

RECLAMATION OF FORESTED SOILS

DAMAGED BY OIL SPILLS

**Results of Preliminary Studies
Under Contract No. OSP3-0073
For the Northern Forest Research
Centre, Edmonton, Alberta**

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RECLAMATION OF FORESTED SOILS DAMAGED BY OIL SPILLS — PRELIMINARY STUDIES *

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SUMMARY

Most oil spills in Alberta take place in forests, and the oil usually accumulates in poorly-drained areas with organic soils. Spills are conventionally treated by diking and pumping off free oil, followed by burning as much as possible of the residual oil. Spill areas generally become re-vegetated only very slowly after that treatment, especially where the oil has penetrated the soil to some depth. Preliminary results from a study on reclamation of such soils shows that decomposition of oil contained in the soil and the re-establishment of vegetation can be accomplished quickly and economically. In field experiments it was possible to establish plants on a previously barren spill, and to decompose most of the oil during a four-month period. Oil decomposition by soil microorganisms is retarded by lack of nutrients and, in some instances, by soil acidity. Poor plant germination and growth in waterlogged soils is an additional problem. Oil decomposition occurs rapidly if nutrients are added to promote bacterial activity. Lack of nutrients for microbial decomposition of oil is a greater problem than lack of organisms since most soils normally contain organisms capable of oil decomposition. When needed, bacteria can be added easily by adding a small amount of soil, containing bacteria capable of oil degradation. Soil from partially reclaimed oil spill areas is suitable for this purpose. Revegetation can be achieved (possibly in one year) if drainage of wet low lying areas accompanies the addition of nutrients. The oil itself appears to be more harmful to germination than to growth of an established plant if nutrients are present. Toxicity per se does not appear to be as serious a problem as is nutrient deficiency at the sites we

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have studied. It is expected that in another year we will have practical, economical techniques established for promoting biological degradation of the oil left after burning and for re-establishing vegetation on these barren areas. These techniques will be designed to be compatible with the total ecosystem so that in solving one pollution problem we do not create another.

INTRODUCTION

Oil pollution of soil is a serious problem from environmental, productivity and ecological standpoints. Productivity considerations are generally associated with agricultural soils. However, the ability of wet forested soils to provide wildlife habitat and their use for recreational purposes is also important. We can not separate soil pollution from productivity, and we can not ignore productivity when dealing with wet forested soils.

Oil spills occurring in forests generally become localized in low lying areas. Spills may occur because of an unattended valve or through leaks in storage pits, in which case the escaping oil runs downhill to low areas. Pipeline breaks are another source of spills, and occur most frequently in poorly drained soils due to anaerobic corrosion. Soil slumping also causes breaks. In all cases the oil collects in low wet areas.

This report will deal with preliminary results from attempts to reclaim two wet, oil-contaminated soils; one west of the town of Swan Hills, and one in the vicinity of Leedale in Central Alberta.

SITES

SWAN HILLS This site has received accidental oil spills over a period of six years. Each spill was burned as part of the original clean-up operation. Drainage is almost non-existent and the peat (3 - 5 feet thick) is floating in water. Free water frequently sits on top of the peat-oil mixture. Prior

to the spills the site was a Black Spruce bog, but all trees were burned in the clean-up operations. Depth of oil penetration varied between 12 inches in dry areas to 1 inch in wet areas with 1-2 inches being the depth in most instances. Oil content varies between 200 and 300% by weight in the top 2 inches, but is only 35% in the 2-6 inch depth.

LEEDALE A pipeline leak spilled oil at this site during the fall and winter of 1971/72. The site is a flat, poorly drained area of willows, sedges and grasses. Severe disturbance of the area during pipeline repair and clean-up resulted in oil being mixed to a depth of 12-24 inches. The area was diked and this accentuated the problems of poor drainage. Oil content is 150% by weight to a depth of 6 inches.

RECLAMATION PROCEDURES

The reclamation procedures were designed to provide information on the effect of drainage and addition of nutrients, lime and a soil inoculum on oil degradation rate, numbers and types of microorganisms, and on plant growth.

A. Box and Frame Experiment

Drainage was provided by placing undisturbed samples of oiled peat into 4' X 8' X 12" boxes raised so that the water table was lowered by 4-5 inches. Frames were placed beside the boxes to serve as control (undrained) areas.

Nutrients were added according to the following table:

<u>Nutrient</u>	<u>Rate (kg/ha)</u>	<u>Source</u>	<u>Times of Application</u>
N	336	NH ₄ NO ₃	June 22, July 22 (213 kg/ha) and Sept. 9
P	112	11-55-0	June 22, September 9
K	112	K ₂ SO ₄	June 22, September 9
S	46	K ₂ SO ₄	June 22, September 9

Amendments included lime (CaCO_3) at 12 tons per acre, and an inoculum of 4 tons per acre of a Black soil (Angus Ridge series) previously contaminated by a similar oil and containing a large population of organisms adapted to oil degradation. Also included in the amendments treatment were micronutrients.

Mixing was used to dilute the oiled surface soil with unoiled soil from below, and to break the continuous oil layer at the surface. The soil was intimately mixed by hand to 10 inches.

B. Piling Up Experiment

Drainage was achieved by piling mixed oiled soil into heaps one foot above the present surface. The piles were contained in 4' X 8' frames with control frames set adjacent to them.

Nutrient and amendment treatments were the same as in "A" except that no additions of nutrients occurred until July 5, 1973.

C. Dike Experiment

A 200' X 200' area was diked off and a ditch was dug within the border of the dike. Water was then pumped from the ditch to lower the water table by 6-10 inches. Nutrient and amendment treatments were as in "A". Mixing was to a depth of 6 inches. This experiment was replicated three times. A control area was located immediately north of the dike.

D. Strip Experiment - Leedale

This experiment did not contain a separate drainage treatment because the entire Leedale site was pumped out and drained. Three 5' X 80' strips were laid out with a 10' buffer between each strip. Treatments were: Nil; Nutrients; Nutrients plus amendments.

The nutrients were added according to the following table:

<u>Nutrient</u>	<u>Rate (kg/ha)</u>	<u>Source</u>	<u>Times of Application</u>
N	326	NH ₄ NO ₃	September 15
	112	(NH ₄) ₂ SO ₄	October 6
P	112	11-55-0	September 15
K	112	K ₂ SO ₄	September 15
S	46	K ₂ SO ₄	September 15
	128	(NH ₄) ₂ SO ₄	October 6

Amendments were the same as in the Swan Hills experiment (A, B and C). This experiment was not started until late August because of a delay in pumping water from the area.

RESULTS

OIL DECOMPOSITION

The rate of oil degradation during the summer of 1973 was influenced by the reclamation treatments used and was most rapid and complete where the soil was mixed, drained and had nutrients and amendments added. During a four-month period some of the treatments had reduced oil content to only a small fraction of the original value (Tables 1 and 2). Drainage had only minor effects on decomposition rate in the Box and Frame experiment, but caused a substantially greater decrease in oil content in the Piling Up experiment (Table 1 and 2). Mixing was beneficial in all experiments and the mixing action during piling the material into heaps for the Piling Up experiment may help account for the greater effect of drainage in that experiment. Downward movement of oil in the drained soil (Piled Up portions) cannot be discounted. Substantial amounts of emulsified oil or its decomposition products appear to be present in some of these treatments. This would allow movement of hydrocarbons into water below the sampling depth. Addition of lime, micro-nutrients and an active soil inoculum appeared to have the greatest single

effect although it is unlikely that this would have occurred in the absence of added nutrients.

Results from the replicated Dike experiment (Table 3) support those of the Piling Up and Box and Frame experiments. In addition, they demonstrate that rapid oil decomposition can be achieved even under cold conditions prevailing in the late fall. The experiment was conducted from August 25 to October 20 and during that time more than half the oil was decomposed on some treatments. Thus reclamation of these cold wet sites is probably not restricted to as short a time as the frost-free period might indicate.

Two factors probably contribute to the beneficial effect of mixing. Oil scums form on the surface of the soil. These scums become hard and dry with the result that very little microbial activity can take place. Mixing breaks them and buries much of the scum below the surface. This provides an effective oil-water interface at which microbial activity may take place. Mixing also dilutes the oiled soil with unoled soil thus reducing the concentration of the oil.

Although mixing during reclamation is advantageous, disturbance of the soil surface during the initial clean-up operation considerably increases the overall problem of the spill. Disruption of the soil surface allows oil to be mixed into soil at depth. Decomposition at depth is slow because of poor aeration and lack of nutrients. Although we do not know how much of this oil or its decomposition products may move into groundwater, the longer persistence increases this possibility.

SOIL MICROORGANISMS AND NUTRIENT CYCLING

The activity and types of soil microorganisms as indicated by total numbers of bacteria and fungi was influenced by the presence of oil and by the reclamation techniques used. In both the Box and Frame and the Piling Up experiments the initial microbial population was primarily fungi. Drainage was favourable to the growth of both fungi and bacteria (Table 4 and 5). Addition of nutrients

alone generally resulted in increased numbers of fungi whereas nutrients plus amendments increased numbers of bacteria at the expense of fungi (Table 4 and 5). At the Leedale site oil increased the number of bacteria and fungi by several fold over the adjacent unoiled control soil (Table 6).

Oil appears to be decomposed by both bacteria and fungi although bacteria cause more rapid breakdown. In both the Piling Up as well as the Box and Frame experiments, addition of nutrients increased the numbers of fungi but not the numbers of bacteria. This increase in fungi was associated with a reduction in the amount of oil in the soil. Addition of nutrients and amendments plus mixing of this soil caused a 10-fold increase in bacteria but a decrease in the numbers of fungi. This shift in the population may be attributed to the increased pH as a result of liming and to the bacteria added with the soil inoculum. This treatment was also associated with the most rapid loss of oil. Hence, there may be some advantage in maintaining soil conditions which enhance the ability of bacteria to compete with fungi for added nutrients.

Proper reclamation should produce an improvement in general fertility and biological activity of soil, compared to that of the soil prior to the oil spill. The increased numbers of microorganisms in the presence of oil and of oil plus nutrients is a result of decomposition of the oil by bacteria. A part of the C present in the oil is converted into microbial biomass; the remainder is oxidized to CO_2 to provide energy. Most of the added N, P, etc. is used for biomass synthesis. Hence decomposition of oil in soil will result in an increase in the amount of microbial tissue in soil and ultimately the amount of soil humus.

Nutrients present in organic form as microbial biomass and humus are remineralized over an extended period of time. Thus fertilizer added for oil spill reclamation can be used by succeeding crops on agricultural soils and by re-established vegetation in forested areas. Because of the re-mineralization of nutrients, it is essential that plant growth be re-established in wet areas as

soon as possible, to promote continual recycling of nutrients within that ecosystem and prevent eutrophication of lakes and streams in adjacent areas.

A simple calculation may serve as an example of the long-term increase in fertility that one may expect.

If 1,500 lb N/acre were used for reclamation, it would be remineralized over a period of several years.

Assuming a half-life of 5 years (from studies on soil N) the following quantities of N would be available for succeeding crops:

	Years After Successful Reclamation							
	<u>1</u>	<u>2</u>	<u>3</u>	<u>4</u>	<u>5</u>	<u>6</u>	<u>7</u>	<u>8</u>
lb N/acre released /year	210	165	150	135	120	105	90	75

The value of released nitrogen can be appreciated if one considers that a 40 bu/acre wheat crop will contain about 100 lb N. Phosphorus and sulfur undergo similar biological transformations from organic to inorganic form in soil.

The amount of oil that can be expected to be degraded after addition of a given quantity of N can also be calculated:

Assumptions:

- 1) C/N ratio of microorganisms = 8
- 2) Efficiency = 40%

i.e. for every unit of C attacked by microorganisms 0.4 units are converted into biomass and 0.6 units are respired as CO_2 -C.

- 3) Turnover rate of microbial population = 3 times/year.

Consider:

500 lb N/acre added to oil-contaminated soil.

Biomass-N produced = 500 lb/acre

Biomass-C = $500 \times 8 = 4,000$ lb/acre

Total C attacked = $\frac{4,000}{0.4} = 10,000$ lb/acre

population cycles 3 times:

therefore $10,000 \times 3 = 30,000$ lb oil-C attacked per year

per 500 lb N added.

This is equivalent to $\sim 2\%$ oil by weight in the top 6" of a mineral soil.

In view of the foregoing, the observation that in one short season the oil content was reduced from 120% to 20% in the Swan Hills Box and Frame experiment is quite reasonable.

For example: Box and Frame Experiment

Basic data:

Bulk density	0.02 g dry oil-free peat/cm ³
N added	672 kg N/ha
Oil content:	
originally	120%
after treatment	20%

Calculations:

Weight of oil: (0-25 cm)	
originally	6.0×10^4 kg/ha
after treatment	1.0×10^4 kg/ha
Loss of oil	5.0×10^4 kg/ha
Loss of soil-C	3.75×10^4 kg C/ha

(assuming 75% C in the oil)

$$\text{C loss per 100 kg N} = \frac{3.75 \times 10^4}{6.72} = 0.56 \times 10^4 \text{ kg C}$$

From previous theoretical calculation:

$$\text{C loss per 100 kg N} = \frac{3.36 \times 10^4}{5.6} = 0.60 \times 10^4 \text{ kg C}$$

The theoretical value is only 7% greater than the value calculated on the basis of observed field results. One should not take this to mean that a simple calculation is now possible for all reclamation problems. The above is being checked further in the field with both forested soils and agricultural soils to evaluate it properly and this will take additional time. However, this type of approach should be possible in the near future. If we are successful, it will

provide management personnel with a simple, effective means of assessing the amount of material and the time required for reclamation of specific spills.

SOIL pH AND CONDUCTIVITY

Soil pH was at 4.5 - 4.7 at the Swan Hills site and was increased to 6.6 - 7.2 by addition of lime (Table 7). The initial low pH is one of the prime reasons for the preponderance of fungi at this site since bacteria can not compete as well as fungi at low soil pH. The native soil pH of the Leedale site was near neutral (6.1) and was raised to 7.5 by lime. Soil conductivity at both the Swan Hills and Leedale sites was unaffected by addition of nutrients. Hence no adverse salt effects of the added fertilizer are to be expected. This may be attributed to the rapid assimilation of added nutrients by soil microorganisms, to the natural high water content and to the fact that the nutrients were added in increments rather than as one large dose.

MINERAL-N LEVELS

The form of mineral-N added appears to affect its rate of disappearance, with $\text{NH}_4\text{-N}$ persisting longer than $\text{NO}_3\text{-N}$. In experiments at both the Swan Hills and Leedale sites, $\text{NH}_4\text{-N}$ levels were higher than those of $\text{NO}_3\text{-N}$ (Tables 8 and 9). Four factors help account for this:

- (a) the high mobility of NO_3 in soil water may allow it to diffuse out of the experimental area,
- (b) the conditions of this study favor denitrification and some NO_3 may have disappeared in that way,
- (c) NO_3 may in fact be utilized more rapidly by bacteria and may be preferred over NH_4 ,
- (d) remineralization of N into the NH_4 form from microbial biomass with little or no nitrification would keep the NH_4 levels higher than NO_3 .

Studies with an agricultural soil also indicate that $\text{NO}_3\text{-N}$ results in more

rapid oil decomposition than urea-N. The reasons for this apparent rapid utilization of NO_3 must be further examined.

The rate at which mineral-N is immobilized depends on the treatment used with the N. In the treatments in which extra amendments were added N immobilization and oil decomposition was most rapid and the numbers of bacteria were increased markedly whereas fungi decreased. This further emphasizes the role of bacteria in oil decomposition as opposed to fungi and the importance of maintaining soil pH near neutrality to favor bacteria.

The mineral N values were generally higher in the drained than undrained areas of both the Box and Frame and the Piling Up experiment. This is partly attributable to the greater amount of water in the undrained samples resulting in a lower N concentration because of a smaller amount of soil in the analysis. However, this does not account for all of the differences because the difference in water content between drained and undrained areas will change the actual values by only 20 - 50% relative to each other.

For example:

	<u>% water</u>	<u>Kg N/ha for each 100 ug N/g wet soil</u>
Box and Frame Exp.		
drained	700	40
undrained	1,100	60
Piling Up		
drained	650	76
undrained	840	94

A more reasonable explanation is simply that with the high water content and completely saturated conditions of the undrained treatment, the added N was able to diffuse out of the experimental area. This is substantiated by the presence of abundant algal growths immediately adjacent to some of the plots receiving nutrients.

PLANT GROWTH

Germination and growth of the test crops did not occur unless the soil was drained and germination was enhanced by those treatments which resulted in most rapid loss of oil. There was no growth or very little growth where the plots did not receive drainage (Tables 10, 11). In the drained portion of the Box and Frame experiment, yield of oats was 200 times greater where nutrients plus amendments were added and the soil was mixed, than where nothing was added or done other than to drain the plot. No seeds germinated unless they were placed in contact with the peat; those lying on the oil or in water did not germinate. This is why yield (Table 10) was so markedly improved by mixing (mixing + nutrients in the Box and Frame experiment increased yield by 47-fold over nutrient additions alone). The beneficial effects of mixing and drainage on germination are further demonstrated by data in Table 11. It would appear that if any attempt at revegetation is considered, proper drainage is absolutely necessary. Drainage is not as important to oil decomposition as it is to plant growth.

Plates 1 - 6 depict the study area and help illustrate the effect of various reclamation techniques on growth of seeded plants.

Tolerance to conditions at the Swan Hills site varies with plant species. Of the seeded species oats is the most tolerant and reed canary grass appears least tolerant (Table 11). Timothy germination averaged 34% over all drained treatments whereas germination was 70% for oats and only 3.1% for reed canary grass. Various native species were observed growing in spots containing a high oil content. Sites with sedge growth (Carex aquitilis Wahlenb.) contained an average of 72% oil in the top 3 inches (dry wt. basis) whereas in adjacent areas where sedge growth did not occur the oil content averaged 83% in the same depth of soil. Sedge penetration into oil spill areas is facilitated by rhizome extension below layers where oil is highly concentrated. A shoot can then develop

up through small cracks in the oil scum on top. Buck-bean (Menyanthes trifoliata L.) was found growing in an area containing 51% oil at the surface and 10% at 6".

Another sedge, Eriophorum chamissonis C.A. May, was able to tolerate even more oil and was abundant in areas with up to 280 - 290% oil at the surface one inch (dry wt. basis) and 11% oil at 6 inches. Efforts should be made to collect some of these wild species and to propagate them for possible use in revegetating many of these barren areas.

PRACTICAL IMPLICATIONS

Indications are that the main problems with oil spill reclamation in these areas are lack of nutrients for oil decomposition by soil microorganisms, low soil pH in some instances, lack of viable seeds or plant stocks for revegetation and a poor environment for germination and seedling growth.

Considerable added damage results from initial clean-up operations when the soil is disturbed thus allowing oil to penetrate into lower soil horizons. Decomposition rates are slower at depth due to lack of nutrients, poor aeration and a low microbial population. While many oil spills in wet forested areas remain barren for extended periods of time if not treated, oil decomposition proceeds rapidly even in cold wet situations when sufficient nutrients (N, P, K, S) are present and an active microbial population (preferably bacterial) is developed. The addition of N is of prime importance in this regard. The effectiveness of added N is increased by insuring (through use of lime and a soil inoculum if necessary) that an active bacterial population is present. We do not yet know which form of N is most effective nor precisely which other amendments are necessary. It is probable that the usefulness of extra amendments will depend on the character of each individual spill site.

Mixing is advantageous to revegetation and enhances the rate of oil decomposition. This is partially a result of the dilution occurring upon mixing. It also places oiled soil in contact with soil organisms in an environment more conducive to microbial growth than the strictly aerial situation existing prior to mixing when a hard dry crust is present. If the soil is uniformly oiled to considerable depth there may be no advantage to mixing.

General soil fertility and soil biological activity can be expected to increase beyond its original level after proper reclamation. Nutrients added for reclamation will be recycled back through the soil and into

newly established vegetation. This will continue over an extended period of time.

Revegetation of low lying oil spill areas requires drainage. This may entail breaking a dike, allowing natural drainage to occur or some other technique such as ridging within the bog to produce elevated dry areas on which to establish plant growth. Revegetation is also essential to proper reclamation so as to prevent subsequent eutrophication of adjacent waters. Our results suggest that soils such as those investigated can be successfully revegetated even though they contain considerable quantities of oil. The major effect of oil appears to be on nutrient availability. We have not observed toxicity per se to be as much of a problem as nutrient deficiency. We must examine a wider range of oils and spill situations to substantiate this observation.

Nitrate-N added during reclamation does not cause a pollution problem because it is rapidly immobilized during oil decomposition. Its subsequent release from organic form occurs over an extended period. It is released as NH_4 and rapid nitrification can not be expected in these wet partially anaerobic areas.

Dispersants or surfactant are probably not necessary for reclamation. We are presently examining the possibility that they may enhance oil breakdown in soil but it would appear that biological breakdown is limited more by nutrient deficiency than by availability of the oil. Biodegradation results in production of natural surfactants; this may eliminate the need for commercial dispersants on soil.

We expect that in another year we will have developed practical, inexpensive reclamation techniques for wet forested areas. Efforts will be made to insure that methods developed are compatible with the total ecosystem so that in solving one problem we do not create another.

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Table 1. Oil contents of soil at Swan Hills after imposition of various treatments - Box and Frame experiment.

<u>Sampling Dates</u>	<u>% Oil in different treatments*</u>			
	<u>Nil</u>	<u>NPKS</u>	<u>NPKS, plus mixing</u>	<u>NPKS, mixing, and amendments**</u>
Drained				
June 22, 1973	124	95	112	151
July 11, 1973	160	135	109	38
August 9, 1973	118	98	37	41
October 20, 1973	107	107	16	21
Undrained				
June 22, 1973	138	131	102	158
July 11, 1973	191	124	115	81
August 9, 1973	103	53	86	53
October 20, 1973	99	55	54	46

* Oil content was determined by Soxhlet extraction from field moist soil using CH_2Cl_2 . The CH_2Cl_2 was then evaporated off and the residue weighed. The soil after extraction was dried and the oil content calculated as a % of the dry soil weight. Prior to extraction, all samples were ground in a Waring blender to allow a uniform sub-sample to be removed. Control, unoiled samples when similarly extracted yielded negligible quantities of material. All oil analyses were done in duplicate.

** Amendments = Lime (12 tons/acre) and oiled soil inoculum (4 tons/acre) plus micronutrients.

Table 4. Numbers of Bacteria and Fungi in soil from the Box and Frame experiment at Swan Hills, August 9, 1973.

<u>Treatment</u>	<u>Numbers per gram field moist soil**</u>			
	<u>Drained</u>		<u>Not Drained</u>	
	<u>Bacteria</u> <u>x 10⁶</u>	<u>Fungi</u> <u>x 10⁴</u>	<u>Bacteria</u> <u>x 10⁶</u>	<u>Fungi</u> <u>x 10⁴</u>
Nil	71	100	2	40
NPKS	33	200	4	34
NPKS + Mixing	47	200	2	3
NPKS + Mixing + Amendments*	200	17	44	3

*Amendments = Lime (12 tons/acre) + oiled soil inoculum (4 tons/acre) + micro-nutrients.

**For methods see bottom of Table 6.

Table 5. Numbers of Bacteria and Fungi in soil from the Piling Up experiment at Swan Hills, August 9, 1973.

<u>Treatment</u>	<u>Numbers per gram field moist soil**</u>			
	<u>Drained</u>		<u>Undrained</u>	
	<u>Bacteria</u> <u>x 10⁶</u>	<u>Fungi</u> <u>x 10⁴</u>	<u>Bacteria</u> <u>x 10⁶</u>	<u>Fungi</u> <u>x 10⁴</u>
Nil	2	24	2	97
NPKS	6	100	3	30
NPKS + Amendments*	100	28	29	24

*Amendments = Lime (12 ton/acre) and oiled soil inoculum (4 ton/acre) + micronutrients

**For methods see the bottom of Table 6

Table 6 Numbers of Bacteria and Fungi in soil from oiled and unoled areas at Leedale, September 15, 1973.

<u>Location</u>	<u>Numbers per gram field moist soil</u> **	
	<u>Bacteria X 10⁶</u>	<u>Fungi X 10⁴</u>
Spill area	87	94
Control area	28	18

** Numbers reported are averages of five replicates and are plate count values, not direct microscopic observations. Suspensions containing 0.1 ml of 10⁻⁵ and 10⁻⁶ dilutions of soil in sterile water were spread on separate plates of Bacto Plate Count Agar (Canlab) and replicated five times. Only bacteria grew at these dilutions. Suspensions of 0.1 ml of 10⁻³ and 10⁻⁴ dilutions of soil in sterile water were spread on nutrient agar containing 30 µg streptomycin per ml and 33 mg rose bengal per liter. The rose bengal and streptomycin restricted bacterial growth and allowed developing colonies of fungi to be counted. Incubation was at 20°C for 10 days.

It is recognized that this provides only an estimate of a broad group of bacteria and not necessarily those specific to oil degradation. The presence of oil in the soil restricted the number of genera represented on the plates. Also the numbers reported for fungi must be treated with caution since they may represent spores and structures which were not active under soil conditions. However the dynamics of the fungal population as represented by the data are still valid. Also the use of soil extract agar for comparison of plate count members on oiled and unoled soil is not valid if extract from unoled soil is used for both. A standard agar was selected to standardise the enumeration technique.

Table 7. Soil pH at Swan Hills four months after imposition of various treatments.

<u>pH (October 20, 1973)**</u>	
Box and Frame experiment	
Drained	
Nil	4.5
NPKS	4.8
NPKS + Mixing	4.9
NPKS + Mixing + Amendments*	7.0
Undrained	
Nil	4.8
NPKS	4.9
NPKS + Mixing	5.3
NPKS + Mixing + Amendments	6.6
Piling Up experiment	
Drained	
Nil	4.6
NPKS	4.7
NPKS + Amendments	7.2
Undrained	
Nil	4.5
NPKS	4.8
NPKS + Amendments	6.7

* Amendments = Lime (12 ton/acre) + oiled soil inoculum (4 ton/acre) + micronutrients.

** Soil pH was measured in a 2:1 soil:H₂O suspension using a pH meter, with a glass electrode and a capillary tip calomel reference electrode.

Table 8. Mineral-N content of Swan Hills soils after imposition of various treatments.

Box and Frame Experiment						
$\mu\text{g N per g moist soil}^{**}$						
Treatment	$\text{NH}_4\text{-N}$			$\text{NO}_3\text{-N}$		
	June 22	July 11 [†]	Aug. 9 [‡]	June 22	July 11 [†]	Aug. 9 [‡]
Drained:						
Nil	1	15	2	3	2	1
NPKS	1	88	111	3	67	25
NPKS + Mixing	0	113	169	4	86	56
NPKS + Mixing + Amendments*	0	79	74	3	19	1
Undrained:						
Nil	2	8	4	3	6	0
NPKS	1	5	21	3	3	1
NPKS + Mixing	2	5	37	3	6	0
NPKS + Mixing + Amendments*	1	7	29	3	7	0

Piling Up Experiment				
$\mu\text{g N per g moist soil}^{**}$				
	$\text{NH}_4\text{-N}$		$\text{NO}_3\text{-N}$	
	July 11	Aug. 9	July 11	Aug. 9
Drained:				
Nil	4	2	8	1
NPKS	350	97	289	54
NPKS + Amendments*	320	10	284	1
Undrained:				
Nil	3	0	6	1
NPKS	83	1	57	2
NPKS + Amendments*	126	0	88	0

* Amendments - Lime, micronutrients and a mineral soil inoculum.

** For analytical procedure see bottom of Table 9.

[†] Three weeks after first addition of N (as NH_4NO_3)

[‡] Three weeks after second addition of N (as NH_4NO_3)

Table 9. Mineral N content of soil at Leedale site after imposition of various treatments.

Treatment	µg N per g moist soil **			
	NH ₄ -N		NO ₃ -N	
	Oct. 6†	Oct. 19‡	Oct. 6†	Oct. 19‡
Nil	20	21	0	1
NPKS	113	106	235	19
NPKS + *Amendments	44	65	10	3

*Amendments - Lime + micronutrients + soil inoculum

**Nitrate and exchangeable ammonium N were extracted from 20 g subsamples of field moist soil with 100 ml 2N KCl after grinding the total sample in a Waring Blender. Ammonium N was steam distilled from a 20 ml. aliquot using MgO, collected in boric acid and titrated to pH 4.7 with 0.005 N H₂SO₄. Nitrate N was reduced to the NH₄ form with Devarda's alloy and the NH₄ distilled, collected and titrated as above. Analyses done in duplicate.

† three weeks after first addition of N

‡ two weeks after second addition of N

Table 10. Effect of various treatments on yield of oats in Swan Hills Box and Frame experiment, September 9, 1973 (2½ months after seeding).

	<u>Yield of oats**(g dry wt./32 ft.²) on various treatments</u>			
	<u>Nil</u>	<u>NPKS</u>	<u>NPKS + Mixing</u>	<u>NPKS + Mixing + Amendments *</u>
Drained	0.8	2.0	95	210
Undrained	0	0	0	0

	<u>Above values converted to kg/ha</u>			
	<u>Nil</u>	<u>NPKS</u>	<u>NPKS + Mixing</u>	<u>NPKS + Mixing + Amendments *</u>
Drained	2.7	6.7	318	702
Undrained	0	0	0	0

Note: 1 Kg/ha = 0.893 lb/acre.

*Amendments = Lime (12 ton/acre) + oiled soil inoculum (4 ton/acre) + micronutrients.

** Timothy, reed canary grass and oats were seeded. Timothy and oats grew but only the oats were harvested. In the nil treatment the surface cover ratio timothy/oats was 50:50 and dropped to 20:80 in the NPKS + mixed + amendment treatment.

Table 11. Effect of various treatments on germination of three plant species in Swan Hills dike experiment September 9, 1973 (10 days after seeding)**.

(a) Average % germination (for each treatment or species)								
	Mixing		Nutrients			Species		
	Yes	No	Nil	NPKS	NPKS + Amendment*	Oats	Timothy	Reed canary grass
Undrained	2.8	1.2	2.4	2.5	1.1	4.5	1.1	0.3
Drained	42	29	36	34	39	70	34	3.1

(b) Average % germination of oats and timothy on various treatments (average of 3 replicates)						
	Mixed			Not Mixed		
	Nil	NPKS	NPKS + Amendments*	Nil	NPKS	NPKS + Amendments*
Oats						
Undrained	5	1	7	4	2	0
Drained	83	75	75	52	60	77
Timothy						
Undrained	0	3	0	3	0	0
Drained	47	43	43	25	18	28

* Amendment - Lime (12 ton/acre) + oiled soil inoculum (4 ton/acre) + micronutrients.

** Germination measured by visual estimation of % of each row containing growing plants.



Plate 1. A view of the Swan Hills oil spill.



Plate 2. The "Box and Frame" experiment at Swan Hills, 3 weeks after crops were sown. Soil at the left received simulated drainage; while soil at the right received simulated drainage, mixing, fertilizers, and amendments.



Plate 3. The "Box and Frame" experiment at Swan Hills, 11 weeks after sowing of crops. The box at the left received only simulated drainage; while the box to the right of that received simulated drainage, mixing, fertilizers, and amendments.



Plate 4. The "Box and Frame" experiment at Swan Hills 11 weeks after sowing of crops. The two frames enclose soil which was not drained and produced little growth of sown crops.



Plate 5. Two treatments in the drained area of the "dike" experiment at Swan Hills, 10 days after sowing of crops.



Plate 6. The un-drained or control area of the dike experiment at Swan Hills, showing little or no germination of crops at 10 days after sowing.