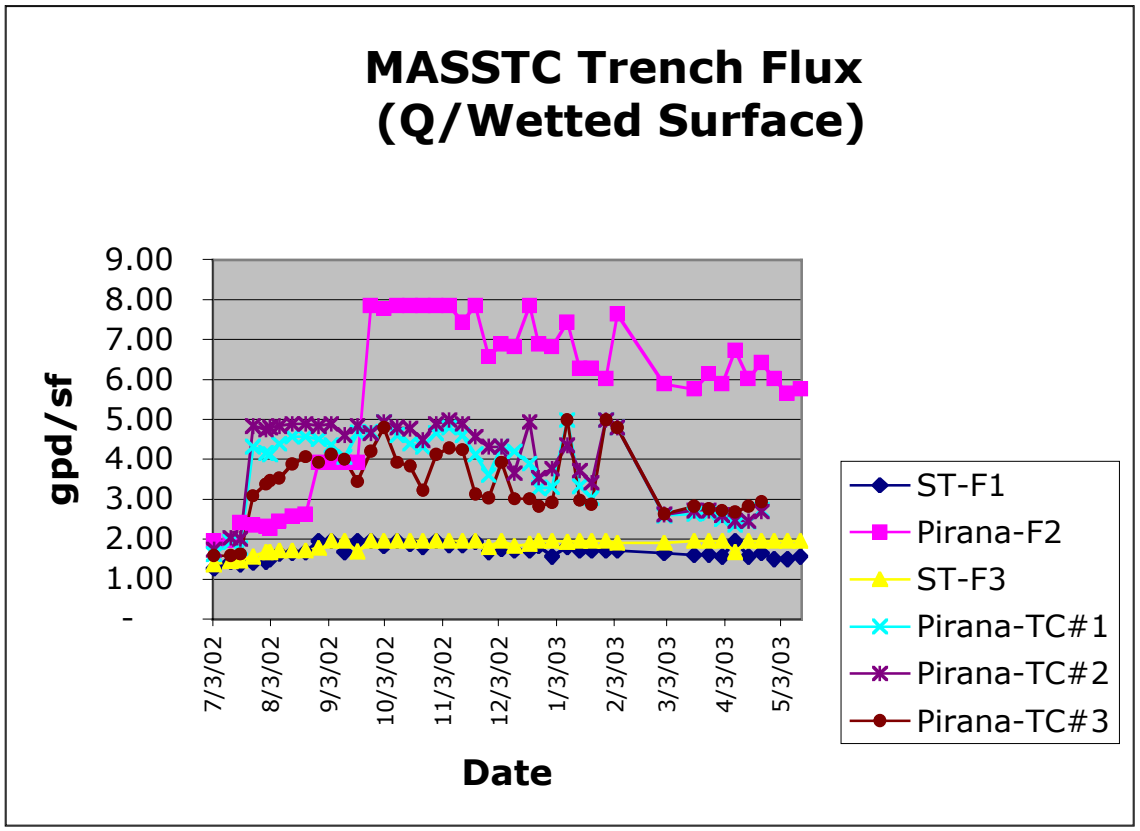

Table 2 F Trenches Sump Effluent Quality.

DateF Sum	Notes	Temp	pH	Sp Cond(uS)	DO	Total Nitrogen	TKN	NH4(mg/l)	Nitrate	Nitrite	BOD5	CBOD	TSS (mg/l)	Alkalinity (mg/l)	Fecal Coliform
10/10		19.60	5.38	407		30	0	1.1	30.2	0.03		1.0	3.5	7	5
10/17		18.60	5.41	443		28	0	0.1	27.3	0.03		1.0	0.6	2	0
10/31		17.10	5.36	394		5	0	1.0	4.7	0.03		1.0	0.5	12	10
11/27		-	5.50	-		19	0	0.2	19.0	0.05		3.0	2.2	7	10

Table 3 F Sump Mass Ballance

Estimated Pirana TN Concentration

75% effluent

Date	Composite F Sump TN (mg/l)	% Effluent	TN (mg/l) Corrected for Rainfall	% Septic	Septic 2' Pan TN (mg/L)	% Pirana	Estimated Pirana TN (mg/l)
10/10	30.48	80%	38.09	33%	39	67%	18.88
10/17	27.57	80%	34.47	33%	39	67%	15.26
10/31	4.97	80%	6.22	33%	39	67%	-
11/27	19.05	80%	23.81	33%	39	67%	4.60
1/22/03	15.20	75%	20.27	33%	39	67%	1.06