Water Resources Center
Annual Technical Report
FY 2003

Introduction

The Water and Environmental Research Center (WERC) at the University of Alaska Fairbanks (UAF) is presently housed in the Institute of Northern Engineering (INE). INE is a stand-alone research institute where the Director of WERC reports directly to the Provost. There is a reorganization effort ongoing at UAF in engineering, so this may change in the near future. The Water and Environmental Center continues to prosper and grow. At present, the basic USGS 104B grant program represents between 3 and 4 % of WERCs total annual budget. Currently there are about 12 faculty that are very active in WERC and about 6 that have minor roles. There are over 30 graduate students; about 20 % are PhD students. An additional ten professional people fill various technical research support functions.

Research needs across the State with three major climate zones (maritime, continental and arctic) are quite diverse and variable. Resource development drives much of the required research. High demand for water in concentrated areas (such as oil development in the Arctic) is an important challenge. Also, the high latitude position of Alaska with the projected amplification of global warming has made Alaska an excellent natural research observatory to document change. Issues of integrated water, wastewater, power and heating systems for rural villages are always an ongoing challenge. WERC faculty derive 54 % of their research dollars from federal agencies (NSF, DOE, NASA, USFS, EPA, NOAA etc.), 10 % from private sources, 29 % from the state and the remainder from a variety of sources.

Faculty, staff and students in WERC continue to be very active in communicating their research results to the various scientific communities and public. This is done through a variety of venues from technical conferences (ASCE, AGU, AWRA, etc.) to presentations in villages and K-12. Results are also distributed on our website (http://www.uaf.edu/water/) and through an electronic newsletter. On the WERC website are also hydrological and meteorological data (near real time) from about 40 field sites distributed around the state. This data is made available for use by others.

Research Program

During the research year 2003-2004 (March 1, 2003 to February 28, 2004), four projects were funded through the USGS 104B program. Since these grants are relatively small in size, they are primarily used for graduate student support. The cost per student is about $25,000 (stipend plus tuition) per year. This allocation allows us to fund between three to four research projects (most years some additional funds are added from outside sources to increase the total funds available.

The four funded projects are:

Molecular Characterization of Organic Matter in Soil Leachates from the Caribou-Poker Creek Watershed by Daniel M. White and Kenji Yoshikawa.

Investigation of Fouling in Membrane Bioreactors for Wastewater Treatment by Silke Schiewer.

Investigation of Immiscible Fluid Movement through Frozen Porous Media by David L. Barnes and Yuri Shur.

Luminescent Bacteria: A New Water Quality Issue? by Joan Braddock.

Results from the study of White and Yoshikawa above demonstrate that the signature of naturally occurring organic leachates can be used as a tracer to determine subsurface flow pathways. This tool can be used in a permafrost environment where groundwater flow has been altered due to the presence of frozen soils. Although people are much more environmentally aware, hydrocarbon spills continue to a problem across Alaska. Where permafrost and seasonally frozen soils are predominant, little is known about the interaction of these two immiscible fluids. Barnes and Shur are looking at this unique problem. The low temperatures associated with the Alaskan environment have the impact of reducing the natural rate of bioremediation. New techniques such as membrane bioreactors are being investigated to reduce the flux of pollutants to the natural environment. Schiewer is examining in the laboratory ways to reduce fouling of membrane bioreactors. Rural people in Alaska rely heavily on subsistence fishing of salmon for both family use and for feeding dogs. In 2001 subsistence fisherman reported that salmon on the drying racks were glowing in the dark (isolated cases had been reported earlier. Braddock and her graduate student set out to study the occurrence of luminescent bacteria. There are no known pathogenic luminescent bacteria.

Investigation of Immiscible Fluid Movement Through Frozen Porous Media

Basic Information

Title:	Investigation of Immiscible Fluid Movement Through Frozen Porous Media
Project Number:	2002AK1B
Start Date:	5/1/2002
End Date:	5/8/2004
Funding Source:	104B
Congressional District:	Alaska
Research Category:	Ground-water Flow and Transport
Focus Category:	Toxic Substances, None, None
Descriptors:	
Principal Investigators:	David L. Barnes, Yuri Shur

Publication

1. Barnes, D.L., S.M. Wolfe, and D.M. Filler. Equilibrium Distribution of Petroleum Hydrocarbons in Freezing Ground. Accepted for publication in Polar Record.
2. Wolfe, S. M., D. L. Barnes, Y. Shur, and D. M. Filler. 2003. Two-Dimensional Movement of Immiscible Fluids Through Frozen Soil. In Proceedings of the 3rd Assessment and Remediation of Contaminated Sites in Arctic and Cold Climates Conference.
3. Barnes, D. L. and D. M. Filler. 2003. Spill Evaluation of Petroleum Products in Freezing Ground. Polar Record, 39(210):1-6.
4. Wolfe, S.M., D.L. Barnes, and Y. Shur. 2003. The Effect of Pore Ice on the Movement of Petroleum Hydrocarbons Through Frozen Soil. Presented at the 54th American Association for the Advancement of Science Arctic Science Conference, Fairbanks, Alaska.
5. Wolfe, S.M., D.L. Barnes, and Y. Shur. 2003. The Movement of Petroleum Hydrocarbons Through Frozen Soils. Presented at the Third Annual INRA Subsurface Science Symposium, Salt Lake City, Utah.
6. Wolfe, S., D.L. Barnes, Y. Shur, and D. Filler. 2004. Two Dimensional Movement of a Petroleum Product Through Frozen Soil. Presented at the Contaminants in Freezing Ground 4th International Conference, Fairbanks, Alaska.
7. Barnes. D.L., S.M. Wolfe, and D.M. Filler. 2004. Movement of Petroleum Hydrocarbons Through Freezing Soils. Presented at the Contaminants in Freezing Ground 4th International Conference, Fairbanks, Alaska.

Overview

Frequent releases of petroleum in regions experiencing seasonal or permanent frozen ground have prompted engineers and scientists to investigate the physical and chemical aspects of immiscible fluid movement in frozen ground. An understanding of these processes is required to engineer methods for protecting both the environment and human health. In Alaska, several recent relatively large petroleum releases in areas of permanently and seasonally frozen ground illustrate the importance of gaining a better understanding of the physical and chemical processes controlling the migration of these compounds through frozen ground to possible freshwater sources.

One key facet of this topic is the relationship between structures of frozen soil (cryogenic structures), moisture content (frozen and unfrozen) and the permeability of soil to petroleum products. It is known that freezing and thawing are very important factors of soil structure. Freezing and ice formation produce cryogenic structure and desiccation of soil below freezing front leads to shrinkage and cracking of soil. Thawing of frozen soil and melting of ice inclusions does not completely destroy soil structure formed during freezing. So-called post-cryogenic structure is the structure of thawed soil affected by freezing and thawing. Such structure makes soil of the active layer highly permeable, especially in horizontal direction.

Proper cleanup response to petroleum releases requires a good understanding of how these cryogenic processes will affect petroleum migration through the subsurface. Currently little is known on this subject. Critical questions include how does pore ice influence the lateral and vertical migration of petroleum, how does soil-water content prior to freezing influence the migration of petroleum, and how do the physical and chemical properties of petroleum hydrocarbon influence the migration? This study is producing results that help answer these relevant questions.

Problem & Research Objectives

The objective of this research is to gain a better understanding of how the movement of immiscible fluids, specifically light non-aqueous phase liquids (LNAPLS), through porous media is influenced by seasonal freezing. This objective is being accomplished through laboratory testing and quantitative analysis.

To study contaminant movement in freezing soil it is important to recognize the main patterns of frozen media:

1. The frozen active layer above unfrozen soil;
2. The frozen active layer above permafrost;
3. The unfrozen active layer above permafrost.

Permeability (saturation) of permafrost depends on soil genesis, soil type, and genesis of permafrost (syngenetic or epigenetic). Soil of the same type can be practically unsaturated or even over-saturated with ice depending on a genesis of permafrost.

The structure of the active layer is greatly dependent upon the freezing system (open or closed). The active layer over permafrost is usually formed in the closed system. In areas of cold permafrost, where soil in the active layer freezes downward from the surface and upward from the bottom of the permafrost table, the upper and lower parts of the active layer can be saturated with ice; the middle part of the active layer is dry and has high open porosity due to vertical and horizontal cracks. In areas of warm permafrost, freezing proceeds only downward from the surface and ice-saturated upper part of the active layer is often underlain by dry soil with cracks. Also, thickness of saturated soil depends on numerous factors; there are several

general patterns in soil structure in the active layer and permafrost, and study of these can provide understanding of permeability of frozen soil. All of these effects influence the lateral and vertical movement of petroleum product that is inadvertently released to frozen soils. We are developing a better understanding of how pore ice influences this movement by using laboratory scale soil flumes performing laboratory studies on the movement of LNAPL through frozen soil.

Characteristics of LNAPL through unsaturated soils and their fate in the subsurface have been under study for several decades. Many definitive manuscripts have been written that describe the physical and chemical fate of these compounds (predominantly petroleum hydrocarbons) in soils that do not experience temperatures below zero degrees Celsius. However, little is known about the chemical fate of these compounds in soils that experience seasonal freezing and thawing. Hence, a quantitative study that is focused on quantifying the chemical fate of petroleum hydrocarbons was undertaken in this research project. Chemical fate in this study is defined as the partitioning between the volatile, dissolved, adsorbed, and liquid fractions in unsaturated soils. The relative fraction of each of these different phases influences the gaseous phase, aqueous phase, and liquid phase migration of petroleum hydrocarbons. A quantitative analysis of phase partitioning in freeze-susceptible soils was used in this study to better understand the influence freezing has on the chemical and physical fate of petroleum hydrocarbons in soil.

Laboratory Methodology

The laboratory flumes used for this study were designed and constructed after extensive literature review on the topic of immiscible fluid migration in soils (Figure 1). Two flumes were constructed. To investigate how pore ice impacts the migration of petroleum hydrocarbons, the flumes were filled and compacted with sand wetted to uniform water contents and placed in the cold room at a temperature of approximately -5° C. The properties of the sand (water retention characteristics, permeability, porosity, density) were measured in the laboratory. Once frozen, 100 ml of colored petroleum was introduced into the column. Progression of the resulting immiscible fluid plume was tracked using time-lapse photography. Two different uniform sand grain sizes were tested at two initial water contents (prior to freezing). Table 1 provides the parameters for each of these tests.

Figure 1. Soil flumes prior to filling with soil.

Table 1. Soil Properties for Each Test Conducted.

Test	Average Grain Size (mm)	Ice Saturation (%)
1	0.60	26
2	0.60	54
3	0.212	26
4	0.212	54

Soil flumes were prepared for testing by mixing the sand with the proper mass of water and compacting the wetted sand in a horizontally placed flume. The packed flume was moved into the cold room and allowed to freeze at –5 C for several hours. Once frozen, the soil flume was placed vertically into supports and 100 ml of colored LNAPL was introduced into a depression made in the top of the soil. The LNAPL was poured in at a steady rate such that all of the LNAPL was in the soil in approximately five seconds. Back lighting added the time lapse photography as the LNAPL moved through the soil.

Results and Discussion

Figure 2 shows the results from tests run on medium sand (Test 1 and 2). As can be seen in this figure, the plume of LNAPL is wider in the flume containing a higher ice saturation. Most likely this result is due to pore ice obstructing the flow of LNAPL by blocking random pores with ice as it progresses down, causing the plume to spread laterally. Movement laterally may also be caused by increases in capillary pressure gradients due to the decrease in average pore space radius. In the flume with the relatively lower ice saturation, the plume measured a maximum of 0.19 m, compared to 0.43 m maximum width in the flume with a relatively greater ice saturation. At the relatively higher ice saturation, the LNAPL ponded during introduction into the flume; however no ponding occurred in the flume with relatively lower ice saturation. Once again this result is most likely due to the decrease in pore space of the sand with greater ice saturation. The maximum depth of penetration was greater (more than 1.2 m) in the flume with relatively greater ice saturation in comparison to the flume with lower ice saturation (0.84 m).

Results from Tests 3 and 4, which were performed on the finer grained sand are shown in Figure 3. Excluding the slight bend of the LNAPL plume to the right in the flume with the relatively greater ice saturation, the resulting plumes for both of these tests are quite similar. Both plumes have a maximum width of around 0.30 m (0.28 m in the flume with relatively lower ice saturation, compared to 0.30 m in the flume with the greater ice saturation). Possible reasons for the consistency in the plume shape for the relatively fine grained soil may be due to blockage of pore space and pore channels by ice for both values of ice saturation tested. Given that the average pore diameter in the fine grained sand is most likely smaller in contrast to the relatively coarse grained sand tested, water expanding into ice has a greater chance of filling pore channels in the fine grained sand flumes. This blockage of pore space appears to be occurring at both ice saturations tested. The apparent blockage of pores is further validated by the limited downward progression of the LNAPL in both tests (Tests 3 and 4 in Figure 3).

These laboratory tests and field studies allowed us to gain a better understanding of the mechanisms that control LNAPL plume migration into a frozen soil. Once a LNAPL has been introduced into soil, the chemical and physical fate of the contaminant during cyclic freeze thaw processes is also of interest. Past documented laboratory measurements have shown movement

of petroleum hydrocarbons to the freezing front in contaminated freezing soils. The mechanisms that are, in part, responsible for the increased contaminant concentration at the freezing front are illustrated in this study with a mass balance model. Results developed in this study show that this concentration increase is due to exclusion of petroleum hydrocarbon from the crystalline ice structure and from physical displacement of liquid petroleum hydrocarbon from the pore space as water freezes and expands into ice. A manuscript discussing this model and the results has been accepted for publication in *Polar Record*.

Conclusion

Through analytical studies, laboratory tests, and field results the influence pore ice has on the movement of LNAPL into frozen and freezing soils has been illustrated. Results from this research indicate that the presence of pore ice has three main effects on the movement of LNAPL through a frozen soil, depending on the pore geometry prior to containing ice and the magnitude of ice saturation. These effects are:

(1) Random blockage of pore space causing LNAPL to spread laterally;
(2) Decreasing of pore channel diameters, resulting in increases in capillary pressure and associated lateral movement; and
(3) Blockage of pore space and channels restricting overall movement.

Further studies that are required are to determine how pore ice is structured in the pore space and how the structure influences LNAPL flow and to determine how to incorporate the presence of pore ice into the flow equations.
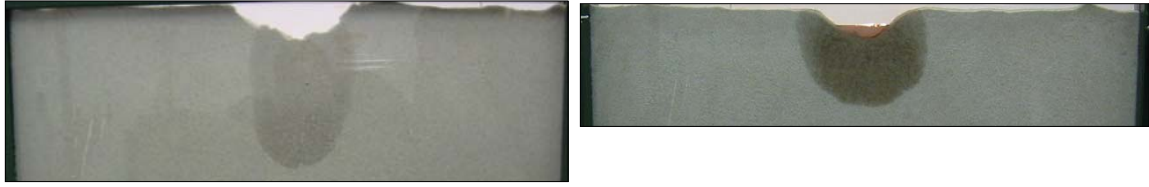
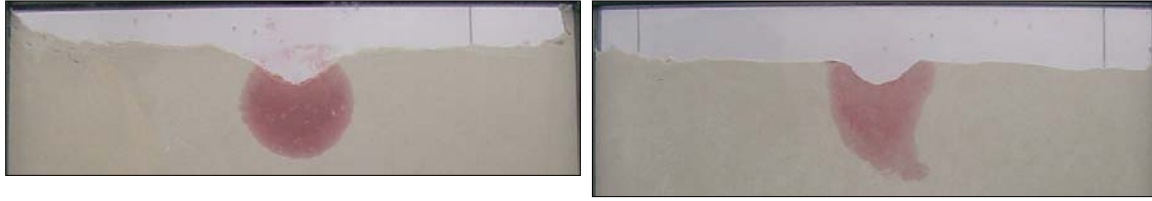Photographs taken at approximately one minute.
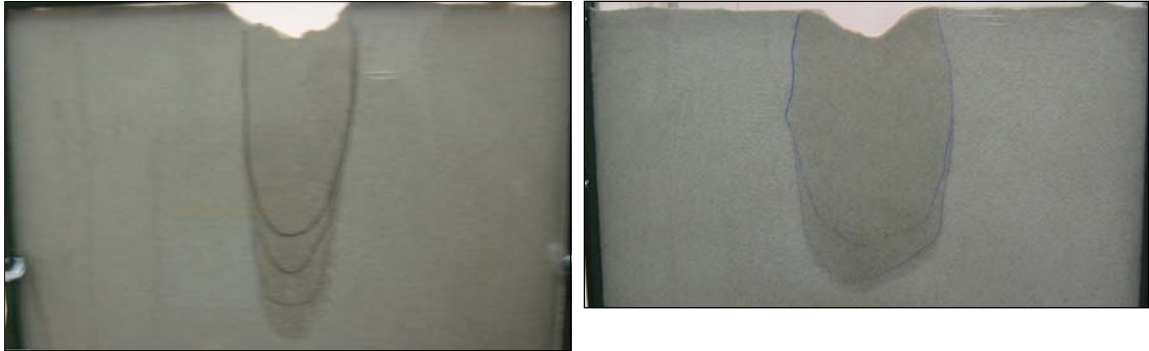
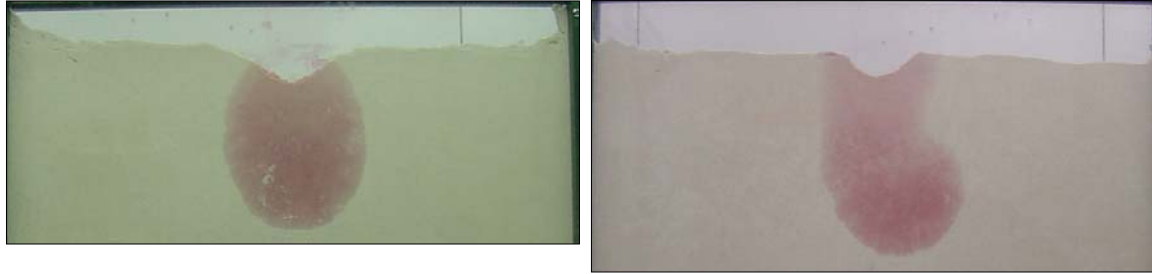Photographs taken at approximately 20 minutes.

Photographs taken at approximately one day.

Figure 2. Movement of LNAPL through frozen medium grade sand at two different ice
saturations. Columns on left side of figure contained an ice saturation of 26% (Test 1).
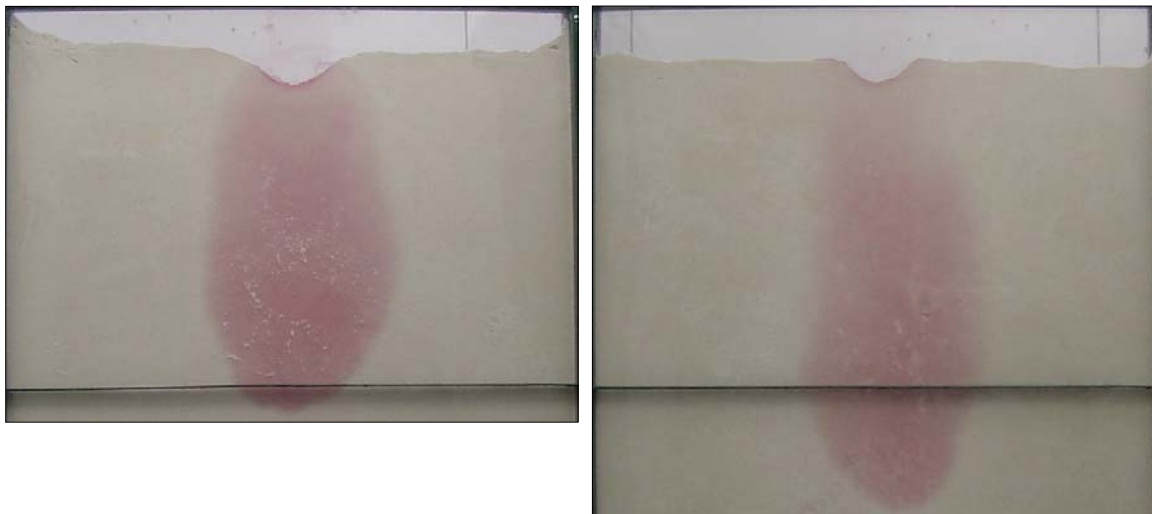Columns on the right side of figure contained an ice saturation of 54% (Test 2).

Photographs taken at approximately one minute.

Photographs taken at approximately 20 minutes.

Photographs taken at approximately one day.

Photographs taken at approximately two days.

Figure 3. Movement of LNAPL through frozen fine grade sand at two different ice saturations. Columns on left side of figure contained an ice saturation of 26% (Test 3). Columns on the right side of figure contained an ice saturation of 54% (Test 4).

Molecular characterization of organic matter in soil leachates from the Caribou Poker Creeks Watershed

Basic Information

Title:	Molecular characterization of organic matter in soil leachates from the Caribou Poker Creeks Watershed
Project Number:	2002AK4B
Start Date:	3/1/2003
End Date:	3/1/2004
Funding Source:	104B
Congressional District:	Alaska
Research Category:	Ground-water Flow and Transport
Focus Category:	Hydrology, Methods, Groundwater
Descriptors:	organic geochemistry, pyrolysis-GC/MS, DOC fingerprinting
Principal Investigators:	Daniel M. White, Kenji Yoshikawa

Publication

1. White, D., K. Yoshikawa, and D. Garland, (2002), Fingerprinting dissolved organic matter to support hydrologic investigations, Cold Regions Science and Technology, 35:27-33.
2. Autier, V. and D. M. White, (2004), Cd mobility in boreal soils, Journal of Hazardous Materials, 106(2/3):149-155.
3. Vincent Autier (2002), Predicting Contaminant Transport Pathways in the Caribou-Poker Creeks Watersheds. MS. Civil Engineering, Thesis, University of Alaska Fairbanks.
4. Autier, V. D. White, and D. S. Garland, (2002), Seasonal variation of organic matter chemistry in a boreal watershed, 53rd Arctic Science Conference, American Association for the Advancement of Science, Fairbanks, Alaska.
5. Seelen, C., White, D.M., K. Yoshikawa, V. Autier, (2004), How chemical differences in soil leachates relate to groundcover, Proceedings of the 2004 Cold Regions Engineering and Construction Conference.

Problem and Research Objectives:

In order to protect the quality of freshwater, sediments, and soils, it is imperative to identify contaminant sources and transport pathways before the contamination becomes wide-spread. While the Clean Water Act focuses on cleaning-up point sources of contaminants, non-point sources are much more difficult to control. Examples of non-point sources include agricultural runoff, road runoff or atmospheric deposition.

Groundwater aquifers are replenished thorough surface recharge. Certain recharge areas in a watershed may have soils with higher hydraulic conductivity than others or may, for other reasons, be particularly vulnerable to the influx of non-point source pollutants. Dissolved organic matter (DOM) is believed to serve as a vector for heavy metal transport in groundwater (Koopal, et al (2001). The properties of DOM are a function of the original parent litter (e.g., leaves, roots) as well as physical, chemical and biological transformations that occur in the soil (Lehtonen et al, 2001; Joly et al., 2001; Schulten and Gleixner, 1999; Page et al., 2002; Suominen, et al., 2003). Whether or not DOM can serve to mobilize non-point source pollutants in a recharge area depends on the properties of the DOM, the pollutant, and the soil/water chemistry.

There were two components to this study. The first was to determine the role of DOM chemistry in the mobilization of contaminants. The second was to determine how groundcover type impacts DOM chemistry in a boreal watershed. Armed with these two pieces of information, as well as information on soil properties, one could identify certain recharge areas in a watershed that may be particularly vulnerable to subsurface mobilization of non-point source pollutants. The subject of this report is the second component of the study, developing a link between cover vegetation and DOM.

Methodology

Duplicate soil cores were collected from 11 sample sites in the Caribou Poker Creek Watershed (CPCRW) in September, 2002. The CPCRW is a 104 km^2 watershed near Fairbanks, Alaska that was set aside for research. Sampling sites were selected so as to represent a variety of vegetation found within a boreal watershed.

Soil leachate was collected from each core using a leaching procedure modified after Wagai and Sollins (2002). Each core was mixed with 7.5L of water that had been treated by reverse osmosis. The soil solution was then sieved to break down aggregates and homogenize the solution. The soil-water solution was stirred and left soaking overnight. The soil-water solution was centrifuged and filtered through 0.45 micron glass filter. The water and DOM were dried under vacuum at 40°C to prevent loss of organic matter to volatilization. The solids remaining from the drying procedure were collected and subjected to molecular fingerprinting by py-GC/MS.

Pyrolysis method

Py-GC/MS was conducted with a CDS Model 2500 pyrolyzer and autosampler in tandem with a gas chromatograph/mass spectrometer. During pyrolysis, the sample was heated from a starting temperature of 25 ºC to 700 ºC in 0.1 seconds and held at a constant 700 ºC for 9.9 seconds. The pyrolysis reactor was mounted to an HP 5890 Series II GC, with a Supelco SPB 35 (35% Ph Me silicon) column, 60 m x 0.25 m x 0.25 μm. The GC interface temperature was set at

235 ºC. The GC temperature program was 40 ºC held for 30 minutes, 1 ºC/min increase for 80 min, 20 ºC/min increase for 50 minutes, 10 ºC/min increase for 10 minutes and then held for 10 minutes. The GC was plumbed directly to an HP 5971A Series Mass Selective Detector on electron impact (EI) mode. The MS scanned mass units 45 to 650. All mass spectra were compared to the NBS54K spectral library. Helium served as a carrier gas at a flow rate of 0.5 cm^3/minute. Each sample was injected with a split ratio of 1:50.

Pyrolysis of NOM produces a complex chromatogram, or "pyrogram," containing hundreds of peaks that represent individual pyrolysis products. The pyrolysis products may have been originally present in the soil, or they may be thermal breakdown products of large macromolecules. It has been found in previous research that a small portion of the pyrolysis products can be used to draw differences between individual, or groups of soil and water samples (White and Beyer, 1999, White et al, 2004). A total of 14 pyrolysis products were selected from each pyrogram to identify similarities or differences in DOM from the 11 soil leachate samples. HP Chemstation software was used to quantify the relative abundance of each of the 14 pyrolysis products. The pyrolysis products were compared on a relative abundance basis and were not individually quantified on a per soil mass basis. In student t-tests, certain pyrolysis products with similar structures were grouped to make the analysis easier. In these cases, the 14 pyrolysis products were grouped into six classes (i.e., benzenes, nitriles, methyl cyclopentenone, phenol, furfural, and carbohydrates). While "benzenes" and "phenols" were used to identify two of the classes of pyrolysis products, there was not benzene or phenol itself in the leachates. Rather, these class names refer to the base structure of the pyrolysis products included in the particular class.

Statistics

The statistical packages in Microsoft Excel© and Minitab v. 14™ were used to analyze the data. Student t-tests were based on a one-tail normalized distribution using Microsoft Excel. Principal component analysis was conducted utilizing Minitab v. 14™. The variables were the relative abundance of each pyrolysis product from each soil leachate. The first and second components were a linear combination of all soil leachates and the relative abundance of each pyrolysis product in combination with the presence or absence of individual vegetation types at each sampling site.

Principal findings and significance

Figure 1 represents the relative abundance of each the six classes in each leachate sample. Table 1 contains a description of the vegetation cover associated with each soil leachate described in Figure 1. The topographic descriptor identifies whether the sample was taken from a north or south facing slope. The tree, shrub and moss columns list the primary vegetation found at each sampling site.

Student t-tests

In order to separate the influence of different vegetation types on the soil leachates, student t-tests were performed. Soil leachates were split into three groups based on the type of tree, shrub, or ground cover at the sampling site. Student t-tests were then performed on each of the three groups.

Student t-tests for the moss groups (Table 2) revealed statistically significant differences between six pyrolysis product classes in the soil leachate and the different types of moss at the sampling site. When leachates from sites with feather moss were compared to those with sphagnum moss, the relative percentages of nitriles, carbohydrates, methyl cyclopentenone, and phenol were significantly different. Leachates obtained from sites containing feather moss and lichen were significantly different among the benzene and methyl cyclopentenone classes. When leachates from sites with sphagnum moss were compared to those with lichen, the relative percentages of benzenes, methyl cyclopentenone, and phenol were significantly different. Student t-tests found a significance level of $p < 0.15$ for 9 of the total 18 comparisons with moss type as a basis. The classes of benzenes, methyl cyclopentenone, and phenol explained most of the difference among moss ground cover. Student t-tests revealed that the relative abundance of pyrolysis product classes were not significantly different when comparing leachates with different tree or shrub cover.

The student t-tests indicated that different types of mosses are more important to the soil leachate chemistry than tree or shrub cover. Therefore, when looking for differences in vegetation type that may make an area more or less vulnerable to contaminant mobilization, the biggest differences will be seen when comparing the different moss types.

Multivariate Statistics

Principal component analysis (PCA) uses the ability to organize data sets into multidimensional planes to better understand patterns and groupings that exist inherently within data sets. In this research project, PCA was used to recognize patterns among leachates with similar and dissimilar vegetation and the 14 pyrolysis products selected from the py-GC/MS pyrograms. The score and loading plots from PCA have two "component" axes. The first and second component axes represent the unique linear combination of each of the 14 pyrolysis products in the soil leachates and the vegetation type. These component axes capture the first and second highest amount of variance within the sample set.

Mosses

Score plot and loading plots in Figures 2a and 2b are the result of PCA analysis that considered the relative abundance of the 14 pyrolysis products from each soil leachate sample and the type of moss found at each sampling site. There was a strong relationship found between sphagnum moss and phenol present in soil leachate pyrograms. That is, when sphagnum was present, DOM in soil leachates produced relatively more phenol when subjected to py-GC/MS than DOM from leachates where sphagnum was not present. While lichen and dimethyl benzene were not well defined by the first component, they were positively defined by the second component. In addition, feather moss was positively defined by the presence of the pyrolysis product, benzofuran. As expected, feather moss and sphagnum moss were dissimilar in the first component. This combination of inputs separated the three mosses into individual areas within the loading plot.

When the score plot was combined with the loading plot, the sample sites corresponded with the projected input positions on the loading plot. Soil leachates P3 and LG fell on the score plot near where lichen moss was positioned on the loading plot. This was consistent with the fact that at both sampling sites, P3 and LG, lichen was the primary moss presence. Likewise leachates AP, UBS, and CSCP fell on the score plot near where feather moss fell on the loading

plot. Feather moss was the primary moss presence at AP, UBS and CSCP. This relationship between score and loading plots did not correlate so strongly for sphagnum moss in both first and second components. However, the position of sample sites P2 and H2 on the score plot corresponded to the sphagnum moss position on the loading plot with respect to the first component. This result suggests that the differences between the soil leachates tested could be principally explained by one pyrolysis product and one moss type.

Shrubs
      The principal component loading and score plots for all pyrolysis products and the influence of shrub cover is shown in Figures 3a and 3b. The loading plot for this specific set of inputs produces a striking pattern. The pyrolysis product phenol was inversely related to all shrubs with respect to the first component. Three of the four shrubs, blueberry, cranberry, and shrub birch, were grouped together in both the first and second components while arctic rose was positively influenced by the first component. When comparing the loading and score plots, soil leachates were not obviously positioned with respect to any groundcover. However, the only soil leachate with arctic rose at the sampling site, AP, fell on the score plot where arctic rose fell on the loading plot. This result suggests that one cannot link the pyrolysis products selected to any of the shrub vegetation except perhaps arctic rose. Additionally, the separation of phenol from the main group of inputs on the loading plot suggests that phenol had no relationship to shrubs and exists in the soil leachate due to other vegetation influences as was shown to in Figure 2a.
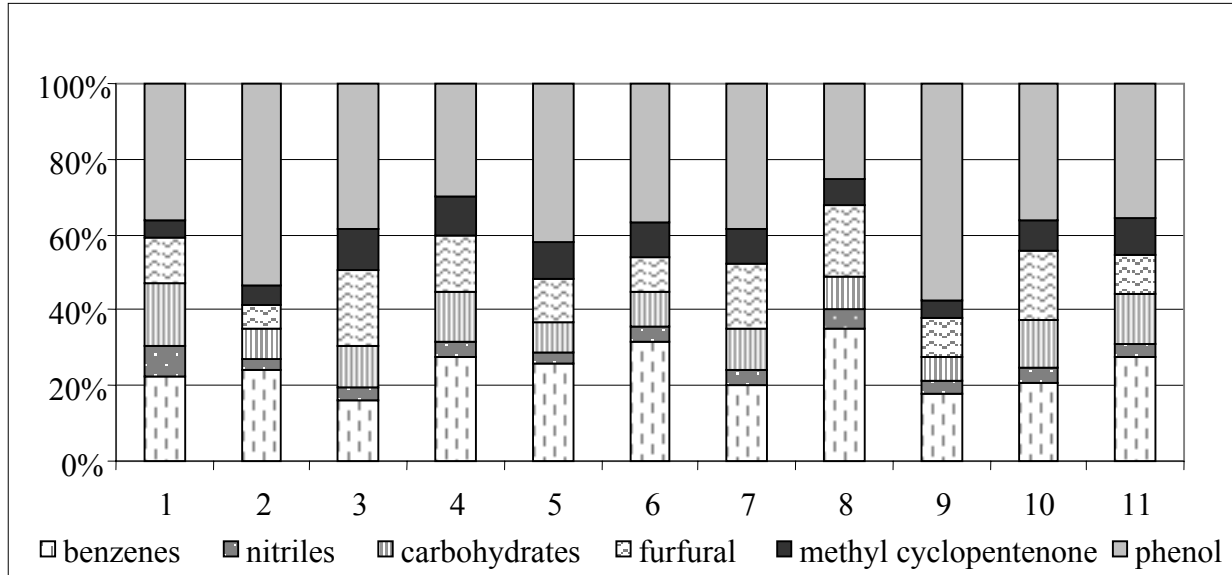
Trees
      Figures 4a and 4b include the loading and score plots for all pyrolysis products with respect to trees cover. The loading plot was strongly influenced by birch and black spruce as shown by their lone positions within the grid. The first component drew a difference between birch and willow, while the second component drew a difference between birch and black spruce.
      Birch trees did not relate to any of the 14 pyrolysis products. Willow related with pyrolysis products benzonitrile and naphthalene and was inversely proportional to phenol in the first component.
   The second component dominated differences within this loading plot. The second component showed similarities between white spruce and birch. Soil leachates SHAY2, SHAY1, and HWLB all fell on the score plot near where white spruce and birch fell on the loading plot. All three sample sites SHAY2, SHAY1, and HWLB had birch and white spruce. Aside from these samples, evidence of tree type was not seen using this method in the soil leachates.
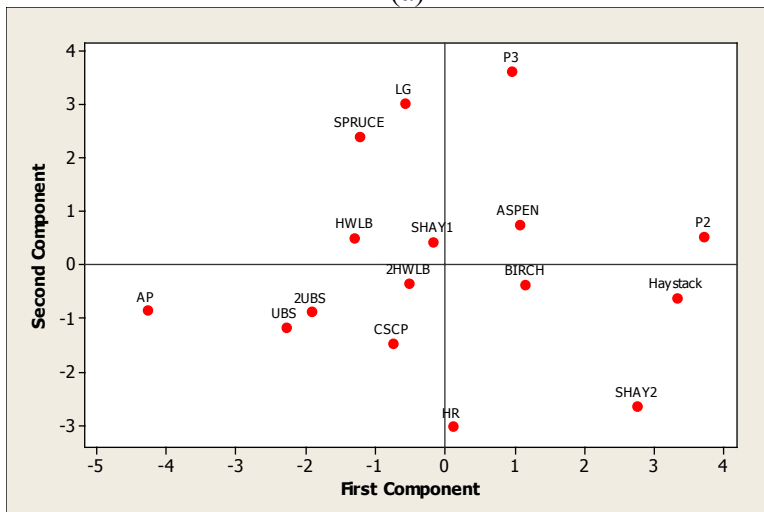
Figures and Tables

Figure 1. Relative abundance of pyrolysis product classes in each soil leachate (see Table 1 for sample site description.

Figures 2a and 2b. Loading and score plot for all pyrolysis products and mosses.
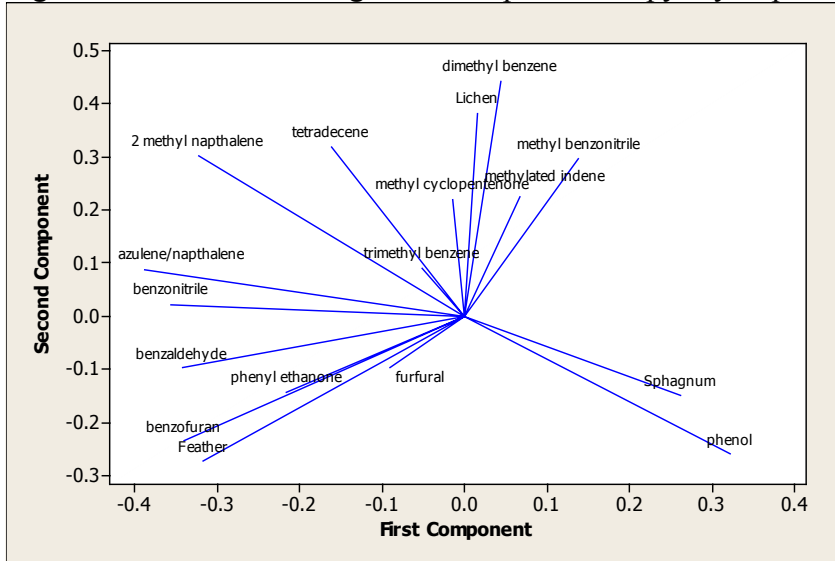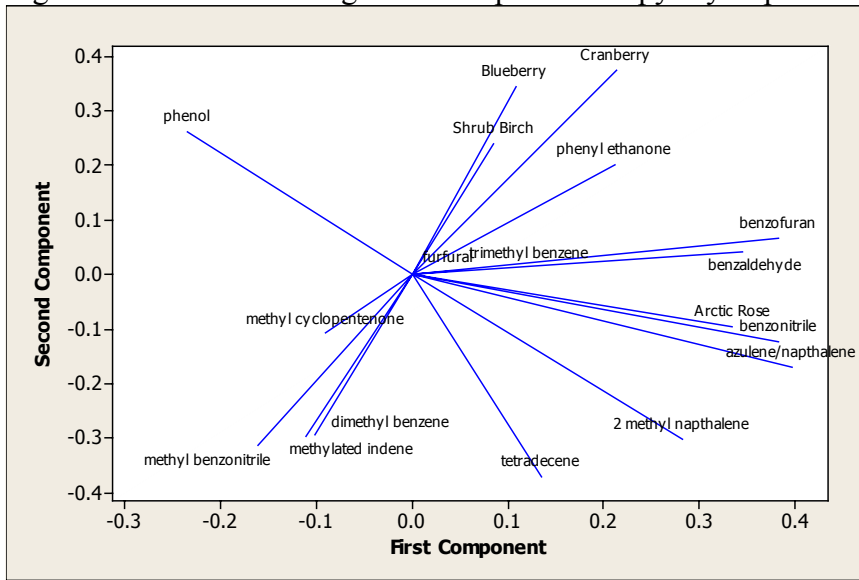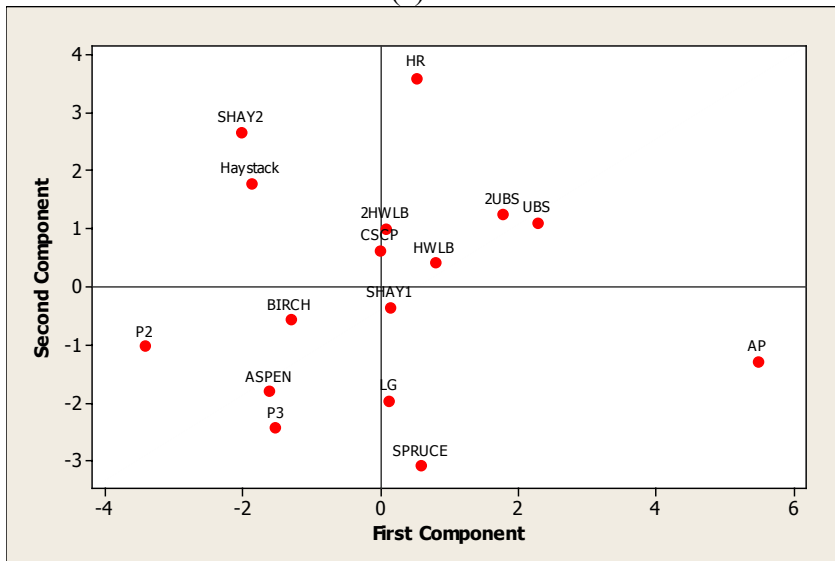

(a)

(b)

Figures 3a and 3b. Loading and score plot for all pyrolysis products and shrubs.
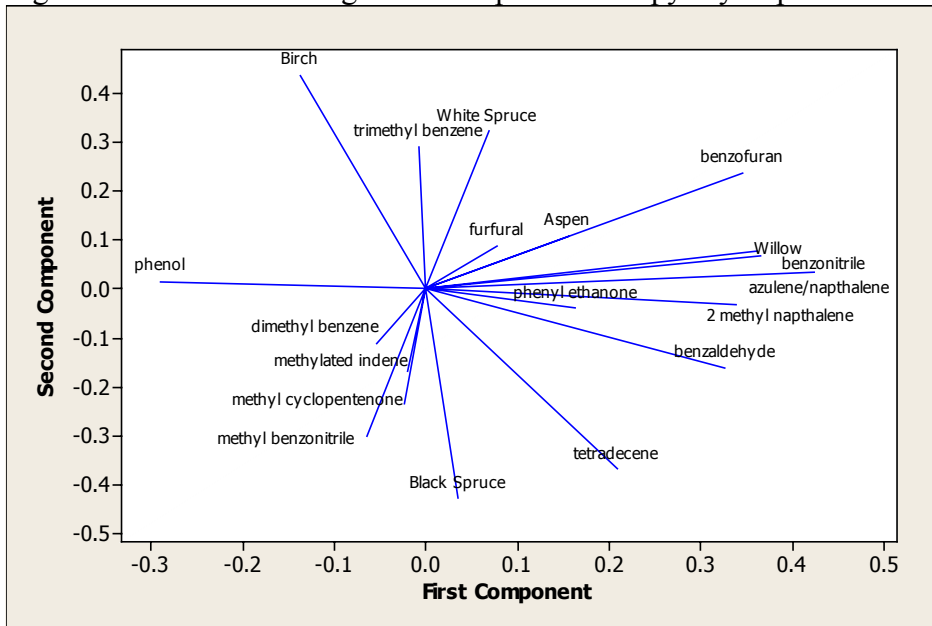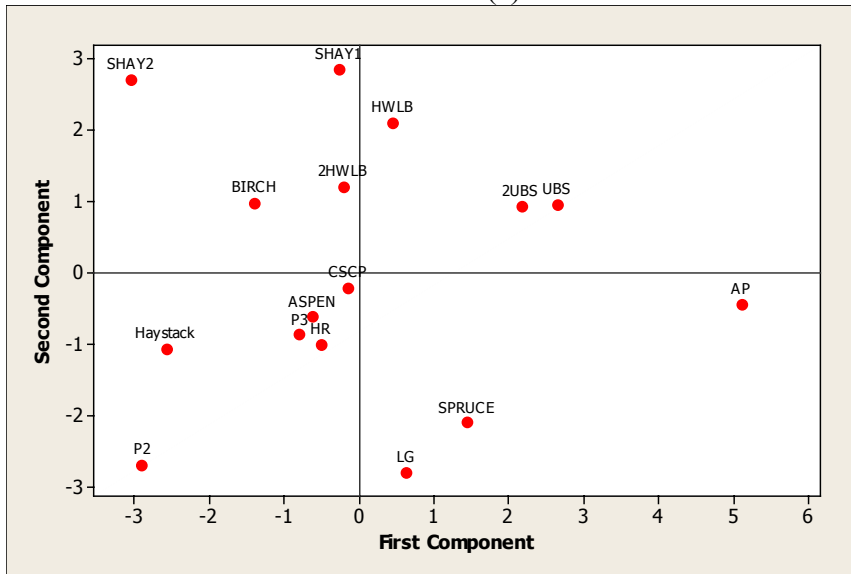

(a)

(b)

Figures 4a and 4b. Loading and score plots for all pyrolysis products and trees.

(a)

(b)

Table 1. Description of cover vegetation at sample sites.

Sample Number	Topographic Descriptor	Tree	Shrub	Moss
1 (AP)	NF	Black Spruce, Aspen, Willow	Arctic Rose	Feather Moss
2 (H2)	SF	Black Spruce	Dwarf Birch, Cranberry, Blueberry	Feather Moss, Sphagnum Moss
3 (HR)	SF	Black Spruce	Dwarf Birch, Cranberry, Blueberry	Sphagnum Moss
4 (LG)	SF	Black Spruce	Dwarf Birch, Cranberry, Blueberry	Lichen
5 (P2)	NF	Black Spruce	Dwarf Birch	Sphagnum Moss
6 (P3)	NF	Black Spruce		Feather Moss, Sphagnum Moss, Lichen
7 (CSCP)	SF		Dwarf Birch, Blueberry	Feather Moss
8 (SHAY1)	SF	Birch, White Spruce	Cranberry	Feather Moss
9 (SHAY2)	SF	Birch, White Spruce		Feather Moss
10 (UBS)	SF	Black Spruce	Dwarf Birch, Cranberry, Blueberry, Labrador Tea	Feather Moss
11 (HWLB)	SF	Aspen, Birch, White Spruce	Cranberry, Labrador Tea	Feather Moss

Table 2. Student t-tests: Moss (one-tail).

	Feather Moss to Sphagnum	Feather Moss to Lichen	Sphagnum to Lichen
Benzenes	0.294	**0.106**	**0.091**
Nitriles	**0.121**	0.323	0.162
Other	**0.123**	0.317	0.295
Methyl Cyclopentenone	**0.121**	**0.054**	**0.144**
Phenol	**0.123**	0.218	**0.074**
Furfural	0.302	0.400	0.431

Conclusions of work to date

The presence of moss had a significant influence on the soil leachates. Student t-tests indicated that from the pyrolysis products of soil leachates, the classes of benzenes, methyl cyclopentenone, and phenol explained most of influence of ground cover on leachate DOM. PCA showed that differences and similarities existed between vegetation groups and pyrolysis products. Sphagnum moss correlated with phenol, feather moss correlated with benzofuran, and lichen correlated with dimethyl benzene. Using the score plots from PCA, the soil leachates were most closely related to their moss cover. A few of the samples suggested an influence by shrub (i.e., arctic rose) or tree (birch and white spruce) samples, but the correlations were weak. Moss seemed to be the vegetation with the most influence on DOM in soil leachates.

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Investigation of Fouling in Membrane Bioreactors for Wastewater Treatment

Basic Information

Title:	Investigation of Fouling in Membrane Bioreactors for Wastewater Treatment
Project Number:	2002AK5B
Start Date:	2/28/2003
End Date:	2/28/2004
Funding Source:	104B
Congressional District:	Alaska
Research Category:	Water Quality
Focus Category:	Waste Water, None, None
Descriptors:	fouling, membrane bioreactors, wastewater
Principal Investigators:	Silke Schiewer

Publication

1. Psoch, C. and S. Schiewer. 2004. Comparison and combination of air sparging and back flushing on a membrane bioreactor. In: Proceedings of the 9th World Filtration Congress (WFC9), New Orleans, April 18-22 2004. 10pp.
2. Psoch, C. and S. Schiewer. 2003 Air sparging in membrane bioreactors to enhance permeate flux and aeration. In: Proceedings of the 5th International Membrane Science and Technology Conference (IMSTEC), Sydney, November 10-14 2003, 6pp.
3. Psoch, C. and S. Schiewer. 2003. Strategies for enhanced performance of wastewater treatment in membrane bioreactors. In: Proceedings of ASCE/EWRI World Water & Environmental Resources Congress, Philadelphia, 2003, 10pp.
4. Psoch, C. and s. Schiewer. 2004Air sparging and backflushing in membrane bioreactors. In: Abstracts of the 7th CST workshop Fouling and Critical Flux, FCF04, Lappeenranta, Finland, June 16-18 2004 (oral presentation and abstract).
5. Psoch, C. and S.Schiewer. 2004. Air sparging in membrane bioreactors (MBR) of different types. In: Abstracts of the 15th annual meeting, North American Membrane Society (NAMS), Hawaii, June 26-30 2004 (oral presentation and abstract).
6. Psoch, C. and S. Schiewer. 2004. Anti-fouling application of air sparging and backflushing for MBR and NOM retention in raw water filtration. In: Abstracts of the Water Environment Membrane Technology (WEMT 2004) conference of the International Water Association (IWA), Seoul, Korea, June 7-10 2004 (abstract).
7. Psoch, C. and S. Schiewer. 2004. Comparison and combination of air sparging and back flushing on a

membrane bioreactor. In: Abstracts of the 9th World Filtration Congress, New Orleans, April 18-22 2004 (oral presentation and abstract).

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10. Psoch, C. and S. Schiewer. 2003. Air sparging in membrane bioreactors for oxygenation in wastewater treatment. 14th annual meeting, North American Membrane Society (NAMS), Jackson Hole, 2003 p. 140, 199 (oral presentation and abstract).

Problem and research objectives

Alaska's ecosystems are sensitive to disturbances such as water pollution. Due to the cold temperatures, organic pollutants still present in discharged wastewater are degraded at a much slower pace than in warmer climates. Therefore, especially in protected areas or ecosystems already exposed to heavier pollution, the amount of pollutants discharged into surface waters has to be limited. Membrane bioreactors, with which a very high effluent quality can be achieved, can contribute to reducing aquatic pollution.

While the use of membrane bioreactors in wastewater treatment is rapidly growing worldwide, this relatively new technique faces the problem of membrane fouling, which is the main drawback of this process. Therefore it is necessary to undertake further investigations with the aim of reducing the problem of fouling in membrane bioreactors.

The goal of the proposed project is to provide knowledge that can help to reduce fouling in membrane bioreactors. The objectives were to:

1. Design and construct a testing facility for an experimental study of fouling in membrane bioreactors, using synthetic wastewater.
2. Experimentally test methods (air sparging and back flushing) to reduce fouling and investigate the effect of operating parameters (MLSS, air injection ratio).

Methodology

The chosen method for fouling reduction was air sparging. Gas sparging, or injecting of air into the feed of the membrane to generate a gas liquid two phase cross flow, which induces a higher shear stress on the membrane surface, helps to fight the build up of a cake layer, thereby maintaining a stable permeate flux over longer time-periods.

For the bioreactor, an activated sludge tank with a capacity of 80 liters was used. The synthetic wastewater and activated sludge were pumped with a submerged pump (Grundfos) to the external membrane module. The polymer membrane (PCI) has a length of 1.20 m and a pore size of 0.2 μm. The module is made of five tubes each with an inner diameter of 6 mm, yielding a membrane surface area of 0.1 m². On each side of the module, the membrane tubes were extended through an acrylic rod. The acrylic extensions with drill holes in the same diameter as the membrane tubes served for the air supply and for observation of the flow pattern in the unit. Each tube features its own connection to the air supply, with separately adjustable air volume stream for the air sparging.

Later, different polymer membranes (Microdyn-Nadir) were used, with a length of 0.75 m and a pore size of 0.2 μm. The modules were made of three tubes each with a channel diameter of 5.5 mm, yielding a membrane surface area of 0.036 m² per module. In some experiments, back flushing was applied for these modules as a second antifouling technique, addressing primarily internal fouling in the membrane pores. Several membrane modules were operated in parallel: one with no improvement (NON), one with air sparging (AS), and one with back flushing (BF), or a combination of both (AS&BF)

Principal findings and significance

The present study is one of the first to investigate a combination of air sparging and back flushing, whereby air sparging focuses on external fouling and back flushing addresses internal fouling. Moreover, very few studies report observation periods of more than 24 hours for air sparging. The project here is one of the very few long term studies, with study periods ranging from one week to several months.
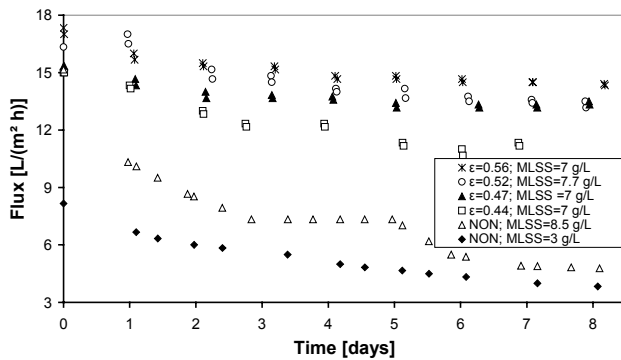

Figure 1. Comparison of Flux with NON-enhancement to combination of Air Sparging (AS) and Backflushing (BF) at different air injection ratios.
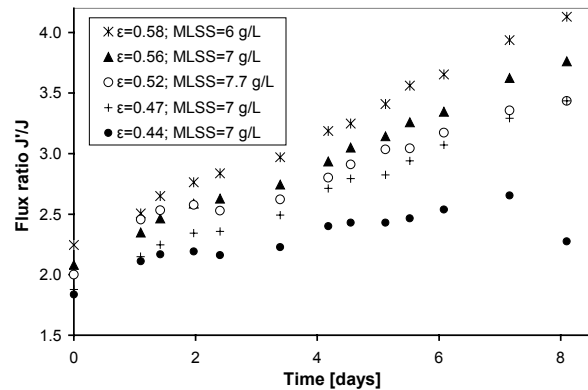
Figure 2. Flux ratios J'/J (between enhanced flux J' and NON-enhanced flux J) for a combination of AS and BF at different air injection ratios.

Figure 1 shows that within the generated slug flow regime (slug flow exists between $0.25 < \varepsilon < 0.9$ (Vera, 2000b), increasing air injection ratios achieves higher fluxes if all other parameters remain approximately constant.

Figure 2 shows the flux ratio J'/J between enhanced flux J' (at approximately constant MLSS) and NON-enhanced flux J. The enhanced flux is significantly increased (up to 4 times) compared to the conventional wastewater filtration after 8 days for $\varepsilon = 0.58$.
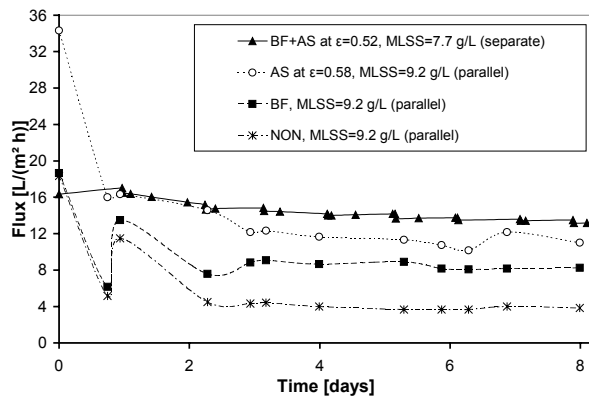

Figure 3. Comparison of NON-enhanced flux to air sparging, back flushing and a combination of back flushing and air sparging.
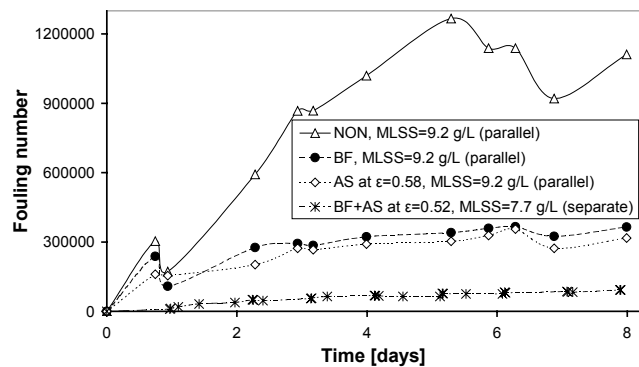
Figure 4. Increase of Fouling number vs time for NON-enhanced filtration, AS filtration, BF filtration and a combination of AS and BF.

Figure 3 shows the flux decrease within eight days, which featured parallel operation of three modules (NON & AS & BF) under exactly the same conditions. In addition the results of AS+BF are depicted. Compared to NON, the flux at Day 8 was about double for BF, triple for AS and highest for the combination of AS and BF.

With the help of dimensionless numbers, the data from Figure 3 was analyzed and interpreted in Figure 4. A gradual increase of the Fouling number can be observed. Without enhancement the Fouling number is about 3.5 times higher than the Fouling number of only AS or only BF. Nevertheless the combination of AS and BF shows again superior performance, with Fouling numbers of less than 10% compared to NON.

In conclusion, for the purpose of maintaining more sustainable fluxes, the combination of AS and BF shows very promising results in a membrane bioreactor for MLSS contents between 4 and 9.2 g/L over a time period of 8 days. The synergistic effects of AS to fight external fouling and BF to fight internal fouling are more emphasized at higher sludge concentrations. Within the slug flow regime higher air injection ratios of 0.58 show better results than lower air injection ratios of 0.44. BF can double the flux already with minimal BF pressures of 45 kPa and a product loss of only 3% due to BF.

Luminescent Bacteria: A New Water Quality Issue?

Basic Information

Title:	Luminescent Bacteria: A New Water Quality Issue?
Project Number:	2002AK7B
Start Date:	3/1/2002
End Date:	2/28/2004
Funding Source:	104B
Congressional District:	Alaska
Research Category:	Water Quality
Focus Category:	Water Quality, None, None
Descriptors:	Luminescent bacteria, salmon, freshwater, Yukon River, Alaska
Principal Investigators:	Joan F. Braddock

Publication

1. Budsberg, K.J., J. F. Braddock, and C.F. Wimpee. 2004. Analysis of the Photobacterium phosphoreum lux operon. In Abstracts of the 104th General Meeting of the American Society for Microbiology, New Orleans, LA, May 2004.
2. Budsberg, K.J., C.F. Wimpee and J.F. Braddock. 2003. Isolation and identification of Photobacterium phosphoreum from an unexpected niche: migrating salmon. Appl. Environ. Microbiol. 63: 6938-42.
3. Budsberg, K. J. 2003. Thesis: Isolation and characterization of Photobacterium phosphoreum from migrating Alaskan salmon.
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5. Budsberg, K.J., C.F. Wimpee and J.F. Braddock. 2003. Genomic cloning of the lux operon from luminous Alaskan isolates. Oral presentation at the Alaska Chapter of the American Society for Microbiology 19th Annual Meeting, Anchorage, AK, April 2003.
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Problem and Research Objectives

  The overall objective of this study was to characterize luminescent bacteria isolated from salmon harvested for subsistence use on the Yukon River. Subsistence fishers from several native villages on the Yukon River reported fish in fall 2001 on fish racks "glowing in the dark" This caused substantial alarm in the rural community as an important and traditional food source appeared to be tainted. It is not clear why this phenomenon has not been more widespread in other years, although fishers on the Yukon River have noted limited occurrences for a number of years (Randy Brown, U.S. Fish and Wildlife Service, personal communication). It is also not known how commonly luminescent bacteria are found on salmon returning to spawn from the marine environment. Furthermore, until this study, it was not known exactly what species of bacteria were responsible for the outbreak seen in 2001.

  It is important to understand that no known luminescent bacteria are pathogenic but several species are closely related to known human and fish pathogens (e.g., several strains of *Vibrio cholerae* and *V. vulnificus* are bioluminescent). In addition, the presence of luminescent bacteria on fish in the cold smoke processes has been used as an indicator of spoilage. Our preliminary data indicated that luminescent isolates from Alaskan salmon may be different from other known luminescent bacteria. Thus to assure the safety of fish on which these bacteria are growing, it was important to determine the identity of the bacteria.

  Subsistence fishers on the Yukon River have observed "glowing" fish in other years (Randy Brown, U.S. Fish and Wildlife Service, personal communication), but the phenomenon was widespread in fall 2001. It is known that luminescent bacteria "light up" only upon reaching a high population density; apparently conditions favored growth of these bacteria in summer 2001. The presence of luminescence on Yukon River salmon led to a number of interesting questions: (1) What taxonomic groups do our bacterial isolates belong to? (2) Are the Alaskan isolates taxonomically different from other previously described luminescent bacteria? (3) How are the bacteria transported to the freshwater environment? Via the fish gut or slime layer or other parts of the fish? (4) Are other species of luminescent bacteria present in fish traveling up Alaskan rivers to spawn? (5) Can planktonic luminescent bacteria be found in the Yukon River? (6) Assuming our isolates are of marine origin, do the bacteria possess physiological adaptations that allow them to survive in the freshwater habitat? (7) Were conditions unique in summer 2001 that allowed more widespread occurrence of luminescence on fish? (8) Are the isolate from 1997 and the isolates from 2001 the same organism? (9) Are isolates found in different locations on a single fish the same organism? (10) Is the occurrence of "glowing fish" likely to be high in the future due to other factors such as global change or the overall health of the fish? The first year of this project focused on answering questions (1), (2), (4), (6) and (8).

  Research during the second year of this project focused on genetic characterization of the *lux* operon of one Alaskan *P. phosphoreum* isolate; as well as one *P. phosphoreum* strain from the Black Sea; one from the Atlantic Ocean near the Canary Islands; and one from the Pacific Ocean near Coos Bay, Oregon. Research focused on the component genes in the *lux* operon from each *P. phosphoreum* strain, genes upstream and downstream of the *lux* operon in each of the strains, exact determination of the *lux* transcript start site from and an analysis of the *lux* promoter from each of the four geographically separated strains of the same species.

Approaches and Methodology:

To characterize our isolates first extensive phenotypic analyses were performed (as described in Reichelt and Baumann, 1973; Nealson, 1978). Second, characterization of the *luxA* genes were performed as previously described (Wimpee et al., 1991). Briefly, genomic DNA was isolated, *luxA* primers were used in PCR amplification, amplified fragments were cloned into pCRII-TOPO (Invitrogen Corp.), and the resultant cloned DNA was sequenced on an automated sequencer. The sequences were subjected to phylogenetic analysis using programs in the PAUP package (Swofford, 1998). Thirdly, a similar strategy was used for the 16S rRNA gene. Universal bacterial primers ("11F " and "1492R") were used to amplify 16S, followed by cloning, sequencing, and phylogenetic analysis. Finally, in addition to the sequence analysis of *luxA* and 16S, a genomic library was constructed, and the cloned *luxA* gene was used as a probe to isolate the entire *lux* operon from our Alaskan strains.

Detailed analyses of the *P. phosphoreum lux* operon were done in several successive steps. First, the lux operon from each of the strains was cloned using the method of Sambrook (Sambrook, 2001). Sequencing of each of the genomic clones was done with a combination of EZ:TN <Kan-2> transposon insertion kit (Epicentre) and primer walking. Sequencing products were submitted for sequencing on an automated sequencer. A bidirectional contig was assembled for each operon sequenced using Sequecher (Gene Codes). Transcript start sites were determined with a method similar to that of Webb (2003). Analysis of the *P. phosphoreum lux* promoter was done by eye, looking for sequences reported to be essential for the operation of the *lux* operon in two sister taxa, *Vibrio fischeri* and *Vibrio harveyi*.

Principle Findings and Significance:

All our Alaskan isolates are short rods, oxidase negative, Gram negative, and require L-methionine for growth in minimal media. Additionally, all Alaskan isolates grow at 4° C, however, optimal growth occurred at 10 - 15° C; no growth occurred at or above 20° C. Growth required the presence of sodium chloride in the medium and cells did not remain viable in river water unless amended with sodium chloride. Comparing our nutritional versatility data to published references, we can place all Alaska strains in the *P. phosphoreum* group. To verify our results, a reference strain, *P. phosphoreum* NZ-11-D, was included in the test. The ability of NZ-11-D to utilize acetate and AK-3 to utilize pyruvate do not present any difficulty in placing Alaskan strains in the *P. phosphoreum* group because of the genetic information described below.

SSU rDNA gene sequences of the seven AK isolates were aligned with six representative sequences from other luminous bacteria. The alignment produced a consensus sequence 1,159 bp in length shared by all 13 taxa. Maximum likelihood analysis of the alignment by PAUP v4.0 revealed all AK isolates cluster identically with *P. phosphoreum* (Figure 1). *E. coli* was used as the outgroup in the maximum likelihood analysis of the SSU rDNA genes.

luxA (a gene necessary for luminescence in all luminescent bacteria) sequences of the seven AK isolates were aligned with six representative sequences from other luminous bacteria. The alignment produced a consensus sequence 554 bp in length shared by all 13 taxa. Maximum likelihood analysis of the alignment by PAUP v4.0 revealed all AK isolates cluster closely with *P. phosphoreum* (Figure 2). *V. harveyi luxB* was used as the outgroup in the maximum likelihood analysis of the *luxA* genes.

Our Alaskan strains of *P. phosphoreum* are nearly identical to other descriptions with respect to nutritional versatility, *luxA* and SSU rDNA sequences; however, our isolates appear to

have a lower optimal growth temperature as compared to the reference strain, *P. phosphoreum* NZ-11-D. Future investigations of the osmotic requirements and temperature tolerances of Alaskan *P. phosphoreum* may reveal adaptations specific to this unique niche.

We observed that the *lux* operon in all *P. phosphoreum* strains examined was *luxCDABFEG* (Figure 3). Also, in all strains examined, the gene immediately upstream of *luxC* was *lumP*, and *ribA* was determined to be downstream of *luxE* in all *P. phosphoreum* strains examined (Figure 4). Interestingly, the *rib* genes code for an enzyme system that synthesizes riboflavin – a necessary substrate for the gene products of the *P. phosphoreum lux* operon. Regardless of which *lux* gene we analyzed, a similar phylogenetic pattern emerged in which the closest sister taxon to the *P. phosphoreum* group is *Photobacterium leiognathi* (Figure 5). Additionally, in phylogenetic inferences that included *S. hanedai*, the phylograms suggest that the *lux* operons fall into two clades: one containing the *Photobacterium* species, *V. fischeri*, and *S. hanedai*; the other containing *V. harveyi*, *P. luminescens*, and *V. cholerae*. Consistent with previous studies, our deduced LuxF amino acid sequences show similarity with *V. fischeri* and *V. harveyi* LuxB amino acid sequences (data not shown). We also observed that the transcript start site for all of the strains examined differ by a single nucleotide position (Figure 6-7). This is a surprising result because the *P. phosphoreum* strains we examined were isolated from widely separated geographic locations. Finally, we were unable to recognize any of the promoter sequences reported to be necessary for normal functioning of the *lux* system in two closely related Bacterial species, *V. fischeri* and *V. harveyi*.

Overall, we have definitively identified the bioluminescent bacteria isolated from Yukon River salmon. We have also determined that the bacteria appear to require salt or some other osmo-regulant for growth and that they have a somewhat lower optimal growth temperature than other known isolates. More detailed analysis of the *lux* operon from several Alaskan isolates revealed surprising similarity to organisms isolated from geographically distant locations. Finally, our results suggest that *P. phosphoreum* may have a regulatory system that is different from other known bioluminescent bacteria. The project supported one master's student and resulted in a number of published abstracts, one published manuscript and a second manuscript that will be submitted for publication this summer.

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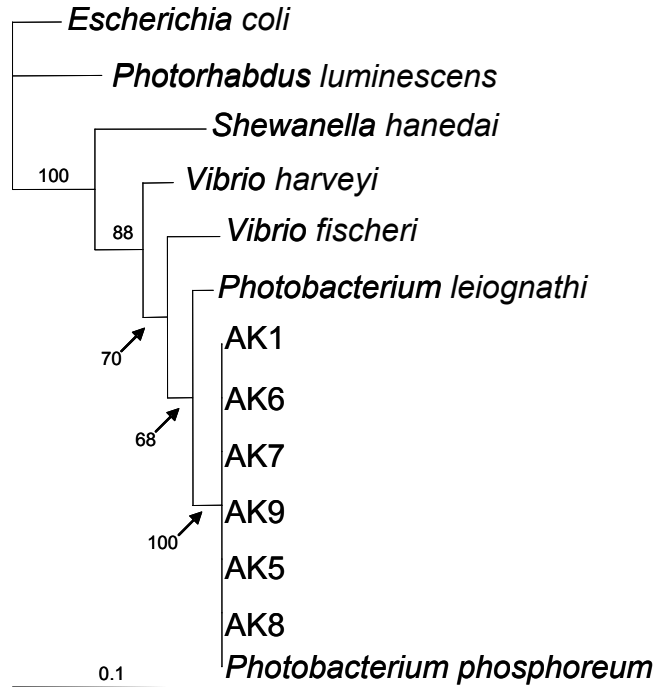
Figure 1. Phylogeny of Alaskan luminous bacteria based on Maximum Likelihood analysis using PAUP* 4.0b10 with SSU rDNA sequences from Alaskan isolates and representative sequences from GenBank. All strains with "AK" are from salmon harvested from the Yukon River, Alaska. *E. coli* was used as the outgroup in this analysis.
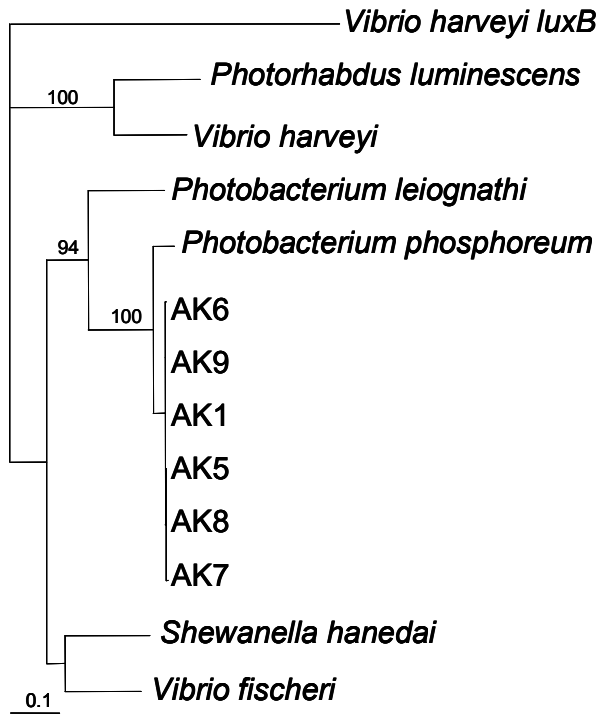
Figure 2. Phylogeny of Alaskan luminous bacteria based on Maximum Liklihood analysis using PAUP* 4.0b10 with *luxA* sequences. All strains with "AK" are from salmon harvested from the Yukon River, Alaska. *V. harveyi luxB* was used as the outgroup in the Maximum Likelihood analysis of *luxA* genes.

luxA phylogeny	species	*lux* gene organization

Photobacterium phosphoreum (*) — lumP — luxC | luxD | luxA | luxB | luxF — luxE | luxG

Photobacterium leiognathi (6) — luxC | luxD | luxA | luxB | luxF — luxE | luxG

Vibrio fischeri (9) — luxR — luxI | luxC | luxD | luxA | luxB — luxE | luxG

Shewanella hanedai (*) — luxC | luxD | luxA | luxB — luxE | luxG

Vibrio harveyi (5) — luxC | luxD | luxA | luxB — luxE | luxG | luxH

Photorhabdus luminescens (23) — luxC | luxD | luxA | luxB — luxE | luxG

Vibrio cholerae (35) — luxC | luxD | luxA | luxB — luxE | luxG

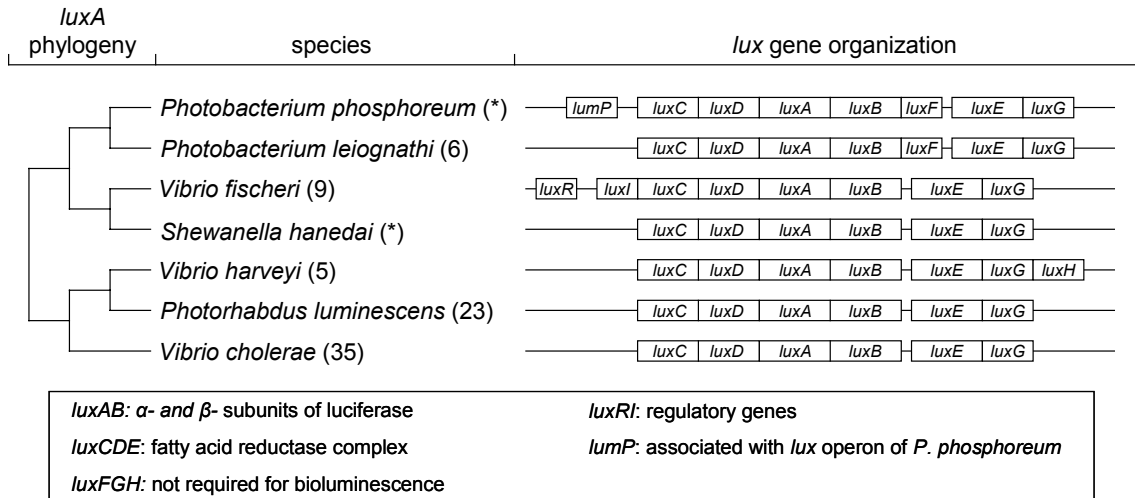|	
luxAB: α- and β- subunits of luciferase	*luxRI*: regulatory genes
luxCDE: fatty acid reductase complex	*lumP*: associated with *lux* operon of *P. phosphoreum*
luxFGH: not required for bioluminescence	

Figure 3. *lux* operon organization of bioluminescent bacteria from which the *lux* operon has been sequenced. Organization of *lux* genes required for bioluminescence, *luxCDABE*, is conserved. Some *Photobacterium* species possess *luxF*, between *luxB* and *luxE*. Numbers in parentheses indicate reference from which *lux* gene organization was reported, asterisks indicate *lux* gene organization determined from DNA sequences deposited in GenBank. Modified from Meighen, 1994.
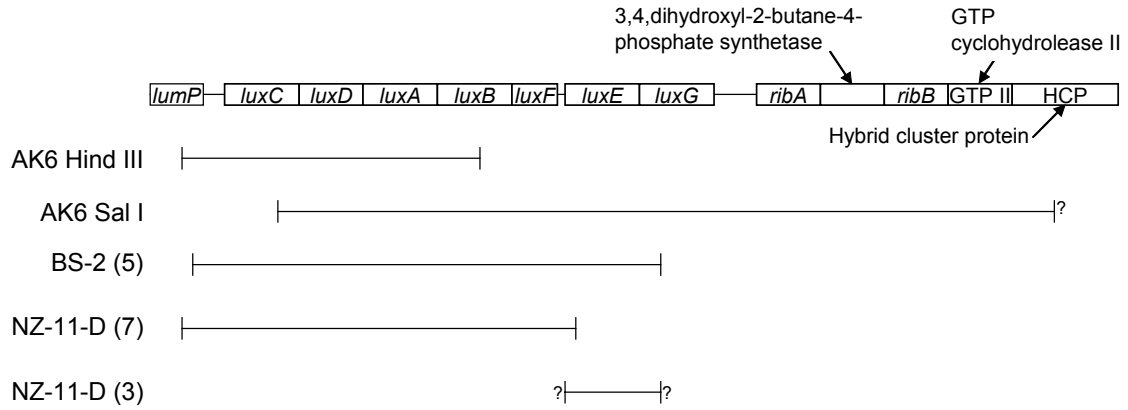
Figure 4. Locations of inserts of genomic clones used in this study.
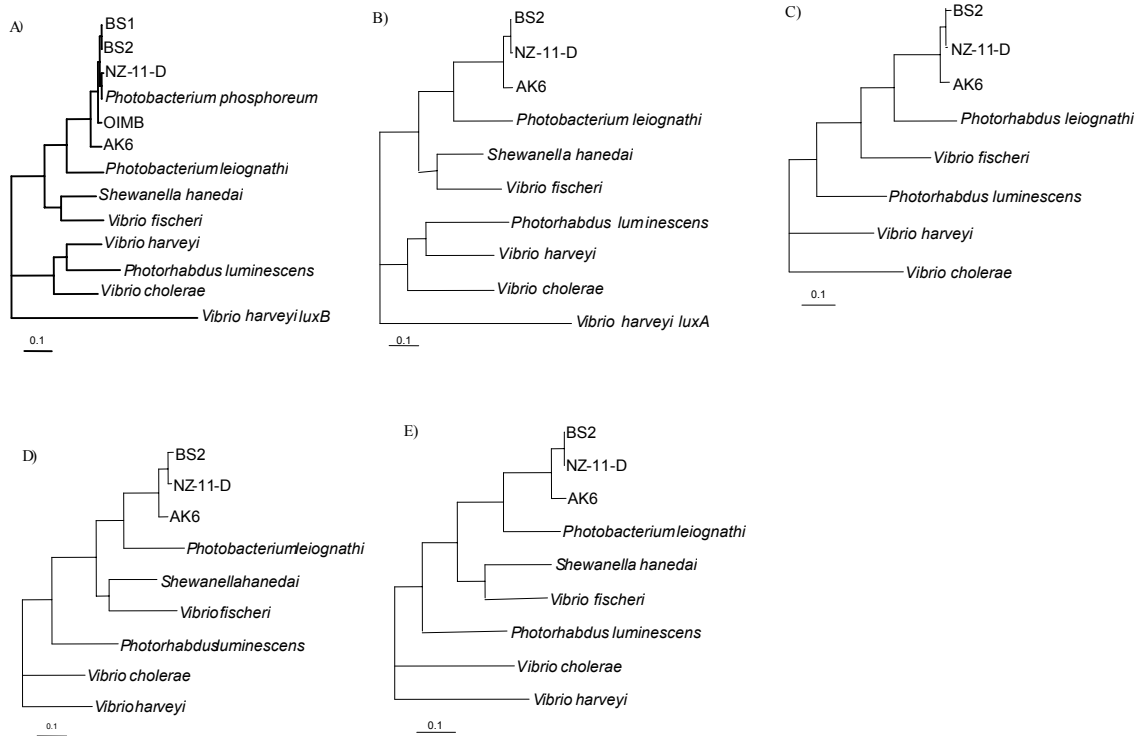
Figure 5. Phylogenetic analysis of the individual *lux* genes. A) ML tree based on *luxA* gene sequence, rooted in *V. harveyi luxB*; B) ML tree based on *luxB* gene sequence; rooted in *V. harveyi luxA*; C) ML tree based on *luxC* gene sequence, rooted in *V. harveyi luxC*; D) ML tree based on *luxD* gene sequence, rooted in *V. harveyi luxD*; E) ML tree based on *luxE* gene sequence, rooted in *V. harveyi luxE*.
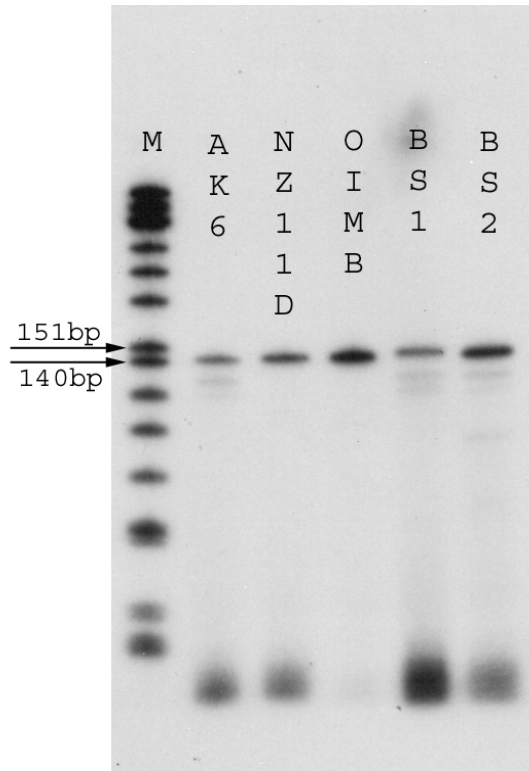
Figure 6. Primer extension products for AK6, NZ11D, OIMB, BS1, and BS2. Marker in first lane is φX174 DNA/*Hinf* I. Volumes loaded were 5 µl 1:50 dilution of φX174 DNA/*Hinf* I marker, 5 µl AK6, 5 µl NZ11D, 5 µl 1:10 dilution OIMB, 15 µl BS1, and 5 µl BS2.
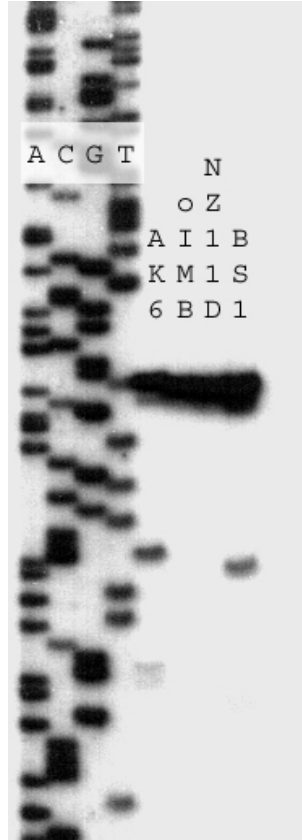
Figure 7. Autoradiogram of primer extension products for the *lux* operon of *Photobacterium phosphoreum* strains AK6, OIMB, NZ-11D, and BS1.   Lanes 1-4 are a sequencing ladder generated for AK6.

Information Transfer Program

Information dissemination is an important activity in any research entity. We accomplish our information dissemination using several mediums: through the organization of seminars, workshops and conferences; by publishing in a variety of outlets; maintaining an active website; participation in state, federal and international committees and professional organizations; professional interaction with K-12; and technical presentations. Every week during the spring and fall semester, we sponsor seminars. Although primarily geared for graduate students, we do have several outside speakers from other universities and countries.

We sponsor workshops, symposia and conferences from the state local to international scale. For example, in 2003 we had a workshop on the Environmental Impacts of Winter Pumping on Lakes on the North Slope of Alaska (attended by 40 people from industry, state, federal, environmentalist and university either in Anchorage or Fairbanks, audio-visual workshop). In February 2004 we convened a meeting in Victoria, BC on Synthesis of Water Balance of Northern Research Basins with 40 people from eight countries. Twenty-four papers from this workshop will be published in a International Association of Hydrological Sciences (IAHS) redbook.

Electronically, we publish a newsletter about every two weeks that also includes pertinent information about WERC (see http://www.uaf.edu/ine/researchflash.htm) . WERC maintains a website (http://www.uaf.edu/water/) that contains information about what is new, ongoing research, publications, student opportunities, general information and more. We have access to data collected at over 40 remote sites around Alaska (North Slope, Seward Peninsula, Interior, etc.); this data is used extensively by state, federal and private sectors. We also maintain some video cams at various research sites like on the North Slope at a Department of Transportation and Public Facilities maintenance camp: htp://www.uaf.edu/water/projects/NorthSlope/images/SagRiverDOT_cam.jpg/

Following is an email received from Edward Plumb (hydrologist with National Weather Service) on March 29, 2004: We are very active users of your real-time data in our operationally driven field. We often rely on your data to fill in the enormous gaps between the NWS and FAA sites. If I have not told you this in the past, the forecast office really appreciates all the data you guys provide over the internet. Thanks!

We encourage faculty, staff and graduate students to participate in K-12 science related activities. These activities range from giving talks and demonstrations in the students classroom, participating as an advisor or judge at science fairs, taking students on field trips or a tour through our research facilities, touring facilities such as the permafrost tunnel near Fairbanks, or participating in longer science camps such as: http://www.uaf.edu/water/projects/ftww/notes2/diagnostics/Outdoor.html

A recent example of advising a high school student follows: The Alaska Stable Isotope Facility (ASIF) in the Water and Environmental Research Center (WERC) recently supported two High School student science projects. One was by Robert Marcotte, a student at West Valley High School who was advised by ASIF staff on a project titled: Determining the stable carbon isotopic composition of plants from the last glaciation in the interior of Alaska - a preliminary survey of the permafrost tunnel, Fairbanks, Alaska. Robert placed 5th overall in the Alaska State High School Science Symposium with this project and moved on to the national symposium in Washington, D.C. The context of the project was related to determining how Alaskan ecosystems respond to environmental change.

Although none of these activities are supported by NIWR funds, NIWR-supported research is disseminated to the research community and the general public through these avenues.

Student Support

Category	Section 104 Base Grant	Section 104 RCGP Award	NIWR-USGS Internship	Supplemental Awards	Total
Undergraduate	0	0	0	0	0
Masters	4	0	0	0	4
Ph.D.	1	0	0	0	1
Post-Doc.	0	0	0	0	0
Total	5	0	0	0	5

Table caption (top of table): Student Support

Notable Awards and Achievements

Publications from Prior Projects

None